

# Manipulating Pig Production XI

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## THE BATTERHAM MEMORIAL AWARD

The Batterham Memorial Award is a prestigious award offered by APSA in memory of the late Dr Ted Batterham.

Ted Batterham's love of pigs began at Wollongbar Research Station in the mid 1960s when he began work with Dr John Holder to solve the problem of variability in the growth of pigs fed meat meals. At that time abattoirs in NSW produced meat meals that were very variable because there was little control on either the raw materials used or the cooking times and temperatures. Ted soon realised that part of the variability was explained by the content of bone but, something much more fundamental that would keep Ted focused and fascinated for the rest of his professional life, was the variability of available lysine in these meals. Ted knew that if proteins were heated in the presence of carbohydrates and fats, lysine would become unavailable to the pig's own enzymes. Ted went to Melbourne University to commence a PhD with Tony Dunkin to develop an *in vivo* assay in rats and pigs to quantify the available lysine not just in meat meals but in a range of other protein sources and cereals. He returned to Wollongbar and became a world leader in the availability of amino acids in feedstuffs for pigs and poultry. Not content just to solve a problem, Ted wanted to find solutions and reasoned that, if the availability of lysine was known, any shortfall could be remedied by supplementation with synthetic lysine. That idea stimulated a long research career that delved into ways that the biological value of proteins could be increased by supplementation with synthetic amino acids.

Ted's research career was always focused on industry problems and driven by a desire to find suitable solutions. He knew that progress was best made by teams of people stimulating and feeding off each other. And he knew that investment in young people was essential.

Therefore, the Batterham award is made to a young scientist, a person within 10 years of graduation. Its aim is to "stimulate and develop innovation and initiative in the pig industry". It is anticipated that the cash award will enable the recipient to broaden his or her exposure to national or international pig science.

### Previous winners of the Batterham Memorial Award:

Rob van Barneveld (1995)

John Pluske (1997)

Kaye Coates (1999)

Darryl D'Souza (2001)

Pat Mitchell (2003)

Eva Ostrowska (2005)

## Acknowledgements

The biennial conference of the Australasian Pig Science Association (APSA) has established itself as an important international conference for pig science. This has only been achieved as a result of the hard work of many people over more than twenty years since the very first conference was held in Albury, New South Wales in 1987. As with previous conferences, a willing band of people have offered considerable time and effort to ensure the 2007 conference reaches the same high standards now expected of APSA. The continued support of members and others associated with pig science and production through submission of papers to these proceedings is acknowledged. Of course no conference is a success without a good number of delegates, and the APSA Committee thanks all those who have attended the 2007 conference and also those who have been instrumental in encouraging others, especially those from overseas, to participate.

APSA invites up to four international speakers to each conference, and the contributions from Keith Behnke, Colm Moran, Matthew Wilson and Ruurd Zijlstra, are greatly appreciated. In addition, I acknowledge the following people from Australia who also contributed to the success of the symposia and reviews: Pat Blackall, Heather Channon, Brett Cowan, Darryl D'Souza, Neil Gannon, John Pluske and Alan Skerman. The A.C. Dunkin Memorial Lecture is an important part of any APSA conference, and the Committee thanks Ian Williams for accepting the honour of presenting the 2007 Lecture. APSA also thanks this year's chairpersons and judges.

There are very few conferences now held where the proceedings are produced prior to the conference and to such a high editorial and scientific standard. The contribution and dedication of the Editors, Janet Paterson and Jenny Barker, in ensuring this happens is acknowledged. The team from Par Excellence who work with Janet, especially Leith Finnie, Julie Taylor and Dickson Poon, are also thanked for their on-going support and contribution to this process. It is becoming increasingly difficult to find people who are prepared to act as referees, and the contributions of these people (named elsewhere in the proceedings) are gratefully acknowledged.

APSA has always had a strong relationship with Australian Pork Limited, and it is pleasing that they again agreed to be the Principal sponsor for the event. The partnership with the newly formed Pork CRC is starting to grow and many of the one-page papers have had direct support from the Pork CRC. It is certainly hoped that the partnership between APSA and both of these bodies can continue to grow.

The organising committee works hard for two years to organise each conference. Accordingly thanks go to Neil Gannon (Vice President), Hugh Payne (Secretary for 2006), Karen Moore (Secretary for 2007), Megan Trezona (Treasurer), Pat Mitchell (Past President), Heather Channon, Frank Dunshea, Darryl D'Souza and Rob Smits. The 2007 conference was organised in conjunction with YRD who acted as the secretariat for this conference, and it has been a pleasure to work with Kate Murphy, Mary Sparkman and Louise Ritchie.

Finally, the XIth Biennial Conference would not have been possible without the generous support of a number of sponsors. These sponsors, listed below, are gratefully acknowledged for supporting the 2007 APSA conference.

Bruce Mullan  
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## Pork CRC

In 2005 the Co-operative Research Centre for an Internationally Competitive Pork Industry (Pork CRC) was established. The Pork CRC is funded by an alliance between the Australian Federal Government, industry and research organizations in Australia and New Zealand.

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An increasing amount of pig research done in Australia and New Zealand is being funded under the auspices of the Pork CRC. Because of the arrangement between the Pork CRC and its contributing partners, the papers in this proceedings that have received funding from the Pork CRC are indicated by the first line of the address: "Co-operative Research Centre for an Internationally Competitive Pork Industry, Willaston SA 5118".

Further details relating to the Pork CRC can be found by visiting the website: [www.porkcrc.com.au](http://www.porkcrc.com.au)

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### Acknowledgement to Referees

The Proceedings 'Manipulating Pig Production XI' contains 95 one-page papers, 3 reviews and 3 symposia. As is the policy of the Association, all one-page papers, reviews and symposia were reviewed by external referees. The APSA committee and Editors gratefully acknowledge the assistance generously given during 2007 by the following referees and those who may have been inadvertently omitted from the list.

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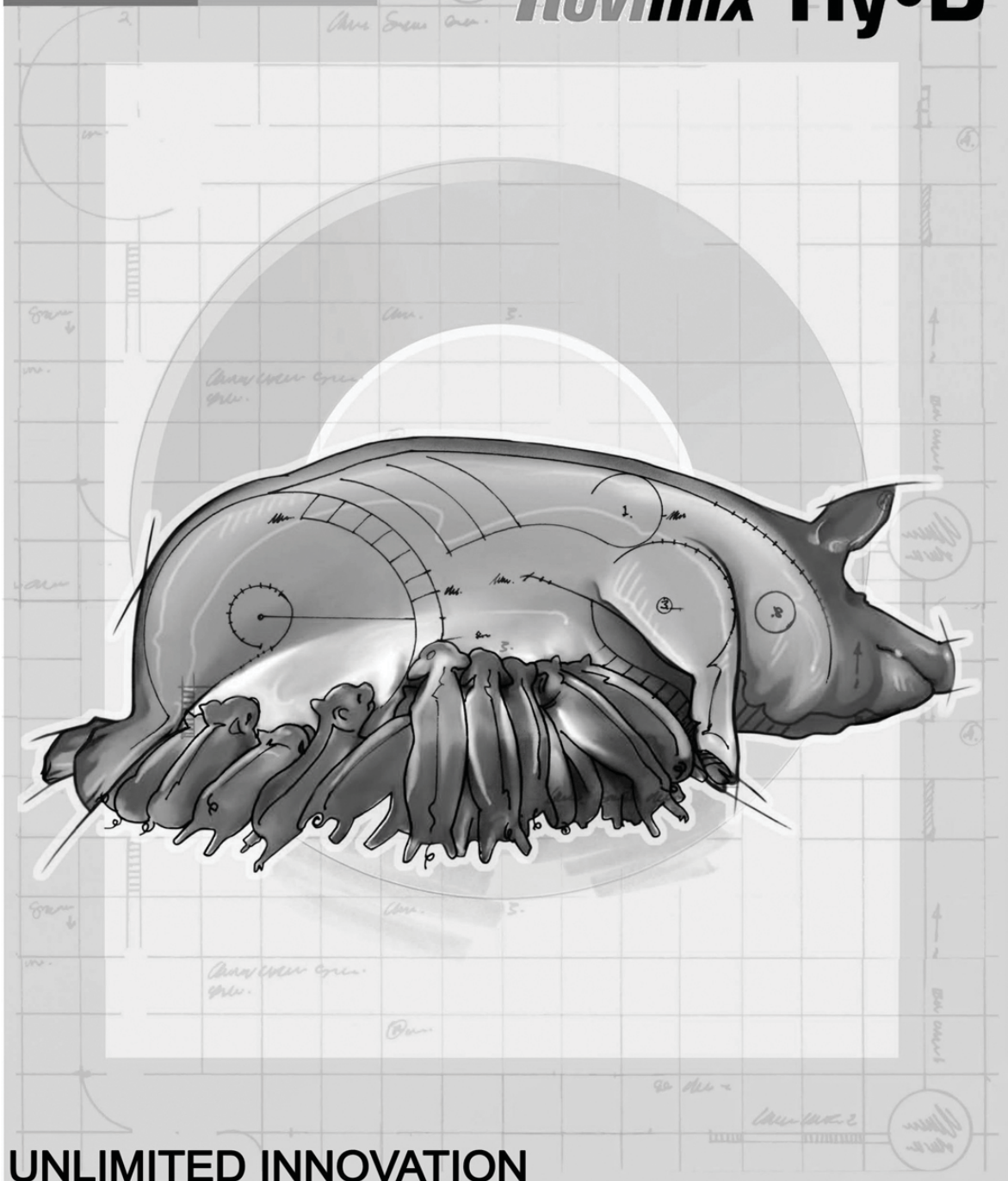


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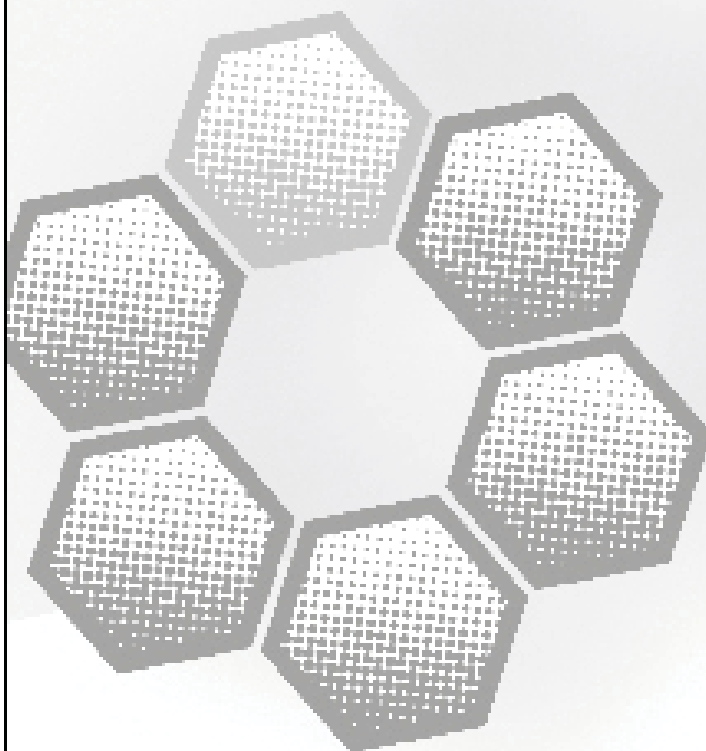
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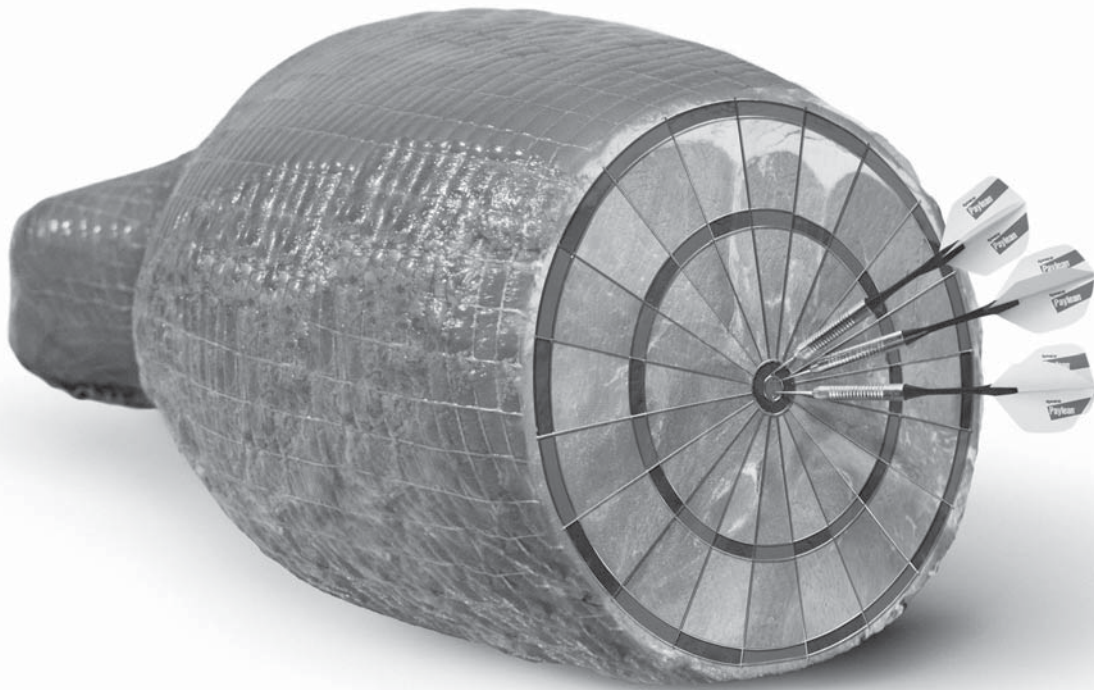
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## Preface

The pork industries in many countries have never seen such tight financial pressures like those of the present moment. Record high grain prices due to a combination of poor seasonal conditions and increased competition for grain have contributed to the financial pressures of pig producers. However, the cost of many other production inputs combined with a difficulty to find and retain a skilled labour force has also increased the cost of pork production. Ideally such cost increases would be passed directly onto consumers, with the profit margins of all those involved along the supply chain being maintained. But in most cases this does not happen.

The industry in many countries has started to make massive changes in the way it operates, including a decision by some to close down production or by others to restructure. For most, the only chance for survival is to make improvements in efficiency by the adoption of new technology or, in some cases, re-introducing old production methods that have regained relevance, and it is here that APSA can and should play an important role.

By providing a forum for people from a wide range of backgrounds and interests to meet and share ideas, APSA provides the opportunity for important changes in pork production to occur. The conference is also an important training ground for new graduates who are integral to the future of the industry and for many it will be the first opportunity that they have to present their findings in public.

Dr Ian Williams has sent all those associated with APSA a very strong message in his presentation of the Dunkin Memorial Lecture. He makes it very clear that APSA has developed high standards for scientific communication and that these standards must be maintained at all costs. With increasing pressures on our time and funding, this goal is often difficult to achieve. However, if we truly want the pork industry to realize its potential then we must carefully consider Ian's thoughts and recommendations.

I am pleased with the good support for the 2007 biennial conference of APSA especially given the difficult circumstances and I think this highlights the positive attitude of all associated with the conference. Whatever the circumstance it is essential to meet with other people from a mix of backgrounds, expertise and nationalities to share their thoughts and problems. It has been a pleasure to act as President of APSA for the past two years. I firmly believe that APSA has a more important role than ever in getting the results from research to the end-user in a clear and concise format. I certainly wish the Association all the very best for on-going success and hope that it continues to spread its reputation throughout the world.

Dr Bruce Mullan  
President  
APSA





## A.C. Dunkin Memorial Lecture

# The history of APSA, its achievements and contribution to science and its possible future directions

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### Introduction

The Australasian Pig Science Association (APSA) has achieved what many other professional bodies only dream about for three good reasons. First, it provides a forum for the discussion of science as it relates directly or indirectly to pig production. Second, it produces a publication that is both refereed and edited and which, most importantly, is accepted internationally as a publication of substance. Third, it has welded together people from all facets of the industry; producers, scientists, extension personnel, feed formulators, equipment manufacturers, providers of specialised products and it has encouraged them to work together on problems faced by the pig industry.

I think that these achievements were only possible because APSA has stuck rigidly to the principles of science since its inception 20 years ago, that is, it has practised well-accepted principles of seeking out the truth. I believe very strongly that if APSA is to continue to be a successful body it must make an even greater effort to adhere to the principles of science. Why? Because I believe that the recent arguments of John Roche and Doug Edmeades (Roche and Edmeades, 2006) are very compelling and that the world is changing from one where reasoning predominated and good science was accepted by all to one in which the non-reasoners are now being heard and sound debate is pushed to the background and stifled.

I wish to argue that we as scientists must adhere rigidly to and demonstrate to others the accepted principles of science to make sure that science survives the challenge from non-reasoning cultures and individuals. We owe this to our teachers, to our society, APSA, to the wider community in which we live, and particularly to ourselves as practising scientists. I suggest that we need to go one stage further and seek better ways to communicate our science because others outside science need to know about the achievements of science.

In presenting the history, I wish to stress that APSA has been successful because of its insistence on good science and scientific presentation. But particularly, I wish to stress that its continued success depends on adhering to these principles.

This paper is a tribute to my supervisor and friend, Tony Dunkin. When I finished this paper, I realised that all the ideas expressed here, particularly the commitment to science and truth, are what Tony stood for and held very dear. I am just passing these ideas on to you, albeit less eloquently than he.

## The history of APSA and pig science

APSA was formed by a group of scientists interested in pigs that broke away from a much larger group, the Australian Society of Animal Production (ASAP). The breakaway began when the pig scientists who attended an ASAP conference in Brisbane in 1982 felt that they were not receiving the recognition they deserved. When the same problem emerged at an ASAP conference in 1984 the breakaway gained momentum and, in August 1986, the first committee meeting of APSA was held with attendees that came mainly from Werribee. To understand and appreciate the breakaway movement it is necessary to trace the history of pig research in Australia because, when viewed in this light, the formation of APSA was not simply a bunch of belligerent scientists acting defiantly but, in fact, was a natural evolution that arose from the rapid expansion of pig research in the previous 15 years.

In the early 1980s pig research in Australia was perceived by many mainstream scientists in agriculture to be small, relatively poorly funded and certainly not a significant force compared with research into red meat or wool. This was not surprising since pig research hardly existed in Australia in the early 1970s but then it experienced a meteoric rise that few, except those within the industry itself, appreciated. For example, in 1969 there were five scientists in Australia working directly on pigs and 12 recognised extension personnel (Table 1). But, by 1985, the number of research scientists had grown to 190 and the extension personnel to 110. How did this happen in such a short time?

Derek Tribe, professor of animal production at Melbourne University, was the mastermind of this research expansion and, in 1969, he orchestrated three masterstrokes. First, he coaxed three eminent pig producers (Melville Charles, Barney Bell and Dudley Smith) to convince pig producers in Victoria that, if sufficient funds could be raised to build a research piggery at Melbourne University, problem-orientated research of great value to the industry would follow. Second, he convinced the federal minister for agriculture, Doug Anthony, to establish a federal funding body financed and administered in the same way as the much larger Wool Research Council. That is, that the Government would match dollar for dollar those funds raised from producer levies. The Pig Slaughter Levy Collection Act was passed by the Federal Parliament in 1971 and the Australian Pig Industry Research Committee was established to administer the funds generated from the levy. This Committee began with about \$500,000 to distribute in its first year. Third and perhaps most important, he sought out Mr Anthony Dunkin, a British scientist working in New Zealand, and convinced Melbourne University to appoint Tony to head the Mt Derrimut Pig Research and Training Centre. Tony was appointed not only to design and run a “state of the art” piggery but also to make Melbourne the centre for post-graduate training in pig science in Australia. Ted Batterham, a young scientist working on pigs with John Holder at Wollongbar Research Station in northern New South Wales, came to Melbourne to be Tony Dunkin’s first PhD student. Many more students from all around Australia followed.

**Table 1. Professional people in pig research and extension**

	Research	Extension
1969	5	12
1985	190	110

In the fifteen years that followed its establishment, the Mt Derrimut Pig Research and Training Centre played a major role in training people to Master and PhD level. But the Mt Derrimut Centre and funds from APIRC stimulated training in pig science at other centres around Australia (Sydney, Wollongbar, Brisbane, Adelaide and Perth) so that, by the mid 1980s, Australia had many scientists trained and, most important, dedicated to the pig industry. Why were so many students stimulated towards training opportunities in the pig science and why did so many dedicate their professional life to the pig industry? I believe that the major influence was the skilful engineering by two individuals working together, Derek Tribe and Tony Dunkin.

Not only did Derek Tribe convince the Federal Government to establish a funding body, the Australian Pig Industry Research Committee, but he also had a major input into the composition of the Committee and suggested as he always did, kindly but forcefully, the way that would make it operate effectively. First, he convinced government to appoint a committee with a majority of scientists who could assess the scientific merit of submissions because he believed that, unless the research was high-quality, it would be of little use to the industry and producers would balk at forking out levies. Second, he suggested that the Committee fund pig science in as many centres as possible. Tribe believed that funding so called ‘Centres of Excellence’ often led to the development of the ideas of a few rather than many and he knew that no one individual no matter how bright had a mortgage on ideas. Third, he suggested that the committee should encourage post-graduate training wherever possible even if it meant producing more scientists than the pig industry could employ at that time. Tribe knew and reasoned to others that good scientists would always find a useful job and, if called on at a later date to come back to serve the pig industry, would always willingly do so to express their gratitude to an industry that had supported them in their initial studies. Last, Tribe insisted that the

Committee organise sufficient time and invest sufficient funds to make sure that scientists could be visited regularly in their own laboratories. He knew from experience that a funding body could make very sound judgements about how research was progressing if they visited scientists on home soil so that they knew scientist's thinking and understood the problems faced.

When Tony Dunkin arrived in Australia in late 1968 he began immediately to design the Pig Centre at Mt Derrimut. He also talked extensively to industry about their problems because he believed that, if the Centre was to be established by producer money, the science conducted should be orientated towards major industry problems rather than esoteric research. He set about organising the first national conference in Australia dedicated to pigs to bring together producers and scientists (Watson, 1969). This was held at Mt Derrimut in 1969 and everyone with even a remote interest in pigs was invited including a notable reproductive physiologist from the United Kingdom, Dr Chris Polge. The conference was a great success but it was noteworthy that there were no contributions from any Australian post-graduate students. A second national conference followed at Gatton College in Queensland in 1975 (Milne, 1975) with a very large contribution from post-graduate students that were training in Australia.

Given the effort of raising the profile of pig research in Australia in the 1970s and early 1980s and the numbers of young scientists now trained in pig science, it was no wonder that these scientists were ready and had the resolve to form their own body. The seed for APSA was sown during the ASAP conference in 1982. A small minority, of whom I was one, argued against a breakaway from ASAP because we felt that concentrating on a species of animal rather than a scientific discipline might weaken us as scientists. How wrong we were. Had pig scientists and extension people stayed with ASAP they too might be in that organisation's present state; a dying society struggling for members and struggling to organise conferences that attract anyone but a few faithful stalwarts.

The founding fathers of APSA led by Ray King agonised over the problem of how to create a society that would be successful in the long term and meet their major goals of providing a forum that the pig industry could use to identify and discuss its problems and work out possible solutions to tackle them. Their definition of industry was wide and it included pig producers and all the people in service industries (equipment, research and extension). They made several key decisions at the outset and, as time has shown, they acted wisely. They decided that the best forum would be a conference but realised that running such an event each year was not feasible because good programs take time to put together and that good conferences require a critical mass of new research findings. The compromise was to hold a conference every second year. They were careful to choose the best site – they chose Albury because it was the demographic centre for most of the pig industry at that time. They realised that, despite the rapid growth in pig research in Australia, research workers in many other countries had much to offer. Their answer was to invite two or three high-profile research workers from outside Australia to address the conference – an expensive proposition. They were also smart enough to realise that paying delegates could not afford to fund overseas speakers and so they sought sponsorship from the federal funding body, the Pig Research Council (formerly the Australian Pig Industry Research Committee). In summary, they took a gamble and decided to run a conference with a top-quality scientific program for three days (Sunday evening to Wednesday lunchtime) that included high-profile, overseas research workers and produced an edited, refereed proceedings that also included the one-page papers from local people and invited reviews. The inaugural conference of APSA took place in Albury, New South Wales in 1987 and about 150 people interested in pig research, production and extension attended. That conference was a great success from both scientific and a social point of view.

Time has shown that the principles adopted by the founding fathers have been extremely successful because the most recent conference in Christchurch, New Zealand, in 2005 was attended by 330 delegates, a massive growth in attendance from the 150 that attended the first conference.

### **Achievements of APSA and its contribution to science**

If we analyse the achievements, APSA has achieved all it set out to do 20 years ago. It was formed with the major aim of providing a forum for the pig industry to identify and discuss its problems and work out possible solutions. I believe that it has been highly successful in achieving this aim because the Society truly encompasses and represents the whole pig industry. For example, the current membership is approximately 38% research and extension, 19% producers and 42% industry (14% consultants including veterinarians and 28% private companies) (M. Trezona, Secretary, APSA, personal communication) - an extremely good mix of personnel to represent the pig industry. In addition, APSA has managed to retain the considerable sponsorship from the principal funding body, Australian Pork Limited (formerly the Australian Pig Industry Research Committee and the Pig Research Council). Australian Pork Limited has always responded to APSA's request and welcomed the offer to send one of its employees to serve on the APSA Committee. It also encourages several of its personnel to attend the biennial conference where they can monitor industry problems and offer encouragement and support for new projects. Australian Pork Limited has long recognised that APSA represents the pulse of pig science in Australia.

But APSA has achieved far more than it set out to do some 20 years ago. I doubt that the founding fathers ever thought that APSA might extend well beyond Australian shores. In 2001 research workers in New Zealand were encouraged to nominate for membership of the Organising Committee and they welcomed the opportunity to join. In 2005 the 10th Biennial Conference went offshore and was held in Christchurch, New Zealand with a record attendance of delegates. In recent years several overseas scientists have joined the Society. The current membership is 77% Australian including 6% students, 8% New Zealand and 15% in other countries.

Not only has APSA served the pig industry well over the last 20 years but it has also made a substantial contribution to science because it publishes the proceedings of its conference that now have international recognition. I believe that this has only been achieved because APSA has maintained its scientific integrity and refereed and edited the contributions it receives. My experience is that most pig scientists want to publish their work in the APSA proceedings. Recently, a very senior pig scientist wrote to APSA asking if he could publish some aspects of his work in the Proceedings that was already published in another journal because he felt members of APSA would be interested.

From time to time the possible downside of a rigid system of refereeing and editing has been discussed. Some people have suggested that it may prevent the latest but still unsubstantiated information from being submitted and aired at the conference or that it may sometimes disadvantage students doing higher degrees. This debate is far from over but my personal view is that the downsides are a small price to pay for “honesty in science” and I would urge the Society to maintain its current system of refereeing and editing no matter what the perceived flaws.

Perhaps the ultimate yardstick of APSA's success is the vigorous elections that take place to form the Organising Committee for each biennial conference. In most scientific societies, members are coerced to join such committees. My experience is that when members volunteer their services the best job always gets done with the minimum fuss.

### **The future of APSA**

I believe that if APSA is to continue to be a successful society it must encourage its members to continue practising good science and make sure that its forums always allow and encourage open debate. This is more important now than ever before because I am persuaded by recent suggestions from John Roche and Doug Edmeades (Roche and Edmeades, 2006) that the world is changing from one where sound reasoning predominated and good science was accepted to one in which the non-reasoners are now being heard and sound debate is being pushed to the background and stifled. The best counter to this change is to keep practicing good science and make sure that the rest of the world knows about it.

I want to summarise what we all know and agree about good science and then I wish to present the arguments for why our world is changing and what we can do about it.

### *The scientific method*

What is it and why has it served us so well?

The scientific method accepted by all of us consists of the testing of hypotheses followed by peer review and then publication in accepted journals. There are five stages. The first involves extensive reading of the literature associated with the area of study. It may also involve talking to colleagues, associates or other scientists who may help clarify issues and even offer ideas. From reading and assessing the evidence, accepted facts are identified and assembled to create an hypothesis or, as some people may prefer, an expectation based on scientific rationale. Stage two involves designing an experiment (laboratory, field, or survey) to test the hypothesis. Both these stages are difficult and often involve many forays up blind alleys and the retracing of steps. Stage three is conducting the experiment and assembling the results in a presentable form and, for trained scientists, this task is often largely mechanical and requires much less brainpower than the first two stages. The fourth stage involves interpreting the results and communicating them, generally in written form. Here the writer tries to convince readers that the hypothesis was soundly based and that the results either support or reject the hypothesis or, in rare cases, are equivocal in their support or rejection. The last stage involves peer review where a manuscript is submitted to a learned journal for publication. After submission the editor approaches referees, generally at least two, who are invited to comment on the paper generally anonymously.

Most of us have experienced the stress of Stage 5 at some time or another, a stage that can drag on for months depending on how much compromise is involved. Referees can reject or seriously question the results and/or their interpretation. The preparation and submission of the paper often represents months or even years of painstaking work summarised into a few pages by writers who have been compelled to weigh up every word that they write. As a consequence, criticism is often hard to accept particularly at that time. A referee may read the paper in an evening, perhaps after a reminder or two from the editor, and is then allowed a ‘free-for-all’ attack on the writer's precious piece

of work. Under cover of anonymity, the editor, who has chosen the referees and appears to agree with their comments, then asks the writer to respond appropriately if the manuscript is to be considered for publication.

The scientific method I have just described has remained pretty much the same for the last 100 years but there are now some significant changes occurring. Keeping up-to-date is more difficult now than it was 20 years ago. The sheer amount of information now available is baffling because of the incredible increase in the rate of publication. Not only are we bombarded in everyday life by information overload but the same applies in our scientific life. Publication rate of journals, the main source of scientific information, has exploded in recent years and is increasing exponentially. Scientists can no longer stroll into the library and find 90% of the information in hard copy in eight or 10 journals. They are forced to rely on computer searches which, although reasonably efficient, are far from perfect. The ‘publish or perish’ syndrome has meant that if scientists cannot get a publication accepted in the most appropriate journal for the topic they are tempted to submit to less-appropriate journals which may be more obscure to many potential readers.

Finding suitable referees is becoming very difficult and many editors or post-graduate supervisors will attest to this. Recently I agreed to referee a paper and the editor replied ‘many thanks indeed, a first acceptance is a rarity these days’. The solutions may be more worrying than these problems. For example, Medwell, who control 19 publications, now offer to turnaround a paper in 2 to 4 days (Medwell, 2007). Can refereeing standards possibly be maintained with a guarantee of such a short turnaround?

Despite all the problems outlined above, the scientific method has served us well because publication makes us explain clearly to others what we have found and peer review makes us constantly recheck that our results explain the real world and fit its great jigsaw puzzle. It’s a slow process and, unfortunately, there are few, if any, shortcuts to speed it up.

### *The changing world*

I have been convinced by the arguments presented in a recent paper, ‘Fact or fiction? How do I know who’s telling the truth’, written by two eminent scientists working in private industry (Roche and Edmeades, 2006). I wish to repeat to you some of their ideas about the changing world of science and possible warnings for the future.

The scientific method evolved about 600 years ago when people began to challenge the authority of the Church and questioned the idea that there was truth other than divine truth that came from worship. People, deemed heretics, picked up concepts and ideas from pre-Christian Greek thinkers and believed that truth could also be revealed by logic, reasoning and experimentation. They started the “age of reason” or Modernism (Table 2) and we, as scientists and extension people in the industry, believe implicitly in this method of obtaining knowledge and establishing truth.

Roche and Edmeades (2006) have suggested that we are rapidly entering an age of political correctness that imposes limits on viewpoints and reduces public discussion to avoid informed debate that might be controversial. All ‘truths’ or opinions must be given equal weighting. We all agree on the absurdity of this last statement but I suspect that many of us feel helpless to do much about it. The louder we yell, the less we are heard. Perhaps the best way to fight the issue of political correctness is to lead by example. I believe that APSA has a major role to play in two ways. First, we must stick rigidly to the principles of science (hypothesis testing, peer review and publication) to increase our knowledge base and discover ‘the scientific truth’. Second we must make a much greater effort to communicate not only our scientific findings but also to give people a better understanding of the scientific method. I doubt that any member of this Society would argue against the first suggestion. So I wish to enlarge on the second.

**Table 2. “Truth” as it was and is changing though time (after Roche and Edmeades, 2006)**

<b>Times</b>	<b>“truth”</b>	<b>Authority</b>	<b>Attitude</b>	<b>Example</b>
Dark Ages	“divine truth” only from worship	Church	Obey laws of God	Pray for a good harvest
Age of Reason	“truth” from logic, reason & experiment	Science	Opinion counts for little, facts are essential	Vaccination can prevent diseases Amino acids are essential for the growth of pigs
Age of non-reason (Post-modernism)	If you believe it is true, it is	Individual or group of believers	Science is behind global misery. A “new truth” must be found	GM crops are bad for human health

One of the problems that many scientists face is that, while they intuitively know that writing is an essential part of science, most loathe actually doing it. Most scientists invest at least 10 years in their training, four years as an undergraduate, three or four years as a post-graduate (one or two years as a post-doctoral fellow, and three to four years as a full professional in a scientific institution or university), but at no stage during that time do they receive tuition or guidance on how to write scientifically. They overcome this deficiency in training by going to the scientific literature and trying to copy its style. They make sure that their method of expression is as dispassionate as possible and they fill the text with jargon to make it appear as scientific as possible. Many years ago the research institutions like the State Departments of Agriculture and CSIRO employed writers to publicise their scientist's findings. These were people who could interpret the scientific language and put it in lay terms so all could understand. As money for research became scarce and more difficult to obtain these people were dispensed with first. It is now left up to the scientists themselves to sell their findings to the wider population. Without the necessary training scientists have little chance of achieving this successfully.

There are, however, some small lights on the horizon signalling that the scientific literature is changing but the change is very slow. Editors know that the reading audience is crucial to their journal's survival because the costs of publication are such that libraries are now forced to choose between those journals they will subscribe to and those that they cannot afford. The times when all libraries could afford all journals are gone. The response by editors has been to start making their journals more readable. Titles of papers are beginning to change from 'The effect of A on B' to something that catches the reader's attention by emphasising a major result or conclusion. Introductions are being shortened to include just the hypothesis and its scientific rationale rather than the "feel good" introduction that lists all the seminal papers in the field but never really suggests what's in the writer's mind. For example, the *Journal of Animal Science* has for some years suggested in its instructions to authors that introductions 'must not exceed 2000 keystrokes (characters plus spaces). It briefly justifies the research and specifies the hypotheses to be tested'. Changes such as these improve reading because they put the message of the paper up front. I am suggesting that changes should go further and the best suggestions that I have come across recently are contained in a delightfully-written paper by Kaj Sand-Jensen from Denmark titled 'How to write consistently boring scientific literature' (Sand-Jensen, 2007). In this paper, Sand-Jensen lists 10 rules that he believes will ensure that scientific writing is unbearably boring (Table 3). Many people are guilty of following his rules. I suggest that if we apply Sand-Jensen's suggestions our contributions will be much easier to follow and may attract a wider audience.

**Table 3. Top-10 list of recommendations for writing consistently boring publications (after Sand-Jensen, 2007)**

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Avoid focus
Avoid originality and personality
Write long contributions
Remove implications and speculations
Leave out illustrations
Omit necessary steps of reasoning
Use many abbreviations and terms
Suppress humour and flowery language
Degrade biology to statistics
Quote numerous papers for trivial statements

---

Most of the suggestions in the table above are self explanatory but I wish to highlight just three of the rules. The first is, in my opinion, the most important and I love how Sand-Jensen puts it. He says 'Introducing a multitude of questions, ideas and possible relationships and avoiding the formulation of a clear hypothesis is a really clever trick. This tactic insures that the reader will have no clue about the ..... direction of the author's thoughts and it can successfully hide his (the author's) lack of original ideas.'. This suggestion to focus a paper by presenting a hypothesis based on sound scientific reasoning is not new. David Lindsay from the University of Western Australia suggested this in his text, "A guide to scientific writing" more than 20 years ago (Lindsay, 1984). Yet it is surprising how few papers follow this format. For example, when I scanned the first 116 paper published in the *American Journal of Animal Science* in 2006, I found only 15% of papers contained hypotheses despite the instruction to authors asking for one. I was more encouraged when I did a similar search of the last APSA proceedings (Paterson, 2005) to find that 40% of the one-page papers contained well-reasoned hypotheses. But, we can do better. Personally, I find reading a paper that has a well-justified hypothesis much easier to follow and understand and so much quicker to read than one without.

Sand-Jensen's second rule, omitting the necessary steps of reasoning is an excellent way to educate a small elite group while the majority of readers are lost. 'This style will also effectively prevent communication with ordinary people – a process which is far too time-consuming'. Many of us are, at times, guilty of this.

Third, he informs us, using as many technical words, abbreviations and acronyms as possible signifies to the reader that, unless they, too, know the 'secret language' of the discipline, they will have little chance of understanding anything about the paper. Most of us are guilty of this from time to time perhaps because, as Sand-Jensen points out, 'it enhances our apparent wisdom as writers and often hides our lack of understanding'.

Sand-Jensen's paper is a delightful read and I strongly commend it to you because, certainly for me, the after effects were most salutary. I suggest that members of APSA should assess Sand-Jensen's simple rules when they next read science and determine for themselves whether they make a difference to understanding and ease of reading. If so, I urge members to think about these rules and apply the reasoning embodied in these rules to their own writing because it just might allow us as a Society to attract a greater audience and allow us to deliver our message better.

In conclusion, APSA is a highly successful organization as evidenced by the number of people that attend its biennial conference and the enthusiasm of the members who design the conference program and make it happen. I believe that this success is underpinned by high-quality science and that APSA should resist any temptation to publish contributions that are not peer reviewed and haven't been subjected to the highest scrutiny. Therefore, I urge the Society to continue to publish refereed proceedings that are properly edited. I also urge members to take a further step forward and concentrate on telling a wider audience about their exciting journey in science in a form that is easily digestible.

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# 1

## Product development and sow productivity

# Pork product development in Australia: global trends, local opportunities

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## Abstract

To a large extent fresh meat remains a commodity product and has a considerably lower degree of differentiation compared to many other food products. Many consider the meat industry in western countries as a mature market and product development and innovation have been crucial to the significant growth of the meat industry compared to other foodstuffs. Interest in new products, particularly convenience-orientated products, has increased dramatically in recent years. However, to develop new products effectively, it is paramount that consumer perceptions and preferences are understood and targeted.

The ability of the Australian pork industry to differentiate its product and provide consumers with healthy, innovative products will enable it to compete more effectively against beef, chicken, lamb and seafood. Additionally the pork industry also needs to differentiate itself to convert some of the 'very infrequent meat eaters' to consumers for pork. If pork is to increase its presence on the menu of contemporary consumers, then it needs to adapt and present itself as a relevant and desirable component of today's modern diets. New product development is therefore crucial to the long-term growth and profitability of the Australian pork industry.

Consumers occupy a crucial position in the meat chain, situated at both the end of the chain as the end user as well as at the start of the chain as inspiration for consumer-driven, market-orientated initiatives. Consumer research is essential for segmenting the market into groups with distinct characteristics and needs. Market segmentation provides a better understanding of the market and its needs as it enables behaviour to be predicted with greater accuracy and potential market opportunities to be identified and captured by industry. It also provides the necessary information that the industry as an entity or individual companies can use as the basis for marketing strategies including product development, pricing decisions and communication of benefits. The ultimate success of a food product depends on its acceptance to the consumer, who is the user, potential user and/or the product purchaser.

## Introduction

The 'New Fashioned Pork' promotional campaigns, including "Get Some Pork on Your Fork" and "Just Think Pork" launched in 1982, successfully presented an updated image for pork - new lean cuts, new meal ideas and awareness of the low fat content of pork to consumers. At the time, pork accounted for only 6% of all domestic retail sales of meat, compared to beef at 53% and was considered to be too fatty, too expensive and suitable only for special occasion meals. The new retail pork cuts developed and promoted to consumers through the campaign were leaner, more versatile and of better value than the bone-in traditional pork cuts. However, a study comparing beef, lamb, chicken and pork conducted by the Australian Meat and Livestock Corporation in 1994 found that only 34% of consumers considered pork to be delicious to eat (AMLC, 1994). Only 15% of consumers considered pork to be tender and juicy, with 32% of consumers considering pork to be dry. Over-fatness was also identified to be an important issue, with 65% of consumers stating pork was too rich and fatty. Pork was the toughest, driest and fattiest of all meats assessed. In contrast, 71% of consumers stated that chicken was delicious to eat, 51% found it normally tender and juicy and only 25% of respondents considered that chicken was inclined to be dry. In summary, although the campaign for lean pork met the 'health' needs of consumers, this was not the case for the eating quality expectations. This highlights the importance of understanding the totality of consumer perceptions, needs and expectations of fresh pork.

In the US, leaner pigs and trimming excess fat from pork cuts have lowered the fat content of US pork by more than 30% over the last 30 years (Resurreccion, 2003). The National Pork Board advertising campaign launched in 1997 has promoted pork as 'The Other White Meat' focussing on leaner and lower fat cuts. As a result of this promotional campaign, US consumers are now less likely to perceive pork negatively in terms of fat and cholesterol (NPB 2004).

The most popular cuts of Australian pork sold in major supermarkets in 2006 were pork roasts (48%), chops (19%), ribs (12%), mince (7%), steaks (7%), stirfry and diced (4%) and pork fillet (2%). Consumer research identified intermuscular fat content as the reason for relatively poor consumer demand at the retail level for scotch fillet, when presented as a steak to consumers unfamiliar with this cut reflected (DSM Consultants, 2004). Pork rump steaks were also identified as having little consumer appeal because of their 'two toned' colour and kidney-like shape.

In Australia, the volume share of fresh beef sold at the retail level declined to 30.6% in the quarter October - December 2006 in comparison to 32.2% in the October -December 2005 quarter (Roy Morgan 2006). In contrast, the volume share of fresh pork increased by 1.8%, from 22.5% to 24.3%, in the same period. This suggests that the growth in fresh pork volume has occurred at the expense of fresh beef. In addition, the total number of pork serves purchased at the retail level increased by 12.5%, from 28.9 million serves purchased during the October - December 2005 quarter to 32.5 million serves purchased during the October - December 2006 quarter. In comparison, the total number of fresh beef serves declined by 1.0%, while fresh chicken and lamb increased by 7.1% and 5.6%, respectively. In addition, in December 2006 fresh pork represented 46.7% of total pork volume compared with 41.9% in December 2005 (Roy Morgan, 2006). In 2006, the market share for Australian fresh pork was estimated to be: Food Service 34%, Woolworths 21%, Coles 17%, Retail butchers 17%, IGA/Metcash 2% and Other 10%.

A change in demographic characteristics of consumers has led to changes in the demand for red meat. Demographic data of Australian pork purchasers highlighted that the number of women who bought fresh pork increased by 7.0% in October - December 2006 compared to the October - December 2005 quarter, with increases amongst all ages except women aged 50-64 years old (Roy Morgan, 2006). Interestingly, an increase of 35.6% was identified in the number of women of 14-24 years of age purchasing pork during October - December 2006 compared to the October - December 2005 quarter. In the December 2006 quarter, the number of men buying fresh pork increased by 17.2% compared to the same period in the previous year. Furthermore, increases in male pork buyers were identified across all age groups during the December 2006 quarter.

These data confirm those reported by Resurreccion (2003), who stated that although the overall consumption of red meat and poultry has not significantly changed, beef and veal consumption in the UK and USA has declined. Resurreccion (2003) considered that the decline in UK beef and veal consumption reflected consumer concerns about the safety of beef as a food, animal welfare and environmental perceptions of beef production, consumer concerns about diet and health, changing consumer lifestyles and the availability of more conveniently prepared foods.

Australian Pork Limited estimates the domestic consumption of fresh pork at December 2006 to be 10.63 kg/capita, a growth of 34.6% from 7.9kg/capita in June 2002. The industry strategic goal is to achieve an additional 30% growth in domestic consumption of fresh pork by 2010. Product development has a significant role to play to enable the industry to achieve this objective. In order to continue to meet consumer demand for lean, healthy pork products, the Australian pork industry is also continuing to implement changes to its production systems including reducing feed costs, improving feed efficiency, effective selection of genetic lines, improved feed formulations and feeding systems.

However, despite these recent improvements in domestic consumption of fresh pork, consumer research (Stollznaw, 2005) identified that pork still compares poorly to its competitors: chicken, beef, lamb and fish. Stollznaw (2005) identified that only 30% of consumers rated pork to have a great flavour compared with its competitors: beef (48%), lamb (43%), chicken (38%) and fish (31%). Ease of cooking pork remains a significant barrier for consumers, as only 25% of consumers considered that this was the case. In contrast, chicken was rated by 60% of consumers to be easy to cook followed by beef (49%), lamb (36%) and fish (36%). These findings suggest consumers have not altered their cooking practices as communicated in the 'Cook it Right' campaign, while this may also reflect issues with inconsistent pork quality. Healthiness of pork was rated relatively poorly. Overall, 94% of consumers rated fish to be healthy followed by chicken (54%), beef (43%), lamb (34%) and pork (33%). The versatility of pork cuts was identified to be very under-appreciated for its range of cuts, uses and adaptability, with only 41% of consumers positively responding to this attribute. Only fish considered to be less versatile (32% of consumers) and 72%, 66% and 42% of consumers considered chicken, beef and lamb, respectively, to be versatile.

### **Pork product development in Australia**

The challenge for the pork industry is to develop innovative pork products that are cost efficient, convenient, nutritious, visually attractive, wholesome to eat and safe (Desmond *et al.*, 2000). This will assist with providing consumers alternatives to traditional products. Pork from low value cuts can be high in connective tissue as well as subcutaneous and intermuscular fat.

Product development can encapsulate: new cuts, ingredients, packaging, pre- and par-cooked products, presentation and different formats – in fact, anything that enables pork to be differentiated and competitive against other meat proteins in the retail and/or food service markets. As stated by Linnemann *et al.* (2006), successful consumer driven food product development can resemble the task of hitting a moving target!

Linnemann *et al.* (2006) categorised new food products as:

- 'Me-too' products – this category represents the largest groups of new food products;

- Line extensions – these include new flavours for existing products;
- Repositioned existing products – this category includes those that are promoted to reposition the product, including functional meat products;
- New form of existing products – where the physical properties of the product have dramatically changed;
- Reformulation of existing products – this category could include meat products with lower sodium content or lower fat content;
- New packaging of existing products – this involves accepted products with new packaging concepts e.g. modified atmosphere packaging;
- Innovative products – defined as products resulting from changes in existing products that can not be included in the categories above. Marketing can be costly for products in this category as consumers may have to be educated to the novelty; and
- Creative products – new products that have never been seen before! Creative products are considered to commonly require extensive product development, may be costly and have a high degree of risk.

In 2003, Australian Pork Limited (APL) established its Product Development Program to work with pork processors, retailers, food service suppliers interested in developing and/or marketing innovative cuts or value-added pork products for the domestic and export markets. The focus of the program targets product development of primals and cuts that are not currently in strong supply in the Australian market (e.g. legs, shoulders and loin-off bellies) to lift whole carcass profitability for Australian producers.

### Moisture infusion

Moisture infusion, by the application of functional ingredient in the brine solution to fresh pork, provides pork companies with a way to improve the overall eating quality of the final cooked product, particularly juiciness and reduce the variation in tenderness. In the USA, the adoption and acceptance of moisture enhanced at the retail level pork is widespread. The major Australian pork processors are now regularly producing moisture infused (MI) pork as part of their operations. From a marketing perspective, it is recognised that continuing efforts are required with the major retailers to better support and stock MI pork lines in store. Many consumers still remain unaware of the eating quality benefits of MI pork and how it differs from 'normal' pork, despite extensive promotional campaigns.

Although the processing and packaging technology required to produce moisture infused pork is not new to the meat industry, the introduction and implementation of moisture infused pork by Australian pork processors required a significant increase in industry capability. The majority of the processors interested and committed to MI pork came from a very low processing base, as their businesses were primarily concentrated on boning and slicing fresh pork. This improvement in process capability by pork processors means that new opportunities for valuing adding to pork that require the utilisation of new equipment, food ingredients, cooking and packaging systems/requirements may now be more easily adopted and implemented.

Moisture infusion can now be viewed as a platform technology whereupon the incorporation of flavours and functional ingredients into brine and development of pre-cooked MI pork products are considered to be potential growth areas for both the domestic retail and food service sectors. It is anticipated that providing marinated pork products to Australian consumers will generate incremental sales and value along the value chain and will also assist in improved carcass utilization, particularly those cuts that are currently under-utilised.

The combination of flavourings with MI pork is a relatively new area for pork, but has been common practice of the fresh chicken industry for many years. It is anticipated that ingredient and equipment suppliers will utilise their existing experience, primarily with fresh chicken, to provide flavour solutions for pork to interested companies. This is very relevant for the development of flavoured MI pork items (e.g. kebabs, strips) suitable for the delicatessen. It is noteworthy that pork merchandising in the US is shifting away from the traditional bone-in products, with 58% of pork packages in 2004 being boneless cuts. The majority of marinated pork products in the US are produced in the meat processing plant rather than at the retail level, with injection and tumbling being the most common techniques used.

MI pork is also suitable for development of several different categories of marinated products according to their end use/market segment: ready to cook (fresh); easy to cook (e.g. microwaveable products) and ready to eat (pre-cooked) products. These opportunities are being developed in conjunction with major MI processors together with retailers.

## Value added pork products

### *Current situation in the USA*

In the US, flavoured pork represents the majority of fresh pork value added business – outperforming all other value added pork products by 50% in pounds and 63% in sales (NPB, 2004). Pork chops and pork roasts/tenderloins have the highest dollar sales in the category with flavoured roasts (including tenderloins) accounting for 34% of flavoured pork sales, followed by ribs (22%), steaks (16%), boneless chops (16%) and bone-in chops (11%). In 2004, pork had the highest percentage of ready to cook/value added packages available at retail (12%), followed by turkey at 8%, chicken at 6% and whole muscle beef (4%) (National Meat Case Study, 2004). Although ready to cook/value added products only represented 6% of the total meat case, this market is continuing to grow. The benefits of value added pork products, including pre-seasoned, pre-marinated chops, roasts and tenderloins, are the ease of cooking and minimal (if any) preparation required (NPB, 2004). The main consumers for US pre-marinated pork are women aged 25 to 84 who are looking for a delicious quick and easy meal that can be prepared and served in 30 min or less.

The National Pork Board refers to ‘value added pork’ as ‘flavoured pork items’ and includes products like pre-marinated boneless pork chops, tenderloins, loin roasts and pre-seasoned ribs. The term ‘value added refers to flavours added to the products by either a marinade or a rub, but not through smoking or curing. Within the fresh pork category there are three product types that comprise the value added segment: ready to cook/pre-marinated; ready to eat/heat and eat and specialty cuts (where the form of the product is altered by not the flavour). These three sectors of the value added segment account for 54% of fresh pork sales. Pork chops and pork roasts/tenderloins have the highest dollar sales in the category with flavoured roasts accounting for 34% of the flavoured pork category (by weight) and chops making up 41.5% of the bone-in and boneless segment (by weight).

### *Current situation in Australia*

The US situation for value added meats is not reflected in the Australian retail case. There are currently only a few value added pork products available in Australian retail stores. Independent butchers conduct this activity in-store, as required, but the major supermarkets have been selling pre-packaged flavoured/marinaded beef, lamb and chicken products, featuring a variety of flavours for several years. Flavoured/marinaded pork products are only now becoming available on retail shelves as flavoured roast products. It may be considered that these beef, lamb and chicken products are directly competing with MI pork products, as the meat content on the ingredients panel is 82% or less. This is clearly a growth area – presenting an opportunity for a range of pork products in this market, particularly case ready products and/or bulk packaged products for the retail sector.

Increased consumer demand for convenience has contributed to poultry’s success in competing with beef, lamb and pork rather than increased health awareness. The increase in poultry consumption may in a large part be due to the production of value added convenient poultry products. The poultry industry has been more responsive to the changes in consumer lifestyles and provides products that address health and convenience concerns. The proliferation of chicken products has also increased the demand for chicken and this has impacted on the consumption and market share of beef and pork. Much of the positive perception enjoyed by chicken has resulted from packaging, positioning and product form as well as pricing. The lessons learned and industry opportunities for growth, arising from providing added value products to consumers, must be adopted by the Australian pork industry in order to sustain current increases in market share for pork.

In 2005, Masterfoods and Australian Pork Limited worked together to develop a range of sauces that are suitable for a range of pork cuts. Consumer research undertaken during the development of the Pork Choices range identified that ‘a pork dedicated cooking sauce product range concept is very appealing: ‘bring it on’; ‘it is a solution to the need for variety, particularly for families’, ‘women appreciate an easy meal solution specifically for pork’ and ‘it is answering the consumer tension of not knowing how to cook pork’. Market research also identified that lack of knowledge of how to prepare pork is an issue for consumers and is solved by inaction – they just don’t buy it. The availability of sauces and condiments suitable for use with chicken has been an extremely successful way of increasing market share for chicken and marketing chicken as a convenient meat.

Retailer pressure to improve margins on fresh pork is also driving innovation on how carcasses and cuts can best be prepared and sold to maximise volume, utilise the entire carcass and reduce the incidence of product markdowns. Retailers are also demanding that their suppliers present innovative solutions to them for their consideration and acceptance. Current initiatives with pork processors are underway to deliver to retailers a new range of superior quality and premium priced pork products. This work is identifying opportunities to increase the percentage of steak and grilling cuts from pork carcasses, while reducing the percentage of large sized roasts.

It is expected that this will improve consumer perception and taste response to pork, increase fresh pork sales (measured by both volume and value), create additional value to pork producers, processors and retailers and reduce waste and markdowns in all stages of the pork supply chain and identify further areas for product development of value added pork products.

### Food service requirements for pork

Choice of cuisine, price and location are the major drivers influencing consumer decisions of fast food outlet type (AC Nielson, 2004). This presents challenges for meat-based products sold in these outlets to maintain market share and volumes sold in light of this within-store competition. The potential demand for convenience foods for in-home usage is being reduced by frequent use of restaurants and takeaway foods.

High volume institutional caterers and pubs and clubs were identified to have the biggest growth potential for pork (BIS Shrapnel 2006). The importance of the food service sector as a market sector for pork is growing due to changing consumer behaviours toward eating out and demand for convenience meals. The food service sector may also provide the pork industry with an opportunity to utilize alternative cuts from larger carcasses, potentially coupled with MI Pork, to present added value products. Currently, only 53% of Australian restaurants have pork as a main on their menu – considerable effort is being made to increase this to 75% within 2 years.

The convenience or ‘fast’ food sector in Australia was worth \$8.8 billion in 2003 (Euromonitor, 2004). Fast food, in value, is the second largest sector of consumer foodservice and by 2008 is predicted to be worth \$10.3 billion, with fast food chains holding 59.5% of the total market value. Consumer foodservice expenditure is continuing to increase due to increased demand for convenience, increased consumer purchasing power and expansion of product ranges to satisfy consumer demands for different styles of food, particularly healthier foods. Currently, 30% of Australians are eating fast food at least once a week and 64% of consumers eating fast food at least once a month.

McDonalds is the most successful consumer foodservice company in Australia, holding 34.7% market share in the fast food sector and 24.2% of market share of total consumer food service. McDonalds is the preferred fast food restaurant for 23% of Australians, followed by Subway (16%) and KFC (13%) (BIS Shrapnel 2006). Fast food chains are increasingly presenting a ‘fresh’ image to consumers, increasing the range of healthy food items. Recently, McDonalds launched a range of meal items that are eligible to be marketed with the Heart Tick. However, pork is not sold in sizeable volumes (other than as ham, bacon and salami) in this market segment and to date this still remains a considerable, but yet untapped, opportunity for the Australian pork industry. This market sector is an important and significant one to target for pork, particularly as suitable pork products that could satisfy food service requirements could potentially be obtained from lower valued and/or lower utilised cuts.

### Consumer preferences for meat

Molnar (1995) considers that the quality of food products is determined by their sensory qualities, safety, nutritional value and its convenience. Food quality is closely allied with the concept of acceptability (Cardello, 1995). Quality improvement must be driven by consumer expectations and perceptions (Issanchou, 1996).

#### *Intrinsic quality attributes*

The appearance of meat determines how consumers perceive quality and influences purchasing decisions (Carpenter *et al.* 2001). Consumer lifestyles, rather than just demographics, are increasingly being used as a more multi-dimensional basis to explain consumer behaviour. As pork at the retail level is mostly unbranded and unlabelled, consumers base their quality evaluation at the time of purchase on intrinsic factors, mainly the appearance of the product.

The major intrinsic quality attributes for meat, as documented by Grunert *et al.* (2004), include:

- cut format (e.g. steak, roast, cubed, minced);
- colour: (consistent colour, no two toning);
- fat lumps: (for steak, roast, and cubed only);
- fat rim: (for steak and roast only);
- intramuscular fat/marbling content: (for steak and roast only); and
- fat content: (for minced only).

Ngapo *et al.*, (2004), in a study conducted in France, identified the most important characteristics of fresh pork that determine consumer choice and showed how consumer segmentation in choice related to socioeconomic and cultural differences. Of the four meat characteristics studied (fat cover, colour, marbling and drip), colour was the most important criteria for pork chop selection, with leanness second. Although consumers dislike meat that forms exudates (Chambers and Bowers, 1993), drip loss rated lowest by French consumers (Ngapo *et al.*, 2004). Market research conducted in Australia (Anonymous 2000, DSM Consultants 2004) also identified that consumers prefer paler pork rather than darker pork and consumers used consistency of colour to indicate 'freshness'. Romans and Norton (1989) found that colour influenced purchasing decisions of US consumers, however, no trend for any particular colour (normal, PSE or DFD) was found. Surface characteristics such as iridescence may also be a negative appearance trait when purchasing meat (Kropf *et al.*, 1992).

Similar findings were found with consumer preferences for pork in Sweden, Ireland, Germany, Italy, UK and Spain (Glitsch, 2000). Grunert (1997), in a study of four European countries, also found that fat content and colour were the most important product characteristics that consumers use to base their quality expectations. German consumers rated leanness and colour similarly, and marbling was the least helpful characteristic (Becker *et al.*, 2000). In the US, Romans and Norton (1989) found that 81% of consumers declared that the predominant reason for selecting pork was leanness. Zuidam *et al.*, (1971), in a study conducted in the Netherlands, also found that fat was more often considered than colour. In a study of Dublin consumers, leanness was found to rank highest when making purchasing decisions, followed by presentation factors, colour and drip was sixth. No reported study has shown any correlation of preference with any particular socio-demographic or behavioural group of consumers.

High levels of intermuscular and intramuscular fat are detrimental to the purchase of pork loins. Brewer *et al.*, (2001) found that highly marbled chops with 3.46% fat appeared lighter coloured, less lean, were less acceptable in appearance and were less likely to be purchased by US consumers. This is despite higher ratings received for tenderness, juiciness and flavour of the highly marbled chops compared with leaner chops. Fernandez *et al.*, (1999a, 1999b) also demonstrated that increased levels of intramuscular fat in pork loins had a detrimental effect on meat acceptability by consumers, due to the influence of visible fat on the willingness to eat and purchase the meat.

#### *Implementation of an eating quality assurance system for pork based on intrinsic quality attributes*

Texture, tenderness and juiciness have a substantial effect on liking of cuts of meat (Chambers and Bowers, 1993). Pork purchased at the retail level in Melbourne was variable in tenderness (Hofmeyr, 1998), based on a WB shear force cut-off value of 5kg, 54% of loins would have been considered to be unacceptably tough. Consumers can misjudge the eating quality when purchasing meat and the improved quality will thus not be recognised by consumers in the shop.

Bickerstaffe *et al.*, (2001) in a NZ study undertaken in 1999, identified that pork was less tender than beef and concluded that the tenderness of pork must improve if the industry is to retain its market share. It was considered that a 20% improvement in tenderness would be possible and could be achieved by extending the aging period of pork beyond 5 days post-slaughter. Bickerstaffe *et al.*, (2001) also identified tenderness variation between pork processors, indicating an opportunity to optimise pig processing conditions.

The Eating Quality Assurance Program for Australian pork identified that post-slaughter factors including correct cooking procedures (cooking to an end point between 65-71°C), moisture infusion and ageing to improve tenderness, carcass handling (including tender stretching and low voltage constant current electrical stimulation) and sex of the pig had the greatest impact on reducing variability in pork eating quality (Taverner, 2001). Channon *et al.*, (2001) found that the incidence of unsatisfactory pork was reduced from 25% to 5% when a combination of the production and processing recommendations were implemented.

Recent market research undertaken by Australian Pork Limited has not identified any problems in consumer acceptability associated with variable tenderness and boar taint (Reference). This may reflect changes in pork distribution practices of major retailers – from carcass to boxed, vacuum packaged pork primals. Furthermore, the centralized packaging of pork into modified atmosphere retail ready packs by some retailers further extends the ageing period of pork, and potentially, reduces variability in eating quality. The improvements in tenderness due to ageing are rapid in the first 1-2 days post-slaughter and continue at a slower pace to plateau at around 6 days post-slaughter (Dransfield *et al.*, 1980-81). Rees *et al.*, (2002) found that 50% of tenderization of pork loin muscle occurred within 2 days post-slaughter and 80% within 4 days. Ageing of individually vacuum packaged pork loin for 7 days post-slaughter, rather than 2 days post-slaughter, was shown to improve sensory tenderness and WBSF by 12.5% and 25%, respectively (Channon *et al.*, 2001). Walker and Channon (2003) also found that sensory tenderness of pork loin improved by 16% and 20% by ageing pork for  $\geq 4$  days as a carcass and  $\geq 7$  days in a vacuumed pork loin, respectively, when compared with pork aged for only 2 days.

Paterson (2000) stated that the ability of the US pork industry to implement production and processing improvements to improve consistency of pork quality would result in positive implications for fresh MI pork products available to consumers. These included:

- Continued sales growth (due to increased frequency of purchase and increased number of people purchasing pork);
- Consistent delivery of guaranteed eating satisfaction; and
- Continued sales growth for flavoured pork products to offer consumers variety, versatility and easier preparation.

#### *Boar taint*

As a result of production of entire male pigs, rather than castrates, coupled with improved efficiencies relating to increased carcass weights, the risk of boar taint in Australian pork is increased. Bennett (1997) identified boar taint to be the major cause of consumer complaints relating to Australian pork. Castrated pigs are fatter and are less efficient converters of feed compared with entire male pigs. As price payment grids for pig carcasses are based on both carcass weight and fatness at the P2 site, castrated pigs more frequently incur penalties reflecting their increased fatness levels. The lack of market signals, in the form of price premiums for castrated and/or immuno-castrated pigs together with increased costs of production, means that producers have little incentive to alter current production practices for male pigs. Yet it is this 'lack of boar-taint signal' which prevents the use of an 'eating quality guarantee' when it comes to pork products in Australia.

In contrast to the 'initial lack of general acceptance' of the findings from the Eating Quality Assurance Program by the Australian pork industry, a number of companies have utilised such technologies to underpin specific eating quality pathways or assurance programs to produce a high quality pork product required by the consumer. 'Select Pork' is one such example of an eating quality pathway that was implemented by a consumer focused alliance in Western Australia in 2001. The Select Pork alliance was formed between a group of producers (10), a processor and a retailer (35 outlets). The eating quality pathway used by the Select Pork alliance involved eating quality interventions at the producer and processor level and was implemented in two stages. The Stage 1 eating quality pathway stipulated (i) Halothane free pigs, (ii) pigs with minimum of 50% Duroc sire lines, and (iii) no entire males (pork from immunological castrates, surgical castrates and females only). Stage 2 involved moisture enhancement of fresh pork.

**Table 1. The effect of the Select Pork eating quality pathway on the sensory quality of the *Longissimus thoracis* muscle (D'Souza *et al.*, 2003)**

Brand	Generic Pork	Select Pork (Stage 1)	Select Pork (Stage 1 & 2)	I.s.d	Significance
Aroma <sup>1</sup>	55	63	57	6.54	0.002
Flavour <sup>1</sup>	54	66	76	6.11	<0.001
Juiciness <sup>1</sup>	43	58	75	6.85	<0.001
Tenderness <sup>1</sup>	41	59	75	7.40	<0.001
Overall acceptability <sup>1</sup>	48	64	76	6.67	<0.001
Quality grade <sup>2</sup>	2.9	3.5	4.0	0.279	<0.001

<sup>1</sup>Acceptability score (line scale); 0 = dislike extremely and 100 = like extremely

<sup>2</sup>Quality grade; 1 = unsatisfactory, 2 = below average, 3 = average, 4 = above average, 5 = premium.

Branded pork from Select Pork (Stage 1) and Select Pork (Stage 1 and 2) were considered by consumers to have better eating quality compared to generic pork. Select Pork (Stage 1) was considered to have better odour compared to generic and Select Pork (Stage 1 and 2). However, Select Pork (Stage 1 and 2) was considered to have the best flavour, juiciness, tenderness, overall acceptability and quality grade followed by Select Pork (Stage 1) and then Generic pork. In addition, the incidence of consumers rating the pork as being below average or the pork eating quality 'fail rate' was 30%, 15% and 3% for generic pork, Select Pork (Stage 1) and Select Pork (Stage 1 and 2) respectively. It was considered that the entire eating quality pathway delivered improved eating quality and attracted a price premium and by eliminating boar taint, the Select Pork brand was able to guarantee tenderness and flavour.



### *Extrinsic quality cues*

The major extrinsic quality attributes that are relevant to meat include:

- price;
- nutritional expectations;
- brand;
- origin; and
- information on animal production system.

Neely *et al.* (1998) stated that consumer preferences are not always related to quality grades. Any form for improved or otherwise differentiated meat quality requires new ways to signal the quality to the consumer and this can be effectively done by branding. Brand can induce a high expectation and positively influence the perception of meat. In the Netherlands, the pork industry has based quality labelling on the improvement of perceived quality and associations of specific attributes. This labelling appeared to reduce quality risk perception and was expected to induce an increase in pork consumption (van Trijp *et al.*, 1995). Oude Ophuis (1994) conducted an experiment to compare the perception of free range and regular pork by Dutch consumers and found that the 'free-range' labelling had a positive effect only for consumers that had prior experience with 'free-range' pork (i.e. those that already had a positive attitude toward this type of production).

Although branding is an obvious method by which a seller can signal a superior quality, reduce consumer uncertainty and encourage consumers to pay a premium for better quality, major Australian supermarkets generally do not allow individual meat companies to market fresh pork under their own individual labels. The strength of retailer brands for fresh meat and the consumer trust that this instills is the approach used by Australian retailers, especially for beef, lamb and pork.

Brand is the predominant quality cue used by low familiarity consumers. Among high familiarity consumers, the brand is also important, but is similar in importance as perceived fat and meat colour (Grunert *et al.*, 2004). Interestingly, low familiarity consumers fail more in their quality predictions at the moment of purchase. Both high and low familiarity consumers use branding as the major cue for forming expectations about the health quality.

Quality can also be perceived during cooking. The degree to which the meat shrinks during cooking was identified by Chambers and Bowers (1993) to be an important sensory cue.

The demand for organic food products has increased dramatically in Europe over the past decade (Beckmann, *et al.*, 2001; Squires *et al.*, 2001). Bjerke, 1992 suggested that health and environmental concerns are two major motives for choosing organic products. Bech-Larsen and Grunert (1998) conducted a study in Denmark and Great Britain on consumer attitudes toward organic pork. The major reasons identified for choosing or not choosing organic pork were: animal welfare, budgetary restraints, health, and enjoyment, with for animal welfare concerns more important to British than Danish consumers. Budgetary restraints were the important motive for not eating organic pork by consumers in both Great Britain and Denmark. Consumers associated organic production with good health, animal welfare and concern for the environment as well as good taste. This infers that 'organic' is no longer a credence characteristic, but is also partly an experience characteristic, where expectations can be confirmed or disconfirmed after the purchase. Scholderer *et al.* (2004) measured the expected and experienced quality of pork chops where production system (conventional vs. organic) and extrinsic cues available to consumers (none/conventional/free-range/organic) were varied. Although organic pork received lower scores for experienced quality after consumers tasted samples, when consumers believed that they tasted organic or free-range pork, they rated the perceived quality of the meat higher, irrespective of which type of meat they actually ate.

Many consumers believe that meat bought from a butcher is of better quality than meat bought from a supermarket (Becker *et al.*, 2000; Bernués *et al.*, 2003; Glitsch, 2000; Grunert, 1997). McIlveen and Buchanan (2001) showed that information available at the place of purchase – a butcher, a low and a high quality supermarket – affected the sensory evaluation of meat samples.

Grunert (2006) conducted a simulated shopping study with 299 German consumers where respondents had to choose between four packs of pork chops and the only information on these pork chops was price, origin, animal welfare in the production process and guarantees for the absence of pesticide residues in the meat. Respondents had to click with mouse on the 'pack' to obtain this information. It was found that the average time for making a purchasing decision of pork chops was 20.2 seconds and during this time, respondents managed to click 3.1 times to get price information, 2.7 times to get information on origin and 0.9 times to get information on either residues or on animal welfare.

### *Influence of intrinsic and extrinsic drivers on consumer meat purchasing behaviour*

Moskowitz (1995) concluded that it is not sufficient to state that a product has a 'high quality' to motivate a consumer, as quality must be supported by a specific concrete benefit for the consumer. Brunso *et al.* (2002), in a review collating research into consumer food choices and perceptions of quality, conducted a Total Food Quality Model to explore consumer attitudes to beef and pork in several European countries. Quality perception of meat was influenced by objective factors, including muscle conformation, fat distribution and colour, as well as by subjective factors (e.g. credibility of health statements, claims for superior farming and animal rearing techniques).

Bredahl *et al.*, (1998) conducted a study to investigate the relationships between intrinsic quality cues, expected quality, experienced quality and physiological product characteristics on quality perceptions of pork loin chops. A total of 200 German pork consumers who had the main household responsibility for food shopping and cooking participated in the study. The physiological product characteristics measured were: presence of the halothane gene, PSE, pH, colour, blood splash and intramuscular fat. Consumers were firstly shown real, fresh samples of three kinds of pork and asked to evaluate the samples by completing a questionnaire for the four intrinsic cues: colour, fat level, fat marbling and meat juice. Each consumer was then given colour-labeled samples of each of the three kinds of pork and asked to prepare (using a familiar preparation method) and consume the meat for dinner at home over the next three days, in a pre-determined order. Nutritional value, wholesomeness, freshness, leanness, juiciness, taste and tenderness were assessed. These quality criteria were used by consumers to form expectations about the quality of pork in a purchase situation and evaluate eating quality after preparation and consumption as well as to measure consumers' quality expectations of the raw and cooked pork. A very strong relationship between visual appearance and expected quality was found. Eating quality was only moderately related to expected quality, with only 24% of the variance in experienced eating quality explained in the model. This study highlighted that consumers experience considerable difficulty in forming quality expectations that is predictive of a later quality experience. Objective measures of product quality were also shown to be weakly related to quality as experienced by consumers. The inability of consumers to predict their quality experience after purchase is partly due to the misinterpretation of intrinsic quality cues, especially intramuscular fat (Grunert *et al.*, 2004).

Process traceability attributes (including production method and origin) are of interest for particular market segments in response to meat quality concerns. However, functional traceability attributes (including organizational efficiency and chain monitoring) were not considered to be as important as a basis of market segmentation as they do not address concerns about food safety, healthiness, environment or animal welfare.

Grunert *et al.* (2002), as quoted by Grunert *et al.* (2004), conducted a study to determine whether additional information at the point of purchase would assist consumers to form quality expectations about fresh meat. A total of 20 objective characteristics for pork were screened with regard to quality dimensions of relevance to consumers. It was found that those characteristics that consumers both believe they understand and which they regard to be as important rated highly. Interestingly, these characteristics were all health or process-related rather than eating quality related. It was concluded that consumers do not believe that their judgments of eating quality are improved by providing more information at the retail level. Consumers tend to entrust sensory dimensions of quality to an expert, like a butcher, than try to arrive at better quality evaluations on the basis of better information. Butchers are also able to directly communicate with their customers about the quality and source of meat sold while in the supermarket, labeling can only partly carry out this role.

These studies demonstrate that the meat industry potentially has greater hurdles to overcome than other food sectors as the measures that consumers use to judge a good piece of meat (i.e. it is tasty, tender and succulent) are often misleading and leave them less likely to choose meat which won't deliver against their expectations.

Despite the increased amount of information available to consumers through labeling, traceability systems and quality assurance schemes, the effect on consumer trust in meat as a wholesome and safe product is limited (Gellynck *et al.*, (2006). This may be due to misunderstanding and misinterpretation of consumers arising from the overload and complexity of information on food products. Trust differs from confidence as trust recognizes the presence of a risk, while confidence does not (Luhmann, 1988). Gellynck *et al.* (2006) argued that retailer interest in traceability schemes is driven more by procurement management efficiency rather than safety or overall quality. Interestingly, quality assurance schemes were found to have a poor impact on consumer perception.

Grunert (2006) stated that the role of extrinsic cues in quality perception of meat is increasing. Meat quality is increasingly being inferred from information about the meat, which opens up further opportunities for more differentiation in meat products. This was considered to also pose new requirements for the organization of the meat value chain to fulfill the functions of delivering both meat and information.

Consumers are requiring other retail channels for specialized meat products, in addition to purchasing meat in supermarkets, particularly when these products rely on the credible supply of extrinsic cues. Fragmentation and diversification of the market for meat and meat products is also occurring, reflecting the marketing methods used for meat products being sold on the basis of extrinsic cues. Grunert (2005) considered that as possibilities for product differentiation develop in response to the increasing diversity of consumer lifestyles and requirements, including the demand for convenience, the more diversified the products will become. This diversification in products may be accompanied by a diversification in retail channels (Grunert, 2005).

### **Drivers of meat purchasing decisions**

Three major trends are influencing consumers' food choices – convenience, health and wellbeing and indulgence or personal satisfaction. Meat products offered for sale in both retail and food service need to demonstrate through their composition, design and marketing positioning that these trends have been fully understood and addressed.

Consumers worldwide are increasingly willing to pay for convenience, variety, health and safety (Haselgrove, 2006). The increase in meat-rich diets for emerging nations is about diet variety. AC Nielson (2006) identified several overarching key global trends relating to consumer purchases in food and beverages during the period of July 2005 to June 2006, namely continued focus on health and freshness; the need for convenience and continuing need for value.

#### *Convenience*

Planning what to cook is a key challenge and it is the planning, rather than the actual cooking, that creates the most amount of pressure on consumers. The demise of formal mealtimes, with increased frequency of informal and fragmented dining is well documented. Furthermore, Calvett (2006) stated that the incidence of 'Stuck at Desk' eating and 'Dash Board Dining' (in car eating) by consumers is increasing.

As many consumers are challenged by the need to come up with a wide repertoire of meal ideas, they require information on new ways to prepare quick, healthy and easy to cook meals. Importantly, meat consumers can be influenced in-store on their product purchases and it has been shown that the largest proportion of time in the supermarket is spent browsing the meat aisle (DBM Consultants, 2004). Consumers generally do little planning of meals, with most decisions made the day of the dinner and at the end of the day (Resurreccion 2003). Consumers are no longer looking for pork to prepare occasional /special occasion meals – they are increasingly looking for suggestions to enable them to confidently and successfully cook pork (DBM Consultants, 2004). For families, it is critical that new and innovative ideas are developed and communicated to encourage children to eat a wide variety of meat to meet their nutritional requirements. This opens up many opportunities for the development and marketing of value added pork products.

Changing lifestyles has led to the shift toward more convenience in food preparation. It has been found that areas with high rates of women in the workforce are associated with a less diverse range of goods purchased. Households with these characteristics purchase fewer traditional meats such as roasts for at-home meal preparation but purchase more prepared products (USDA/ERS 2002). Multi-income households do not pay as much for fresh meat due to the preparation time required.

Senauer (2001), in a US study, found that 55% of respondents indicated that convenience is 'very important' in their food purchases. In Western countries, the share of meals eaten outside the home is increasing. Darian and Cohen (1995) suggested that convenience can cover any savings of time, physical or mental energy that occurs during meal preparation at home: deciding what to eat, purchasing, preparation, consumption and cleaning up. This definition of convenience therefore encapsulates more than just ready-made meals or eating out.

Many cuts are interchangeable between pork, beef and lamb. Overall, consumers look for good value for money when purchasing meat and are not prepared to forego quality for a cheaper price. Many consumers also tend to purchase the same cuts due to familiarity and previous enjoyable eating experience with the cut. For roasts, consumers will look to find the lowest cost roast available regardless of species as the roast meal experience is not something that is dependent upon the cut or species used (DSM Consultants 2004). It is also possible to interchange the type of meat and cut used to prepare stir fry meals and casseroles. The challenge for the Australian pork industry is how to successfully develop and market products that are differentiated from its competitors and be cost effective to produce by utilising significant volumes of lower valued pork cuts.

Convenience represents a desire by consumers to seek out products that effectively and efficiently allow them to juggle the multiple tasks that need to be accomplished each day and to increase the time for priorities (including family, social, leisure time). Convenience can be delivered in a number of ways – prior preparation of meat items removes the need for consumers

to have an in-depth knowledge of meat cuts (both reflecting and acknowledging the skills gap that exists in the general public). Meat that is partly or fully prepared and ready to cook in its tray also removes the need for consumers to handle meat. There is a continued need to position pork as a quick and easy solution to fit into consumers' busy lives. More consumers are choosing to eat away from home or purchase more products that are prepared outside the home or partially-prepared.

Over the past several years, Australian supermarkets have begun to offer a variety of value-added, pre-packaged and case ready meat products. On a global level, the meat industry has responded to the convenience trend mainly in the ready meal category, where many products have a meat component. de Boer *et al.* (2004), in a major study of demand for convenience products and services in Ireland, demonstrated that different lifestyle dimensions are associated with demand for different types of convenience products and services. The range of products available differs considerably between countries, but the bulk of these products are targeted more at the 'uninvolved' than at the 'food-loving' consumer segments.

Perceived scarcity of time and stress in daily life can affect consumer behaviour. Scholderer and Grunert (2005) showed that convenience orientations act as a mediator between perceived resources (in terms of disposable time and money) and convenience-oriented behaviour (like buying convenience foods). Convenience orientation was also shown to be influenced by food-related motives. Resurrecion (2003), quoting USDA/ERS (2002), stated that a 10% increase in income was associated with a 0.7% increase in demand for ready-to-eat meals.

Meal preparation time has also decreased, from 3 hours per day after World War 2, to less than 30 minutes today (Calvett, 2006). Consumers are therefore looking for products that can be quickly and easily prepared. Knowledge about cooking has also declined, so meat cuts that require some skill for preparation and a longer cooking time have been substituted by poultry. This has also been reinforced by a larger proportion of single person households that are more convenience orientated since they are only cooking for one person (Jensen *et al.*, 1994).

The increased interest in faster and easier to prepare main meal dishes such as pizza and pasta and the introduction of new dishes have also induced a substitution for meals with less or no meat. The erosion of formal 'at the table' and 'meat and three veg' eating occasions could equate to the erosion of our own market for meat. From a home cooked meal where 50% of the meal's weight could be meat, a more convenient alternative e.g. pizza or a pasta meal may contain only 10% of its weight as meat. This has the potential to diminish our 'share of stomach' and our sales volumes unless we can make our meat products highly relevant to the consumers' new eating patterns. This poses new and significant challenges for our industry to respond to.

However, Grunert *et al.* (2001) stated that the more a product is differentiated, the less it is likely to appeal to consumers at large, because consumers differ in their preferences, their ways of shopping, preparing meals and eating. Consumer-oriented product development, also in the meat-sector, will therefore typically require a segment-specific approach (Grunert and Valli, 2001).

### *Meat avoidance*

There is also an increasing trend towards meat avoidance by consumers, particularly young females (Kubberød *et al.*, 2002). Although these consumers may be offended by blood, raw meat and links to certain animal body parts, this does not necessarily imply that they will become vegetarians. This does however identify that there are opportunities to deliver meal preparations where meat, and its animal origin are less prominent, for example from higher forms of processing. However, it is not clear how strong the meat avoidance trend is nor whether it is growing. Convenience and meat avoidance may, in terms of product development, point at similar directions, namely at meat-based products with a high degree of processing, detached from their animal origin, and adapted to different motives of different consumer segments (Grunert *et al.*, 2002).

### *Animal Welfare*

Van Trijp (1994) demonstrated that Dutch consumers express doubts about the animal friendliness of pork and poultry production. Consumers who are concerned with animal welfare may stop eating meat if production systems do not reflect their concerns (Lister 1995). Several European studies, including Holm and Mohl (2000) and Ngapo *et al.* (2004), demonstrated that although consumers are concerned about animal production systems, the link between negative images of production and purchasing behaviour were very weak. Lister (1995) suggested that consumers will not pay extra for what they consider to be a normal production system.

### *Food Safety*

Under normal conditions, most consumers are not anxious about product safety, however a certain fear may be present in a latent state. One of the major outcomes from implementing food safety measures is to reduce uncertainty and increase consumer trust in food. As consumers like to assume that all food on sale in supermarkets is safe, Grunert *et al.* (2004) considered that trying to position a differentiated product on food safety issue could hurt the category as a whole. Consumer concerns about avian influenza and bovine spongiform encephalopathy and development of Creutzfeldt-Jakob disease have devastated poultry and beef markets in recent times. The return to the previous confidence levels is difficult to restore, as demonstrated in Japan.

### *Healthiness*

Healthiness is a traditional credence characteristic. Most consumers believe that meat can be part of an inherently healthy diet. The healthiness of pork is related to the natural composition of pork and therefore its healthiness for consumers (Kanis and de Greef, 2003). Although pork is commonly perceived as relatively fatty and unhealthy, the intramuscular fat content of pork is on average lower than beef and lamb (Verbeke *et al.*, 1999). Channon *et al.* (2001) found that the intramuscular fat content of Australian pork averaged 0.98% in pork loin.

Fourteen fresh pork cuts, out of 22, are eligible to be labelled and sold with the National Heart Foundation's Heart Tick (Barnes *et al.*, 1996), as they meet the criteria for fatness under the category 'Meat – Plain'. However, moisture infused pork, with a neutral flavour, is not eligible for the Heart Tick as the National Heart Foundation view this product to be 'Meat – Plain' despite it being classified as a 'Manufactured Meat' under the Food Standards Code. As the National Heart Foundation regard marinated meats to be under the Meat – Processed category, any development of flavoured brines for MI pork production will enable products produced to be eligible to use the Heart Tick as long as the products meet fatness and sodium limits stipulated by the National Heart Foundation. These limits are unable to be published as they are only released to food companies interested in being involved in the Heart Tick program. All rights in the Guidelines for Tick Approval (including copyright) are owned by the National Heart Foundation of Australia. The Tick Guidelines (and related documentation) is not able to be distributed to any third party or used other than for the purposes of assessing a product for potential Tick approval.

In the US, the number of evening meals prepared at home from scratch accounted for only 32% of all evening meals (MSI, 2005). Convenience foods represented 26% of evening meals, with 23% were consumed in a restaurant. When consumers buy their ready meals, they are increasingly turning to refrigerated options, as these are perceived to be healthier and fresher. 'Fresh' is the most desirable food label claim and was extremely/very important to 62% of food shoppers (Health Focus, 2005). Consumers also believe that refrigerated meals use fresh ingredients and contain fewer additives (R&M, 2005).

Cowan *et al.* (2001), in a study conducted in Ireland, found that Irish meat processing companies are committed to meeting retailer demand for a reduction in additives and preservatives and requires assistance in developing processes that can result in additive free and preservative free products. Market growth opportunities for novel, nutritionally enhanced meat products targeting the adolescent market was also identified (Cowan *et al.*, 2001). There is great potential for convenience foods, particularly quality chilled meat products.

### *Nutritional enhancement of pork*

Lean pork is an important component of human diet, when eaten in moderation (O'Dea and Sinclair, 1993), as it is a good source of protein, omega-3 fatty acids, vitamin B12, niacin, zinc and iron (D'Souza *et al.*, 2005). As previously reviewed by D'Souza *et al.* (2005), meat consumption, including pork, has been linked with an increased incidence of obesity, Type 2 diabetes, and cardiovascular disease. Cardiovascular disease is Australia's largest health problem and greatest killer (50, 294 deaths in 2002) and affected 3.67 million Australians in 2001 (National Heart Foundation 2004). Currently, 60% of Australians are overweight or obese, similar to levels reported in adult Americans (Bray, 2005). Increasing body weight increases the risk of early mortality as well as enhances the risk of developing diabetes, high blood pressure, cardiovascular disease, gall bladder disease, osteoarthritis and some forms of cancer. The consumption of diets high in fat, increasing portion sizes, excessive consumption of sugar-sweetened drinks and food products, abundant variety of foods in supermarkets and food service outlets are key dietary factors in the development of obesity (Bray, 2005). Consumer studies conducted by Mintel (2005) highlighted the need for new meat products that span more than one 'mega' trend. It was considered that for a product to succeed, it must 'convey a bundle of intrinsic and extrinsic attributes that better meet the rational and emotional needs of the different consumer groups within the overall shopping base'.

Arihara (2006) stated that the consumption of meat and meat products is increasingly avoided by consumers

to reduce the risk of cancer, obesity and other diseases, even though meat plays a critical role in the maintenance of human health. The outbreak of bovine spongiform encephalopathy (BSE), foot and mouth disease, avian influenza in poultry, *Escherichia coli* O157, *Salmonella* spp., *Listeria monocytogenes* and other food safety outbreaks have impacted on global meat demand, particularly beef and poultry. The demand for meats in the United States has changed due to increased consumer emphasis on nutrition, health and diet, saturated fat, cholesterol and obesity (Resurrecion 2003). Increased health concerns have resulted in the inclusion of more fresh vegetables and fruit, a shift from high fat, high protein diets, in the American diet. Nutritional concerns about fat and cholesterol have resulted in increased demand and production of leaner animals and removal of greater amounts of fat from retail cuts.

Dietary fat intake, rather than protein, is the major contributor to the increased incidence of these three health conditions (Scrimshaw and Guzman, 1968). Over the past five years, the food industry has paid significant attention to the fortification and the nutritional enhancement of a number of functional properties of everyday foods. There are a number of opportunities to develop and market pork products with enhanced nutritional value and with a higher bioavailability of nutrients. It is also considered that as the population ages, the increased cost burden on health care may be eased by the development of functional food products from animal proteins. As consumers experience expands, this will drive higher expectations of the meat industry to deliver the ultimate combination of attributes demanded by consumers. Issanchou (1996) concluded that health quality becomes a determinant when there are also dietary recommendations related to the use of the products.

As the pig is a monogastric, it is possible to improve the nutritional quality of pork via dietary manipulation of the pig. However, this comes at a cost and would need to be recouped from consumers to make the venture viable (D'Souza *et al.*, 2005). Arihara (2006) concluded that hurdles still exist in developing and marketing novel functional meat products as these products are unconventional and many consumers perceive that meat can negatively impact on their health.

Goldberg (1994) identified twelve broad groups of ingredients to have potentially beneficial effects for human health, including dietary fibre; oligosaccharides; sugars/alcohols; amino acids, peptides and proteins; glucosides; alcohols; isoprenes and vitamins; choline; lactic acid bacteria; minerals; unsaturated fatty acids and antioxidants.

Grunert *et al.* (2004) stated the differentiation of products based on health should aim at positive health effects, such as the development of meat-based functional food products. Bech-Larsen and Grunert (2003) detailed pitfalls in the market for functional food products in Europe, stating that legal restrictions hampering the communication of health benefits and scepticism of European consumers to any product modifications that are regarded as 'unnatural' are major issues. Perceived unhealthiness of animal fat was suggested to be one area of potential development (Bech-Larsen and Grunert, 2003). The Pork Co-operative Research Centre for an Internationally Competitive Pork Industry (Pork CRC) is focussing, as part of Strategy 3, on the development of retail ready, fresh pork products produced from pigs that have received diets enriched with omega 3 fatty acids, selenium or conjugated linoleic acid (CLA). Therefore, marketing and promotion of functional pork products will be an important issue to consider at the outset. In Australia, functional foods are available at the retail level, but to date not in the fresh meat cabinet.

### *Selenium*

The relatively low levels of selenium (Se) in soils in Australia and New Zealand has resulted in low Se content of primary products produced in these areas. Although Se intakes of consumers in Australia and New Zealand are sufficiently high that deficiency symptoms are not expressed, Dunshea *et al.* (2005) suggests that there is growing evidence that the relatively low intakes may contribute to elevated levels of risk for some cancers. D'Souza *et al.* (2005), in a review on pork as a functional food, detailed many studies that have been conducted that indicate that increased dietary Se intakes, particularly organic Se, impart additional health benefits on the immune system, including reduced viral virulence, reduced cancer risk, reduction in HIV symptoms and progression, reduced risk of cardiovascular disease, and reduced pain from rheumatoid arthritis. Mahan *et al.* (1999) found that organic selenium was incorporated into pig muscle at a linear rate, with no response observed when selenium sulfite was supplemented to pigs. Selenium also has a positive impact on pork quality as it has been shown to significantly reduce cell membrane oxidation which then reduces muscle drip loss.

Selenium enriched pork is now available at the retail level in Canada (by Prairie Orchard Farms – also able to label its pork as being a source of omega- 3 fatty acids) and Korea (as SelenPork).

### *Unsaturated fatty acids*

Fish meals and oils used in pig diets contain high concentrations of polyunsaturated long-chain fatty acids. Fatty acids C18:2 and C18:3 predominate in some oils whereas eicosapentanoic (C20:5), docosapentanoic (C22:5)

and C22:6 occur in high concentrations in others, particularly in fish species from cold waters. Feeding fish oils to pigs can increase the concentration of long chain polyunsaturated fatty acids in their tissues (Irie and Sakimoto, 1992; Leskanich *et al.*, 1994). Dunshea and D'Souza (2003) found that the amount of fat increases rapidly during growth of the pig and originates both from the diet and from de novo synthesis. Madsen *et al.* (1992) reported that 75% of carcass fat resulted from de novo synthesis indicating that it is relatively easy to manipulate the fatty acid composition of pork. Howe *et al.* (2002) evaluated the inclusion of 15% PorcOmega, a stabilized tuna fishmeal formulation, for 42 days per-slaughter as a source of DHA for enrichment of pork products. It was shown that the long chain n-3 PUFA in pork forequarter and leg chops was increased 3-fold and no adverse effects of sensory qualities of pork were found.

There is increasing interest to manipulate the fatty acid composition of pork to achieve a ratio of polyunsaturated to saturated fatty acids of higher than 0.4. More recently, the ratio of n-6:n-3 polyunsaturated fatty acids has been investigated. In pork, this ratio is 7:22 compared with 2:11 and 1:32 in beef and lamb, respectively (Enser *et al.*, 1996). Wood *et al.* (2004) stated that this ratio should be less than 4 to reduce risk of cancer and coronary disease.

However, increasing the amount of unsaturated fatty acids in pork may result in an increased possibility of lipid oxidation. This can be minimized by supplementing animals with antioxidants including Vitamin E and selenium to reduce rancidity, prolong shelf life and retain colour.

### *Conjugated linoleic acid*

Conjugated linoleic acid (CLA) is composed of a mixture of positional and geometric isomers of octadecadienoic acid, with conjugated double bonds located at positions 7,9-, 8,10-, 9,11-, 10,12- or 11,13- on the carbon chain. Dietary CLA is normally supplemented as a mix of these isomers, with the predominant isomers being the cis/trans-9,11 and the trans/cis-10,12 isomers (Dunshea *et al.*, 2005). CLA has antioxidative and immunomodulative properties (Azain, 2003) and may also play a role in the control of obesity, reducing the risk of diabetes and modulation of bone metabolism.

### *Functional pork products*

Goldberg (1994) stated that the three basic requirements for a food to be regarded as functional are: i). it is a food derived from naturally occurring ingredients; ii). It can and should be consumed as a part of the daily diet and iii). it must regulate specific biological processes, once ingested.

Although low fat meat products have been developed, to varying degrees of commercial success, in many countries, Arihara (2006) stated that the meat industry has been hesitant to adopt the functional trend and to introduce additional physiologically functional properties into meat products. Dietary fibre from oats, sugar beet, soy protein, apples, peas, probiotic lactic acid bacteria have been used in the formulation of meat products, such as sausages and patties (Fernández-Ginés *et al.*, 2005; Jiménez-Colmenero *et al.*, 2006). Soy proteins are considered to be effective for preventing cardiovascular disease, cancer and osteoporosis and are commonly used in the manufacture of processed pork products. However, some products that are suitable for addition to pork as functional ingredients may also impart allergenic responses in humans.

## **Packaging**

### *Fresh meat*

US demand for packaging used by the meat, poultry and seafood industries will exceed \$11 billion by 2008, partly driven by the trend toward convenience food. Case ready packaging, including modified atmosphere packaging and vacuum packaging, to extend shelf life and provide protection from puncturing and product tampering during shipping, is also expected to drive above average demand for trays and films. In the US, continued growth of packaging will reflect additional branded meat products often in ready to cook formats. Increasing demand for pre-packaged meat in food service will also drive demand for packaging.

Eilert (2005) noted that the majority of the significant innovations taking place in the packaging sector are occurring in other industries outside of the meat industry. Innovations in meat packaging equipment appear to be limited to increasing speed and efficiency of current packaging formats. The ability of materials to offer flexibility in primary processing and reheating as well as enable wider distribution and extended shelf life of meat products will be critical to its market success. However, innovations in packaging must also be mindful of consumer scepticisms of meat processors, in that meat retailers are out to deceive consumers, hide poor quality, fatty and grisly meat under labels and use brightly coloured opaque trays to obscure it. Transparency of packaging is critical as is building a reputation through ongoing adherence to high quality standards to allow consumers to have full confidence in the credence of quality claims.

Mize and Kelly (2004) reported on a study conducted by the Cryovac Division, Sealed Air Corporation, the National Cattlemen's Beef Association and the National Pork Board to audit and report trends in US fresh meat packaging at the retail level. It was found that in 2002, 69% of the linear footage of the retail meat case in supermarkets was occupied by fresh meat and poultry. However, by 2004, this had declined to 63%, reflecting an increasing market share of products catering to increasing consumer convenience. These products included fully cooked products, marinated meats as well as processed products including hams and sausages. It was also found that the proportion of case ready packages (those not repackaged in back of store) increased from 49% in 2002 to 60% in 2004. For pork, in 2002, 37% of pork was presented in case ready packaging compared with 50% in 2004. In 2002, ready to cook products represented 7.1% of the linear feet space of US retail outlets while 10.6% of linear feet shelf space was occupied by ready to eat products. Case ready penetration of chicken and turkey has continued to increase at the retail level with 95% of packages audited in 2004 being case ready (Mize and Kelly 2004). This was considered to reflect the marketing efforts of poultry companies, their distribution systems, and uniformity in product quality, particularly colour, throughout the distribution period.

Mize and Kelly (2004) also identified that as the proportion of case ready meats increased, meat packaged in Styrofoam trays with a polyvinyl chloride wrap declined to 47% in 2004 from 51% in 2002. The use of modified atmosphere packaging (MAP) and vacuum packaging increased, from 9% in 2002 to 13% in 2004 and 10% in 2002 to 13% in 2004 for MAP and vacuum packaging, respectively. In Australia, centralised packaging of fresh meat and delivery to retail stores, as either case ready product or as boxed packaged primals, has changed the types of packaging used for retail sale of meat in supermarkets. This has been in response to economic efficiencies, increased shelf life of meat as well as reflects the lack of skilled labour required for meat fabrication at the retail level. Furthermore, increased opening hours of Australian supermarkets has also driven retailer demand for pre-packaged fresh meat to be delivered.

In Europe and UK, case ready packaging is growing at 15% per year and is one of the fastest growing segments of the self service sector. A large variation in the use of case ready packaging in European countries was identified by Salvage (2005). In the UK, 90% of meat is presented in case ready packaging while in Italy, only 10% is presented in this format.

In the UK, the major supermarket chains all utilise tray sleeves to improve product presentation of modified atmosphere packaged meat products, both value added and raw cuts. These adhesive backed labels, that can contain graphics and information including price, weights, use by and packed on dates, can be applied as either top, top and side, c-wraps, top and two sides or full wrap, eliminating the need for use of non-recyclable backing papers. A sleeving machine is used to apply these labels to packs to eliminate the need for additional package handling and is able to operate at high packing speeds. This equipment and tray sleeves is only now available for use in Australia.

#### *Current case ready packaging of fresh meat*

Modified atmosphere packaging of fresh meat generally use clear or colored barrier lined trays (made from polystyrene, polypropylene or polyethylene) with a clear or printed barrier film. This package style has a headspace ratio of about 1:1 and contains an atmosphere of 70-80% O<sub>2</sub> and 20-30% CO<sub>2</sub>. This headspace ratio is required to expose the meat product to a minimum oxygen concentration of 55% during its shelf life, in order to optimize meat colour (Jakobsen and Bertelsen, 2000). The presence of oxygen in the pack results in the formation of oxymyoglobin and CO<sub>2</sub> acts as a bacteriostatic agent (Bartkowski *et al.*, 1982). Modified atmosphere packaged meat is centrally packaged and then distributed to retail stores. However, the product shelf life is limited to about 10–12 days for pork.

Carpenter *et al.* (2001), in a study conducted to investigate whether consumer preference for beef colours (red, purple and brown) and for packaging of fresh beef and patties (MAP, vacuum packaging, conventional overwrap with polyvinyl chloride), found that although consumer preferences for beef colour and packaging influenced likelihood to purchase, it did not bias eating satisfaction.

#### *Use of carbon monoxide in case ready meat packages*

The inclusion of carbon monoxide (CO) in fresh meat packages has been practiced in Norway since 1985 to maintain a stable bright red meat colour, reduce lipid oxidation and prolong microbiological shelf life without compromising operator or consumer safety (Sorheim *et al.*, 1999). However, this is no longer practiced as the use of CO is not permitted by the EU. The gas mixture used for pork packaged in case-ready, modified atmosphere packs in Australia is 70-80% O<sub>2</sub>; 20-30% CO<sub>2</sub>. The shelf life of pork packaged in MAP can be 10-12 days depending on cold chain maintenance. CO can improve the colour stability of meat due to the formation of carboxymyoglobin (COMb), which has a similar colour to oxymyoglobin. At a storage temperature of ≤ 4°C and using a gas mixture of 0.3-1.0% CO/60% CO<sub>2</sub>; balance N<sub>2</sub> in packs, a desirable colour of red meats can be maintained for at least 21 days while the



product can remain microbiologically stable for 11-21 days. Levels of CO of less than 0.3% may be suitable for lighter coloured meats including pork. Storage of bone-in meat in anaerobic atmospheres containing CO can also prevent bone discolouration. Furthermore, the use of low CO gas mixtures are not explosive unlike the high risk of explosions resulting from the use of high concentrations of O<sub>2</sub>. The use of CO eliminates the need for including oxygen in the gas mixture and the anaerobic environment also prevents the development of lipid oxidation.

Despite this, there has been considerable concern, particularly in the US where the US Food and Drug Administration permitted its use as a processing aid when included at 0.4% CO in a master pack system, that this practice could mask meat spoilage. The FDA stipulated that “the case ready meats would be removed from the MAP system prior to retail display” unlike the use of CO in sealed case ready trays in MAP in Norway. However, Eilert (2005) stated that in 2004, the FDA issued a decision that low levels of CO did not mask spoilage and although colour did not degrade in a package containing CO, offensive odours could still form in the presence of CO. This led to a new format of case ready packaging to be used in the US.

Australian retailers do not accept the use of CO in case ready packaging for retail sale of meat. This reflects that CO can only be used as a processing aid rather than as a food additive according to the Food Standards Code (FSANZ 2004). The main distinction between a processing aid and an additive in the Food Standards Code is that a processing aid does not perform a technological function in the final food. An application would be required to be submitted to FSANZ to consider the use of CO as a food additive.

#### *Low oxygen systems for fresh meat*

Low oxygen packaging systems have not been widely implemented in the US (Eilert 2005) or in Australia, compared with the use of high oxygen packs. This system extends the shelf life of meat by distributing it in its myoglobin state and presenting the product at retail in the oxymyoglobin state. This package consists of a barrier lined tray and a clear barrier film. The film has two layers: an exterior peelable barrier exposing an oxygen permeable film, sealed to the tray, that enables the product to bloom at retail once the exterior film is removed. The package has no headspace requirement and can contain an atmosphere of 80% N<sub>2</sub>:20% CO<sub>2</sub>. Fresh meat is particularly susceptible to discoloration due to the formation of metmyoglobin by a partial oxygen pressure in the range of 5–10 mm of mercury (Sebranek and Houser, 2006). The residual O<sub>2</sub> requirement must be below 0.05% after packaging, and essentially zero within 24 hours following packaging (Solomon, 2004). Although there has been interest in this packaging format for use with pork in Australia, additional labour required to peel the exterior layer together with the requirement for in-store labelling onto the oxygen permeable film has limited its introduction at the retail level. If the external layer is not removed, the product will be displayed in the myoglobin state - not preferred by retailers. This packaging type can increase distribution life by 5–10 days in the low-oxygen state depending on the cut and the headspace of the product is reduced by 50%.

#### *Thermoform vacuum packaging innovations for fresh meat*

The use of thermoform packaging systems, or form-fill-seal machines, is not new and is not widely used for the vacuum packaging of fresh pork primals due to the requirement for different sized dies to package different sized primals.

Shrinkable films have now been developed for use with horizontal thermoform packaging machinery (Salvage and Lipsky, 2004). These films are being used by the Australian meat industry, but not yet for pork. This development addresses issues related to the formation of wrinkles and excessive film use. It is considered that this application will continue to evolve due to the advantages it offers over the use of vacuum bags and traditional form-fill-seal operations. This package style involves the use of a barrier tray (styrene or polypropylene) and a barrier film that forms around the product to reduce any purge coming out of the product. An additional web of film or a header can be added for pricing and labelling during packaging. The product shelf life can be 15–22 days depending on the cut. As the product is displayed in the myoglobin state at the retail level, colour deterioration and oxidation are minimized in the display case.

#### *Packaging formats for value added meat products*

It is estimated that the meal kit category in the US will increase from US\$11 million in 1998 to \$50 million in 2008, in response to providing consumers with convenient meat products. This increase in demand is considered to be in response to an ageing population minimal cooking skills of typical consumers and reduced time for at-home meal preparation due to employment and lifestyle pressures.

Packaging solutions for prepared meat items are continuing to evolve to meet the needs of a widening number of eating venues and situations. These include: on the move, easy open, non-spilling/resealable, ovenable/microwaveable, self heating. The development of Simple Steps by Sealed Air Corporation (Parlin, 2004) combines vacuum skin packaging of meat into a thermoformed cook-in tray. This package can be processed to just below 90°C and can be placed in the microwave for reheating without using any utensils to puncture the film. The lidding material is self-venting, and the package is designed for optimal reheating in the microwave. The package is also fitted with an easy opening, peelable tab. Unfortunately, due to the relatively small size of the Australian market, the Simple Steps system has not yet been introduced into this market.

Modified atmosphere packing trays that have compartments that enable segregation of sauces/condiments from raw meat are increasing in acceptance and prevalence in overseas markets, particularly in the UK. These packs provide consumers with convenient meal ideas that still require consumers to cook the product and then easily and quickly assemble the meal.

Microwaveable trays that contain a pre-cooked meal, packaged in a low oxygen gas mixture with 70% N<sub>2</sub>:30% CO<sub>2</sub> and lidded with a barrier film are currently available in the US. Controlled temperatures, excellent hygiene and high barrier packaging are pre-requisites during production in order to achieve an extended shelf life of 21 to 35 days. These types of products do not typically lend themselves to any post packaging pasteurization steps. This packaging format is considered to deliver convenience and have excellent product presentation (Belcher, 2006). Several large US companies have achieved national distribution of ready meals, which may contain gravy or sauce, packaged in vacuum films that can withstand a hot water cook of up to 12 h at 90°C. The packaged product is then placed in a microwaveable tray, lidded with a non-barrier film and then placed into a sleeve or box for distribution. This package can also be assembled with a pre-cooked entrée and then go through a post packaging pasteurization step to eliminate any vegetative pathogens. The consumer can then re-heat the product in the microwave or in hot water. The disadvantage is that the presentation of the entrée is not very appealing and is usually marketed in a box or a sleeve. The consumer also has to puncture the product prior to microwave heating and remove the hot product from the package prior to serving.

In March 2007, new marination packaging for value added meat products was unveiled in the US by Cryovac Food Solutions. This new two-part package will contain separate compartments for fresh meat and marinade and the unique design will allow the end user to combine the marinade with the meat by breaking the seal between the meat and marinade. This packaging design is being targeted to foodservice establishments, with benefits including reduced labour and improved quality of marinated meat items, including shelf life.

#### *Opportunities for value added frozen pork meals*

The market share of the frozen food category in major Australian supermarket chains has been reported to have increased by 30% in 2005. Frozen meals alone have grown by 5.1% (MAT 03/06) and frozen savouries (Synovate and Aztec, 2006). It is noteworthy that Nestlé's Lean Cuisine range does not currently include a pork meal in its Bowl Food range (1 fish, 1 vegetarian, 5 chicken, 2 beef and 1 lamb) or in its Larger Serves Range (1 fish, 4 vegetarian, 7 chicken, 2 beef and 1 lamb), so providing quality pork alternatives in this market segment may be an area worthy of exploration. Furthermore, while Lite'N'Easy have a very extensive range of pre-prepared meals (in excess of 50), only two meals are prepared from pork (pork roast and pork sausage) ([http://www.liteeasy.com.au/chefskitchen/dinner\\_menu.cfm](http://www.liteeasy.com.au/chefskitchen/dinner_menu.cfm)). Importantly, the challenges of creating great tasting products that are convenient must now be addressed by processors/manufacturers. Raw material selection, seasoning characteristics and plate presentation for the consumer are key issues that must be addressed as part of the process development, to ensure that quality products are produced.

#### *New packaging developments*

Eilert (2005) reported that a film structure that is able to be refrigerated and then reheated conventionally or by microwaving has been developed. This technology resulted from combining crystallised polyethylene terephthalate (CPEI) with amorphous polyethylene terephthalate (APET).

New and alternative packaging systems are required to further meet the needs for high quality, convenience foods. There is considerable interest and development in biomaterials (refer to Nettles Cutter, 2006) however, limited functionality with meat and high cost has affected its ability to effectively compete with plastics, derived from petroleum-based materials. With rising petroleum costs, there is concern with finding cost-effective ways to manufacture packaging materials. Furthermore, consumers are demanding that food packaging materials be more natural, disposable, potentially biodegradable as well as recyclable (Lopez-Rubio *et al.*, 2004).

Changes in consumer preferences have led to innovations and developments in new packaging technologies, especially film properties. Active packaging, used to extend the shelf life of fresh and cooked meat products, include oxygen scavengers, carbon dioxide scavengers and emitters, drip absorbent sheets and antimicrobial packaging. Antimicrobial packaging is gaining interest from researchers and industry due to its potential to provide safety and quality benefits.

### *Equipment innovations*

Meat processors are faced with four main operational challenges: variety/flexibility in production, packaging cost optimisation (per kg or per pack), ensuring food safety and environmental issues. Processors are also demanding highly automated plants with continuous processing lines providing complete control and traceability of the entire process, increases in productivity, reduction in labour costs, safe and ergonomic operation, with minimal maintenance and ease of cleaning and sanitation. The challenge for Australian pork processors is how to cost-efficiently incorporate new equipment and technology with current volumes – relatively smaller in scale in comparison to US and European processors.

As detailed in this review, changing and evolving consumer trends and demands are driving a continual need for new and more innovative food products. Changing consumer tastes and the need to satisfy the requirements of convenience and different portion sizes are also driving more research and new product development initiatives. This has resulted in closer co-operation between pork processors, equipment suppliers/manufacturers and ingredient companies working together to test for example, new recipes, yield targets, packaging requirements.

### **Conclusion**

Product development is difficult and risky. Most new products launched on consumer markets are failures. While the exact figures vary a great deal (and naturally depend on the way one defines success and failure), it is commonly accepted that the failure rate for new products on consumer food markets is somewhere between 60 and 80%. Having success with new products, also in the meat sector, requires constant input from the market, and, on consumer markets, especially from consumers.

The ultimate success of a food product is dependent on its acceptance to the consumer, who is the user, potential user and/or the product purchaser (Moskowitz 1985). Globally, sensory testing and market research studies are crucial to the success of new products and Australia is no different. It is envisaged that new pork product development in Australia will require significant market research to determine consumer needs and the new pork products that will meet these needs. General indications are however very positive for the Australian pork industry with significant areas of opportunity for new pork products already identified.

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## Dietary selenized yeast increases the selenium content whereas organic iron (sqm) has no effect on the iron content of pork

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The human health benefits of dietary selenium (Se) and iron (Fe) are well established (Rayman, 1997; Kristensen *et al.*, 2005). Meat and meat products are primary sources of dietary Se and iron for humans. Pork from Se and Fe-supplemented pigs may provide an additional source of these nutrients. However, the effects of supplementation of Se and iron on their status in muscles and effects on pork quality are not clearly defined. This study examined the effects of dietary Se and Fe supplementation on the Se and Fe status and meat quality of pork.

Crossbred finisher pigs (n=18 boars and 18 gilts), were offered *ad libitum* access to one of six experimental diets: 1) Basal: 0.13 mg/kg sodium selenite + 50 mg/kg iron (II) sulphate; 2) 3 mg/kg Diamond V Se (Diamond V Mills Inc); 3) 9 mg/kg Diamond V Se; 4) 100 mg/kg SQM Fe (Quali Tech®); 5) 1000 mg/kg SQM Fe and 6) 3 mg/kg Diamond V Se + 100 mg/kg SQM Fe, and were slaughtered after 28 days. *Longissimus dorsi* (LM) and *Biceps femoris* (BF) muscles were analyzed for Se and Fe levels. Pork quality measures were taken 24 hours post-slaughter in LM muscles. Instrumental colour ( $L^*$ ,  $a^*$  and  $b^*$ ) and Warner-Bratzler shear force were measured up to five days of aging. Data were pooled across sexes and analyzed using analysis of variance.

Dietary Se supplementation significantly ( $P < 0.0001$ ) increased the Se concentration of pork in a linear manner (Table 1). Conversely, the Fe supplements had no effect on the Fe content in both LM ( $P = 0.88$ ) and BF ( $P = 0.14$ ). The Fe content of LM and BF muscles were higher ( $P < 0.0004$  and  $P < 0.056$  respectively) in gilts than boars. Neither Se nor Fe supplements had an effect on pig performance, carcass traits or meat quality parameters ( $P > 0.05$ ). The results indicate the potential for healthful fortification of pork with organic Se, whereas no beneficial effects of feeding higher levels of organic Fe were identified. Higher Fe levels observed in gilts over the boars warrant further investigation.

**Table 1. Effect of the dietary Se and Fe supplements on Se and Fe status in LM and BF muscles, live pig performance, carcass composition and pork quality**

Attribute	Se ppm			Fe ppm		Se/Fe ppm	SE	P-value
	Basal	3	9	100	1000	3/100		
ADG, kg	1.03	1.04	1.01	1.07	0.97	1.05	0.06	0.84
LM- Fe, mg/kg	12.3	12.5	12.3	12.2	11.7	13.2	0.71	0.79
BF- Fe, mg/kg	11.7	14.8	9.3	10.0	10.8	12.3	0.72	0.15
LM- Se, µg/kg	133.3	903.3	2433.3	125.0	131.7	980.0	57.5	<0.0001
BF- Se µg/kg	138.3	935.0	2533.3	138.3	138.3	976.7	54.3	<0.0001
Ultimate pH (24-h)	5.6	5.5	5.4	5.6	5.5	5.5	0.05	0.84
Drip loss, % (48-h)	4.3	6.5	6.8	6.3	6.0	6.7	0.72	0.19
Shear Force, kg (5-d)	3.2	3.5	3.0	3.4	3.5	3.0	0.24	0.37
$L^*$ (5-d)	48.3	48.2	49.4	48.3	48.8	49.7	0.88	0.76
$a^*$ (5-d)	6.3	6.2	5.7	6.1	5.6	6.0	0.31	0.58
$b^*$ (5-d)	5.4	5.4	5.5	5.8	5.4	5.9	0.29	0.69

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## Expression of colonic selenoproteins from selenium-enriched milk assessed in artificially-reared neonatal pigs

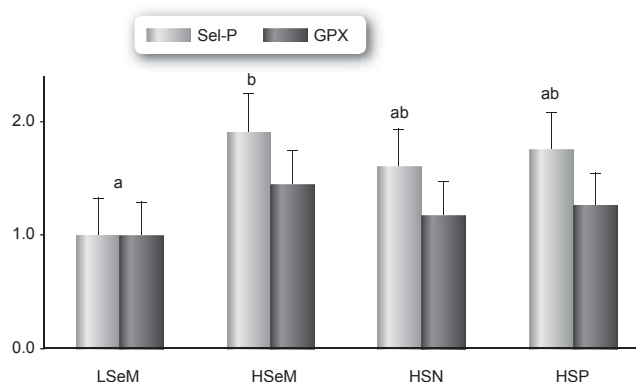
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While it is generally accepted that selenium (Se) intakes of Australian and New Zealand consumers are sufficient to prevent overt signs of deficiency, the relatively low intakes of Se in these countries may contribute to an elevated risk of bowel cancer (Clark *et al.*, 1996). As protein-bound Se is more bioactive and less toxic than other forms of Se, there is interest in increasing the amount of Se in animal proteins for human consumption. Since milk protein-bound Se is more bioavailable than inorganic Se and yeast-bound Se (Uglietta *et al.*, 2006), the aim of this study was to determine the effect of different forms of Se on the expression of selenoproteins gastrointestinal glutathione peroxidase (GI-GPX) and selenoprotein P (SelP) which may be involved in protection against colon cancer.

Milk enriched with Se (1070 µg Se/kg DM; HSeM) was obtained from cows fed a diet containing Sel-Plex<sup>®</sup> (Alltech Biotechnology Pty Ltd) while the control diet (135 µg Se/kg DM; LSeM) was made using commercial milk powder. Additional diets were formulated by adding Sel-Plex<sup>®</sup> (HSP) or selenate (HSN) to the LSeM diet to give Se concentrations of 1070 µg Se/kg DM. Neonatal pigs (n=54) at two days of age were trained to drink cow's milk and after a further three days were randomly allocated to their respective diets (1.7 MJ/kg BW) and slaughter times (0, 7, 14, 28 and 42 days of feeding). Frozen colon samples were homogenized and RNA extracted and transcribed to cDNA before real time polymerase chain reaction. Primers for GI-GPX and SelP were designed using the pig (*Sus scrofa*) genome database.

The gene expression of colonic SelP was significantly greater (P=0.024) in the pigs fed HSeM than in those fed LSeM while the expression in pigs fed the HSP and HSN was intermediate (Figure 1). There was no significant effect of dietary Se on expression of GI-GPX (P=0.33). Muscle Se contents were 47, 106, 237 and 486 µg/kg for LSeM, HSN, HSP and HSeM pigs, respectively (P<0.001) (Uglietta *et al.*, 2006). These data suggest that Se in milk from cows fed selenized yeast is highly bioavailable and bioactive and may be a means of increasing Se intake and reducing colonic cancer risk in humans.



**Figure 1.** Gene expression of important colonic selenoproteins in pigs fed various forms of Se

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## Does gestation housing affect the lying behaviour of sows after farrowing and hence piglet survival to weaning?

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During parturition and the first 72 hours of lactation the most important non-infectious cause of piglet death is overlaying. Karlen *et al.* (2007) found sows had more feet and leg problems late in gestation in stalls than in group housing and as this condition is likely to continue during early lactation, it may contribute to the sows' apparent unwillingness to change posture during lactation. In this study, we tested the hypothesis that stall housing during gestation increases the risk of piglets being overlaid after farrowing.

During gestation, sows were kept in either conventional stalls (0.6x2.1 m) (Stall) or in large groups of 85 sows on deep litter (Group). Each week for eight weeks, 13 sows from each of the two gestation treatments were selected on entry to the farrowing house. From the 208 sows selected, 203 farrowed and weaned (99 from the Stall treatment and 104 from the Group treatment). Once each week on days 1-4 after farrowing, sows were videotaped from 0700 to 1100 h as they changed posture from standing to lying on their belly/udders in farrowing crates. Recorded data included: the number of possible 'risk behaviours' (e.g. slipping, rapid/uncontrolled descent of all or part of the body); latency to lie down; the time sows took to change from a standing to a lying position; the number of unsuccessful attempts relative to a successful attempt to lie down; and piglet survival. Data were analyzed by analysis of variance (Genstat v8) using replicate means. Data on latency to lie down were log transformed before analysis.

Sows from the Stall treatment took longer and had more unsuccessful attempts to lie down than sows from the Group treatment (Table 1). One interpretation of these results is that sows from the Stall treatment were more uncomfortable than sows in groups due to feet and leg problems carried over from gestation. However, housing during gestation did not affect pre-weaning piglet survival. These changes in posture behaviour may be an indicator of poor welfare and may have long term effects on the productive life of breeding sows. Lameness is an important cause of early culling and need to be further investigated.

**Table 1. Effects of gestation housing on the lying behaviour of the sow**

Risk and behaviours	Stall	Group	SED
Risk posture change	1.8	1.7	0.41
Latency to lie down (log transformed)	1.2 <sup>a</sup>	1.1b	0.03
Number of attempts per successful attempt to lie down	1.5 <sup>a</sup>	1.2b	0.11
Survival of born alive at weaning (%)	87.8	89.1	0.12

<sup>a,b</sup> Different superscripts represent significant differences between means (P<0.01).

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## Piglet mortality in farrowing pens and farrowing crates

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Piglet mortality is generally higher in farrowing pens than crates (Edwards and Fraser, 1997), which affects the economics and acceptability of pens as a practical alternative for crates. However, Weber (1997) and Cronin *et al.* (2000) reported no difference in piglet mortality between pens and crates and suggested that improved pen design, appropriate sow genetics and training and selection of stockpeople needed to be addressed before pens were likely to replace crates commercially. The objective of this experiment was to compare piglet mortality in an 'improved design' of the Werribee farrowing pen (WFP) and farrowing crates.

The experiment was done with 72 Large White x Landrace sows on a commercial farm over 18 months and with 12 replicates in time. Two WFPs and four farrowing crates were located within the same shed. WFPs measured 2.0 m wide x 3.7 m deep and were 'improved' by incorporating a triangular (rather than rectangular) piglet creep in one corner of the 'nest' area of the pen. The 'nest' area measured 2.0 x 1.9 m, had a solid concrete floor and piglet protection panels inside the 'nest' area, including in front of the creep area but not across the front of the 'nest' area (i.e. the 'non-nest' area). The 'non-nest area' had a mesh floor measuring 2.0 x 1.8 m and contained the sow feeder and drinker. The farrowing crate had a fully-slatted floor, except for the piglet creep area which was solid. While the creep areas in the two accommodation types had identical heaters, 10 L of rice hull bedding was added to the creep floor in the WFP before introducing the sows, which occurred at least five days before the anticipated farrowing date. Sows were fed 2.5 kg of lactation sow diet plus 0.5 kg bran per day until farrowing and thereafter were fed *ad libitum* with lactation sow diet without bran. Sows and litters were husbanded according to the standard practices of the collaborating piggery and weaning occurred on day 21 of lactation. For each replicate and measurement a pen mean was calculated from the two WFPs. Similarly, a crate mean was calculated over the four crates. The WFPs and crates were then compared using a paired t-test statistic with the calculated means as the experimental units, and with the replicates as the pairs. Exact P values were calculated using a permutation test (Genstat v9.1).

There were no differences ( $P > 0.05$ ) in production parameters for litters in WFPs and crates (Table 1) supporting the findings of Weber (1997) and Cronin *et al.* (2000). Based on piglet mortality, the results suggest the WFP with a triangular creep design is a practical alternative to farrowing crates.

**Table 1. Piglet production and loss per litter in farrowing pens and crates**

Measurement	Pen	Crate	SED	P-v
Parity of sows	2.0	1.8	0.23	0.480
Total born	10.4	11.3	0.63	0.223
Born alive	9.7	10.7	0.56	0.127
Pre-weaning mortality	1.2	1.4	0.24	0.469
Fostered (net)	+0.3	-0.3	0.35	0.169
Weaned	8.8	9.0	0.37	0.612

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# Season and parity effects on the feed intake of lactating sows in an Australian commercial piggery

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Insufficient nutrient consumption during a sow's lactation period will cause a loss of body condition. This can adversely affect the subsequent reproductive performance of the sow and can lead to an increase in sow cull rates through extended weaning to service intervals or failure to enter oestrus (Eissen *et al.*, 1999). Gilts, due to lower body reserves, are particularly prone to weight loss during lactation. Numerous factors influence the feed consumption of lactating sows including parity number and seasonal effects. The aim of this paper was to investigate the effects of season and parity and their interaction on the feed consumption of sows in a south eastern Queensland (Kalbar) commercial piggery.

Records for 2234 litters from 774 sows of three breeds (75.6% Large White, 17.4% Landrace, 7% Crossbred) were analyzed for lactation feed intake. Records were from May 2002 through to November 2006. Sows had a mean lactation length of 21 days and a mean feed intake of 5.11 kg/day. Sows were fed twice daily from Monday to Saturday and once on Sundays. The farm used a step up system that increased feed quantity incrementally from the beginning of lactation. Lactation feed intake was analyzed using Proc GLM (SAS Inc. v.8.2). Significant fixed effects modeled ( $P < 0.001$ ) were breed, parity, farrowing month and year with lactation length included as a linear covariate.

Primiparous sows averaged significantly less daily feed intake than higher parity sows (Figure 1). Feed intake increased steadily from January to July and then fell again for primiparous and multiparous sows (Figure 2). Primiparous sows lactating in January ate 2.19 kg/day less than multiparous sows lactating in July (Figure 2) and averaged below 4 kg/day from November to February. The parity by season interaction was not significant. In comparison, primiparous and multiparous Large White sows of European origin fed once daily over a 28-day lactation period consumed 4.0 and 4.4 kg/day respectively (Gourdine *et al.* 2004) across seasons in a tropical environment. Feed intake was 4.7 kg/day during winter (Nov-Apr) and 3.9 kg/day during summer months. The low feed intake levels of summer farrowing primiparous sows may be compromising the future reproductive performance of these animals due to the adverse effects of mobilizing body reserves.

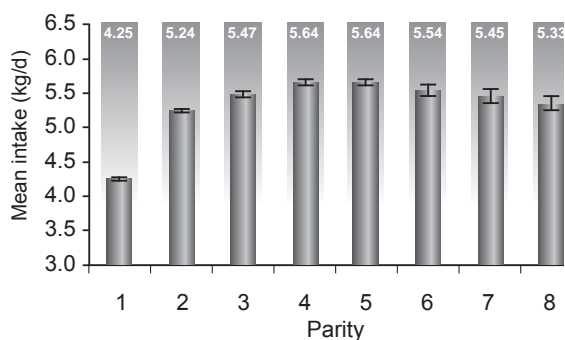


Figure 1. Mean daily feed intake by parity

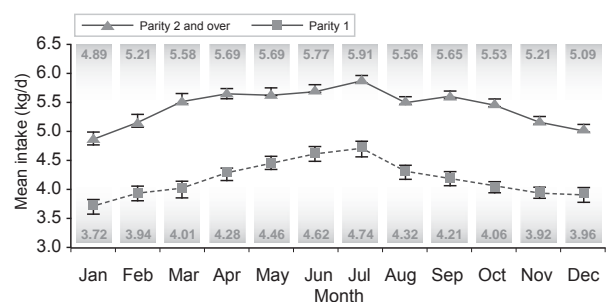


Figure 2. Monthly mean daily feed intake

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## The effect of cooled drinking water on the lactation performance of gilts and parity 1 sows

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The recommended temperature for optimum pig water intake is 15-20°C (Kruger *et al.*, 1992). During summer the temperature of drinking water is often similar to the ambient temperature (Willis and Collman, 2005). Poor water intake during lactation may have direct effects on milk synthesis and ultimate weaner performance (Jones and Stahly, 1999). This experiment investigated the relationships between cooled drinking water on daily water intake, incidence of agalactia, lactation feed intake and weaner growth performance.

Two hundred and eight Large White x Landrace gilts and parity 1 sows were housed in farrowing crates from 112 days of gestation. The sows were provided with either the standard shed drinking water, or cooled water (cooled to 15°C via a refrigerated unit) from the point of entry to the farrowing house. The sows were fed *ad libitum* with a mash diet once they had farrowed. Individual sow water usage was recorded with a water meter. The litters of piglets were weighed post fostering (24 hours post birth) and prior to weaning at an average of 12 days of age. The ambient temperature and the temperature of water in the drinker lines were recorded. The sow and litter were used as the experimental unit and the data were analyzed by GLM analysis of variance and chi square analysis.

The temperature of the control drinking water treatment was similar to the ambient temperature (average 33°C) and the cooling system cooled the drinking water to a constant 15°C. Providing cooled drinking water significantly ( $P < 0.05$ ) increased the daily water intake of the sow (Table 1). There was no significant ( $P > 0.05$ ) impact of treatment on daily lactation feed intake, incidence of agalactia (first seven days after farrowing) and weaner growth performance. It is speculated that sows and piglets may require more time (i.e. greater weaning age) to respond to the cooled water treatment.

**Table 1. Average reproductive performance, daily lactation water and feed intake, and growth performance of piglets in control and cooled drinking water treatments**

	Control	Cooled	SEM
Daily sow water intake (L/day)	22.0 <sup>a</sup>	23.9 <sup>b</sup>	0.52
Daily sow feed intake (kg/day)	4.6	4.7	0.08
Number of pigs weaned/sow	8.8	8.4	0.17
Rate of gain of piglets	0.181	0.182	0.004
Rate of gain of piglets <sup>1</sup>	0.179	0.184	-

a, b Within rows, means with different superscripts are significantly different ( $P < 0.05$ )

<sup>1</sup>Weaning number used as covariate

The financial support from Australian Pork Limited and the technical assistance from the management and staff at QAF Meat Industries Pty Ltd is gratefully acknowledged.

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## Teat order may affect within-litter weight variation but supplemental milk does not

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Fraser and Thompson (1986) reported that piglets feeding from anterior teats grow faster than those feeding from posterior teats in sow-reared but not gilt-reared litters. While it has been shown that providing piglets with supplemental milk increases average gain in litter weight (Azain *et al.*, 1996), it is not known if this milk is consumed equally by piglets on all teats or whether it impacts on litter weight variation. Our first hypothesis was that piglets on anterior teats grow faster than those on posterior teats, independent of parity. Our second was that supplemental milk impacts equally on the weight gain of piglets, independent of teat order.

Seventy-two gilt litters and 72 parity 2-5 sow litters, housed in five commercial farrowing sheds, were randomly assigned to two treatments ( $\pm$  supplemental milk). Full cream milk was provided *ad libitum* 2-3 times daily to piglets from 3-26 days of age (weaning). Piglets were fostered within 36 hours of birth to standardize litter sizes to 10 piglets. Individual piglets were identified at birth and teat positions were recorded three times in the first two weeks of lactation. An average teat position was used if the teat position varied. Piglets were weighed individually at birth, 21 days of age, weaning and 10 weeks of age. The experiment was done in winter and repeated in summer.

Log individual piglet weights were analyzed with REML in Genstat. The average teat position, birth weight, dam parity, milk ( $\pm$ ), season, piglet gender and fostered ( $\pm$ ) were fixed effects. Udder section (US) was divided into teat pairs; US1 (anterior two pairs), US2 (middle two pairs) and US3 (posterior pairs). Fixed effects were adjusted for other model terms (ie. birth weight). Log litter standard deviation (SD) was analyzed using REML in Genstat. Dam parity, milk ( $\pm$ ), log litter birth weight SD, stage (21 days/weaning) and season were fixed effects.

Piglets on anterior US grew faster than those on posterior US for both parity groups (US 1>2>3; all  $P<0.01$ ) but this effect was not significant post-weaning (Table 1). There was an interaction of birth weight and US on subsequent piglet weight ( $P<0.001$ ). Piglets grew faster on US2 than US3 across all birth weights but US1 growth benefits were more obvious for lighter compared to heavier birth-weight piglets. Milk-supplemented piglets were significantly heavier than non-supplemented piglets at 21 days (6.5 kg and 6.1 kg respectively;  $P<0.01$ ) and weaning (7.8 kg and 7.2 kg respectively;  $P<0.001$ ) irrespective of udder section ( $P=0.89$ ). Litter standard deviation was not influenced by supplemental milk ( $P=0.829$ ). Season did not alter the effect of milk supplementation across udder positions ( $P=0.66$ ) or litter standard deviation ( $P=0.13$ ).

**Table 1. Model-based means for piglet weights (kg) for each udder section (US)**

	21 days	Wean	10 weeks
US 1 (anterior)	6.55 <sup>a</sup>	7.70 <sup>a</sup>	24.31 <sup>a</sup>
US 2 (middle)	6.38 <sup>b</sup>	7.54 <sup>b</sup>	24.14 <sup>a</sup>
US 3 (posterior)	5.99 <sup>c</sup>	7.13 <sup>c</sup>	24.22 <sup>a</sup>

<sup>abc</sup>Different superscripts within column indicate significance ( $P<0.01$ ). Values adjusted for 1.6 kg birth weight.

Our results suggest that piglets feeding from anterior teats grow faster than those on posterior teats independent of dam parity in the pre-weaning period, but that this effect declines over time. Placing low birth weight piglets onto the anterior teats of the dam may reduce within-litter weight variation. Supplemental milk appears to provide a growth advantage to piglets equally across all udder sections and was not found to change within-litter weight variation.

This project was supported by Australian Pork Limited and QAF Meat Industries Pty Ltd.

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## Identifying the relationship of gilt rearing characteristics to lifetime sow productivity

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Sow longevity is an important contributor to the overall profitability of a swine operation. Several researchers have studied the influence of gilt development programs on age at puberty, litter size at first farrowing and productivity throughout early adult life. However, few researchers have studied characteristics of gilt development as they might influence lifetime sow productivity and longevity. We hypothesized that low body weight (BW) and backfat depth (BF) of gilts at selection would compromise lifetime productivity of females selected to enter the breeding herd.

A retrospective analysis of gilts (n=11,571) entering the nucleus herd of an Australian commercial piggery from 1999 through to 2003 was done to determine the effects of BW and BF on lifetime sow productivity. Gilts represented five hybrid genetic lines and were selected at 22 weeks of age to enter the gilt pool at 29 weeks of age. At selection, BW and BF at the P2 location were recorded for each gilt. After entry to the gilt pool, total productive days, live pigs farrowed and pigs weaned were recorded over each female's life in the herd. A productive day was any day that a female was pregnant, presumed pregnant or lactating. Gilts that entered the gilt pool at 29 weeks of age but never mated, or failed to produce a litter remained in the dataset for analysis (Productive days = 0 or <114, respectively; litter size born or weaned = 0). Records were ranked lowest to highest by BW or BF then divided into six equally sized groups. Data were analysed using analysis of variance with productive days, live pigs farrowed, and pigs weaned as dependent variables and BW or BF group and genetic line as the independent variables.

Both BW and BF at 22 weeks of age (Table 1) influenced ( $P<0.05$ ) lifetime productive days, live pigs farrowed and pigs weaned. Genetic line explained a meaningful ( $P<0.01$ ) portion of the variation in all analyses but did not interact with BW or BF. Increases in BW at 22 weeks had no effect on lifetime performance until gilts reached a BW more than 101 kg. Body weight more than 101 kg compromised lifetime performance. Backfat had no effect on lifetime performance unless females were very lean (<9 mm) with a reduction in all three measures of lifetime performance. Stalder *et al.* (2005) reported similar results for US Landrace sows. We conclude that maximum BW and minimum BF threshold values for gilts at selection within specific genetic lines may be required to optimize lifetime productivity of breeding sows.

**Table 1. Relationship of body weight (BW) and backfat (BF) depth at 22 weeks of age to lifetime sow productivity**

BW category (kg)	n	Mean BW or BF	Productive days	Live pigs	Weaned pigs
67 – 89	1958	85.3	285 <sup>ab</sup>	17.9 <sup>ab</sup>	15.9 <sup>a</sup>
89 – 93	1959	91.1	298 <sup>a</sup>	18.7 <sup>a</sup>	16.5 <sup>a</sup>
93 – 97	1958	95.3	288 <sup>a</sup>	17.8 <sup>ab</sup>	15.8 <sup>a</sup>
97 – 101	1959	99.3	301 <sup>a</sup>	18.6 <sup>a</sup>	16.4 <sup>a</sup>
101 – 107	1958	103.8	284 <sup>ab</sup>	17.5 <sup>ab</sup>	15.4 <sup>ab</sup>
107 - 138	1959	112.3	269 <sup>b</sup>	16.6 <sup>b</sup>	14.4 <sup>b</sup>
<b>BF category (mm)</b>					
6 – 9	1958	8.1	260 <sup>a</sup>	15.8 <sup>a</sup>	14.1 <sup>a</sup>
9 – 10	1959	9.4	277 <sup>ac</sup>	17.1 <sup>ac</sup>	15.2 <sup>ac</sup>
10 – 11	1958	10.1	286 <sup>bc</sup>	17.8 <sup>bc</sup>	15.5 <sup>bc</sup>
11	1959	11.0	302 <sup>b</sup>	19.0 <sup>b</sup>	16.7 <sup>b</sup>
11 – 13	1958	12.1	295 <sup>bc</sup>	18.4 <sup>bc</sup>	16.4 <sup>bc</sup>
13 – 24	1959	14.2	304 <sup>b</sup>	19.1 <sup>b</sup>	16.7 <sup>b</sup>

<sup>abc</sup> Means within column and category with different superscripts differ ( $P<0.05$ ).

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## 2

## Poster session

## Digestible energy content of sunflower meal

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There are no recent Australian data on the nutritive value of sunflower meals currently available for use in pig diets, despite sunflower seed production having shifted significantly over the past decade from seed for polyunsaturated oils to monounsaturated oil production. Sunflower breeding programs and commercial processing of sunflower into either expeller or solvent meals may also have influenced the nutritive value of the meals. In diet formulations using Feedmania the following figures for digestible energy (DE) content are frequently used: 8.3 MJ/kg for solvent extracted sunflower meal and 10.1 MJ/kg for expeller sunflower meals. The original source of this information is unknown. Dinusson (1990) reported that partially de-hulled sunflower meal and sunflower meal with hull have DE values of 10.1 MJ/kg and 9.8 MJ/kg respectively. The current study examined whether these values were still accurate.

We used a pig metabolism study to assess the digestible energy (DE) value of solvent-extracted monounsaturated and polyunsaturated sunflower meals and a mono-unsaturated expeller sunflower meal. Sixteen entire male Large White pigs (~22 kg) were housed individually in metabolism crates. Pigs were given eight days to adapt to the treatment diets after which total faecal collections were carried out over five days. Ferric oxide was added to the diet as a faecal dye to indicate the start and end of collection. There were four replicates for each experimental diet. Diets were based on 60% sorghum, 30% sunflower meal and a 10% basal component consisting of casein, vitamins, minerals, lysine and oil.

The higher residual fat present in the expelled sunflower meal increased the faecal digestible energy content of this sunflower meal over the solvent extracted sunflower meals (Table 1). The energy digestibility coefficient of the sunflower meals (0.45-0.54) was less than sorghum (0.78).

**Table 1. Nutritive and digestible energy content of various sunflower meals and sorghum**

Sunflower Type	Ingredient content (DM basis)					(As fed basis)	
	DM %	Protein %	Fibre %	Fat %	GE MJ/kg	DE MJ/kg	
Mono-unsaturated expeller meal	93.5	32.6	21.7	14.9	22.5	12.1	
Mono-unsaturated solvent meal	90.0	36.3	22.9	1.9	20.0	9.0	
Poly-unsaturated solvent meal	90.1	37.0	24.2	1.8	19.9	9.6	
Sorghum	89.3	10.4	2.4	3.6	18.7	14.7	
LSD (P=0.05)	-	-	-	-	-	1.45	

These results indicate that the faecal digestible energy content of current sunflower meals produced in Australia today are higher than previously accepted values. This could be a result of changes in breeding, production or processing of sunflower seeds.

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# Towards predicting *in vivo* digestion in pigs: is there an *in vitro* quotient?

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Energy delivery to pigs is currently topical. The various options (additives, grain treatments, types and varieties of grain, and processing) for high digestible energy are usually evaluated by *in vivo* procedures. Although valuable, *in vivo* studies are expensive in time and money and do not separate enzyme digestion from microbial fermentation. *In vitro* studies have some advantages as many samples can be analyzed to elucidate the mechanisms available to engineer pig feeds. *In vitro* techniques have successfully predicted *in vivo* digestion of some foods (Goñi *et al.*, 1997). This paper reports the analysis of *in vitro* enzyme digestion of some cereals to obtain characterization parameters that may predict digestion in pigs.

Ground grains were treated with artificial saliva, pepsin at a low pH and  $\alpha$ -amylase and amyloglucosidase in a reciprocating water bath at 37°C. Time-courses for enzyme digestion of starch by measuring glucose generated (digestogram) in duplicates were analyzed with adaptations of the Michaelis-Menten (Eqn. [1-2]) and empirical (Eqn. [3]) models (Goñi *et al.*, 1997; Duggleby, 2001; Amato *et al.*, 2004):

$$\text{Eqn. [1]- } D_t = D_o + \frac{V_{Mo}}{\alpha} (1 - \exp[-\alpha t]) - \left( \frac{V_{Mo} K_M}{V_{Go}} - M_o \right) [1 - \exp\{-V_{Go}/\alpha K_M (1 - \exp(-\alpha t))\}]$$

$$\text{Eqn. [2]- } Dt/t = V_{max} + K_M \ln(1 - [Dt/Do])/t$$

$$\text{Eqn. [3]- } Dt = D_o + D_\infty (1 - \exp[-K t])$$

where  $D_t$  = digested starch at time =  $t$ ,  $D_o$  = digested starch at time = 0 and  $D_\infty$  = digested starch at time =  $\infty$ ,  $K$  = rate constant,  $K_M$ ,  $\alpha$  = Michaelis-Menten constant or its function,  $V_{max}$ ,  $V_{Mo}$ ,  $V_{Go}$  = maximum velocity of starch hydrolysis or its function,  $M_o$  = initial maltose.

Some parameters in Table 1 are potentially useful in understanding enzyme-substrate relationships and therefore how various factors influence enzyme digestion of starch in feeds. The developed *in vitro* procedure can be adapted to screen 10-12 feeds in five hours.

**Table 1. Selected parameters of the models for starch hydrolysis**

Grain	Eqn. [1]				Eqn. [2]			Eqn. [3]			
	$D_o$	$V_{Mo}$	$\alpha$	$r^2$	$V_{max}$	$K_M$	$r^2$	$D_o$	$D_\infty$	$K$	$r^2$
Barley	0.3	2.42	0.04	0.999	-0.43	-93.39	1.000	8.3	48.2	0.11	0.926
Sorghum	1.8	4.12	0.05	0.999	-0.66	-98.76	0.993	5.5	75.3	0.07	0.989
Triticale	0.4	2.72	0.05	0.996	-0.81	-89.58	0.999	11.4	52.2	0.17	0.891
Wheat	0.5	2.11	0.04	0.998	-0.43	-92.46	1.000	9.1	44.0	0.13	0.903

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# Minimum inclusion levels of copper and zinc proteinate maintain performance and reduce faecal excretion in growing and finishing pigs

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Hernandez *et al.* (2005) found that total dietary levels of 25 ppm copper (Cu) and 40 ppm zinc (Zn), in the sulphate or proteinate form (Bioplex®), reduced the concentration of Cu and Zn in faeces by 80 and 60% respectively and maintained the growth of growing pigs, compared to a diet containing 160 ppm of both Cu and Zn. These results indicated reduced levels of supplemental Cu and Zn might be possible. In the present study we therefore measured the impact of increasing inclusion levels (IL) of Cu together with low (treatments 1-4) or high (treatments 5-8) IL of Zn in the Bioplex® form on performance and faecal levels.

The experiment was designed as a 2x4 factorial arrangement of treatments, with the respective factors being two IL of Bioplex® Zn (40 and 80 ppm) and four of Bioplex® Cu (0, 10, 30 and 50 ppm). A control treatment was included (treatment 9), which used sulphates at levels of Cu and Zn similar to the high Bioplex® treatment. The study used 216 Large White x Landrace pigs from 25-107 kg LW housed in three pens of eight pigs/treatment. Pigs were fed *ad libitum*. Individual LW was measured weekly and faecal samples were collected from the same random sub-sample of four pigs/pen on days 21 and 49 of the experiment. Analysis of variance, using the pen as a unit for average daily gain (ADG), voluntary feed intake (VFI) and feed conversion ratio (FCR) and the pig for faecal levels, was used to examine main effects and all interactions on ADG, VFI, FCR as well as Cu and Zn faecal levels. Growth performance was analyzed over the entire experimental period, and faecal levels were analyzed using repeated measures analysis of variance.

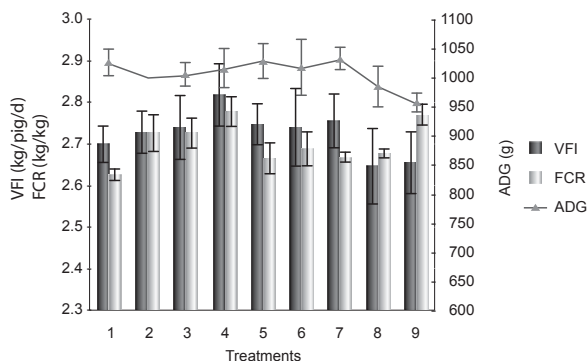


Figure 1. Pig performance ( $\pm$ SEM)

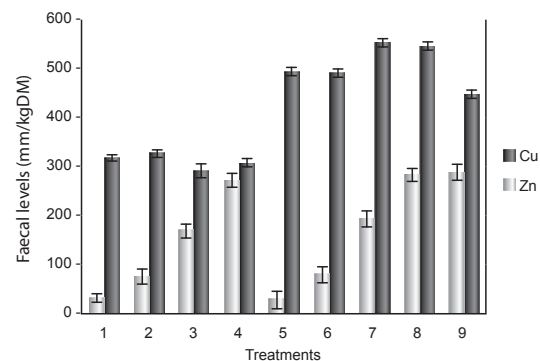


Figure 2. Faecal mineral levels ( $\pm$ SEM)

Over the entire experiment, there was a trend for better ADG in pigs on treatments 1, 5 and 7 (0 ppm Cu and 40 ppm Zn; 0 ppm Cu and 80 ppm Zn; and 30 ppm Cu and 80 ppm Zn, respectively) than that for the pigs fed inorganic Cu and Zn ( $P < 0.1$ ). There was also an advantage to FCR when diets contained Bioplex® Cu and Bioplex® Zn, especially when Zn was included at 80 ppm ( $P < 0.1$ ) (Figure 1). A significant reduction in the levels of Cu (90%) and Zn (40%) in faeces was achieved when the IL in the diet decreased from 50 to 0 ppm Cu, and from 80 to 40 ppm Zn, both in the Bioplex® form (Figure 2). These results indicate that IL of 0 ppm Cu and 40 ppm Zn in the Bioplex® form significantly reduced the amount of Cu and Zn excreted without a detrimental effect to pig performance. However in commercial practice pigs are challenged and marginal levels could compromise performance. Further experimentation to establish minimum levels under commercial conditions is warranted.

The financial support of Alltech Biotechnology Pty Ltd is acknowledged.

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## Mandelup lupins (*Lupinus angustifolius* L.) and enzyme supplementation do not affect carcass composition and meat quality of pigs

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Feeding albus lupin seed to pigs is known to decrease dressing percentage due to gut fill and intestinal cell proliferation. In addition, feeding lupins to pigs has been shown to decrease backfat thickness, without influencing carcass leanness and lean meat percentage in the ham. Enzyme supplementation can alter protein digestibility and this may have an effect on carcass composition. The impacts of including high levels of recently released cultivars of Australian sweet lupins (ASL) (*Lupinus angustifolius*) and the interaction of lupins with supplemental enzymes on carcass composition and indices of meat quality have not been examined. The purposes of this experiment were to examine 1) whether increasing use of the current variety of ASL seed (cv. Mandelup) and addition of multi-enzyme influences carcass characteristics of pigs and 2) whether high inclusion levels of ASL influences meat quality traits.

Two hundred and twenty-four (Large White x Landrace, 27.2±0.22 kg) male pigs were allocated to a 4x2 factorial design with the respective factors being lupin inclusion level (200, 250, 300 and 350 g/kg) and multi-enzyme supplementation (± added enzyme; Allzyme® SSF, Alltech Biotechnology Pty Ltd). All diets were formulated to contain equal amounts of ileal digestible amino acids and the same ileal digestible lysine to digestible energy ratio by progressively substituting lupins for soybean meal. Pigs were fed grower, finisher and pre-sale diets to 50 kg, 75 kg and 107 kg, respectively and then slaughtered at 107 kg. At 24 hours post-slaughter, longissimus thoracis muscle from the left side of each carcass were collected between the 10th and 15th ribs and indices of meat quality were measured in pigs at the two extremes (i.e. 200 and 350 g/kg). The GLM procedure of SPSS (SPSS Inc) was used (using pig as the experimental unit) for statistical evaluation.

Carcass composition and P2 fat depth were not significantly influenced by lupin concentration or enzyme supplementation (Table 1). Enzyme supplementation did not alter the growth of pigs or carcass characteristics. Including either 350 g/kg or 200 g/kg lupin seeds in the diets did not alter meat quality (Table 2). Under the conditions of this experiment, feeding up to 350 g/kg cv. Mandelup showed no negative effects on carcass composition or meat quality.

**Table 1. Effects of lupin concentration and enzyme addition on carcass compositions**

	Lupin (g/kg)				Enzyme		SEM	Main effect		
	200	250	300	350	-	+		Lupin	Enzyme	LxE
HCW <sup>1</sup>	69.9	70.0	69.7	69.5	69.7	69.9	0.18	0.824	0.710	0.793
Dressing %	65.0	64.9	64.8	64.5	64.8	64.8	0.18	0.771	0.876	0.984
P2 Backfat	13.6	13.5	14.0	14.0	13.7	13.9	0.23	0.821	0.719	0.532

<sup>1</sup>Hot carcass weight: AUSMEAT trim 13- head off, flare off, fore trotters off, hind trotters on.

**Table 2. Effects of lupin concentration on meat quality**

Lupin inclusion rate	200 g/kg	350 g/kg	SEM	Significance
Relative lightness (L*)	49.2	50.1	0.53	0.403
Relative redness (a*)	6.84	6.46	0.214	0.377
Relative yellowness (b*)	3.41	3.44	0.157	0.942
Ultimate pH	5.23	5.23	0.017	0.862
48-hour drip loss (%)	3.40	3.13	0.186	0.486
Cook loss (%)	36.5	35.9	0.32	0.348
WB shear-force (kg)	6.91	5.96	0.290	0.102

This work was funded by the WA Agricultural Produce Commission: Pork Producers' Committee.

## Dietary supplementation with fish oil increases long-chain omega-3 polyunsaturated fatty acids in pubertal gilts

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Polyunsaturated fatty acids are essential nutrients for pigs as they lack the necessary enzymes for bio-synthesis from palmitic acid via elongation and desaturation reactions (Enser, 1984). Polyunsaturated fatty acids are incorporated into cell membranes and are precursors to prostaglandins and leukotrienes. These metabolites are involved in reproduction and inflammatory responses. Fish oil contains a high level of omega-3 long-chain polyunsaturated fatty acids including eicosapentaenoic acid (EPA; C20:5) and docosahexaenoic acid (DHA; C22:6). In this experiment we measured the polyunsaturated fatty acid profile in plasma of breeding gilts offered increasing levels of fish oil supplementation to the diet.

Thirty-six Large White x Landrace F1 generation gilts were housed in individual pens (4 m<sup>2</sup>) and offered diets from 20 weeks of age *ad libitum*. After three weeks on a basal diet (13.9 MJ DE; 150 g crude protein; 8.2 g lysine; 56 g total fat per kg), gilts of similar live weight and backfat P2 (100.8±1.7 kg; 13.1±0.4 mm; mean±SE) were assigned to one of six diets. The diets differed in inclusion levels of tallow and fish oil (Feedworks Pty Ltd) from 30 g tallow/kg and zero fish oil to 21 g tallow/kg and 9 g fish oil/kg. Dietary linoleic acid (LIN) and arachidonic (ARA) did not differ between the diets (30 g/100 g; 0.2 g/100 g fatty acids). Gilts were bled at 23 and 30 weeks of age and their plasma analyzed for fatty acid profile as described by Parks and Goins (1994).

**Table 1. Mean±SE plasma fatty acid profile (g/100 g total fatty acids) from gilts at 30 weeks of age for samples taken after seven weeks**

Fatty acid	Fish oil supplementation to diet (g/kg)						P-value
	0	1.0	3.0	5.0	7.0	9.0	
C18:2 LIN	27.5±1.8	28.4±1.4	26.9±1.0	28.0±0.7	29.0±0.7	26.1±2.0	NS
C20:4 ARA	8.0±0.5	8.2±0.7	8.0±0.8	6.5±0.5	6.2±0.3	5.7±0.4	P<0.010
C20:5 EPA	0.30±0.03	0.42±0.07	0.67±0.06	1.0±0.1	1.5±0.1	1.9±0.2	P<0.001
C22:6 DHA	0.65±0.05	0.77±0.11	1.3±0.11	1.3±0.3	1.5±0.2	1.9±0.2	P<0.001
n-6:n-3 ratio	12.6±0.6	11.4±0.5	8.6±0.4	8.1±0.6	6.7±0.3	5.5±0.4	P<0.001

The fatty acid profile was similar before treatment diets were imposed with an n6:n3 ratio of 12.3±0.23 (mean±SE) (Table 1). There was a substantial increase in plasma EPA and DHA with fish oil supplementation of more than 1 g/kg, and a decrease in ARA level at supplementation levels more than 3 g/kg. There were no observed ill-effects of high rates of supplementation on sow health nor did live weight or P2 differ between treatments at 30 weeks of age (145.3±2.0 kg; 14.0±0.7 mm). Supplementing the diet of pubertal gilts with 3 g/kg or higher changes the long-chain polyunsaturated fatty acid profile in plasma without adversely affecting growth or health.

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## Straw bedding affects growth performance and carcass fat distribution in grower-finisher pigs

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Inherent differences between the environments of conventional (C) and deep-litter (DL) pig housing systems are large and can affect the energy metabolism of the growing pig resulting in differences in growth and fat distribution (Trezona *et al.*, 2005). The presence of bedding is a major difference between C and DL housing. Bedding may affect energy metabolism in the pig via its thermal properties or via ingestion. The aim of this experiment was to quantify the effects of straw, as bedding and via ingestion, on growth and fat distribution in growing pigs.

Ninety-six Large White x Landrace female pigs were stratified by live weight (LW) ( $16.1 \pm 0.26$  kg) at eight weeks of age into groups of six and housed in commercial grower-finisher pens within a naturally ventilated shed. The experiment was a 2x2 factorial design with two dietary treatments: 1) CD, commercial grower (13.4 MJ DE/kg 0.99% lysine, 17.6% NDF, 6.7% ADF) and finisher (13.0 MJ DE/kg, 0.68% lysine, 18.7% NDF, 7.8% ADF) diets and 2) SD, commercial grower-finisher diets fortified with 10% wheat straw (grower: 12.3 MJ DE/kg, 0.91% lysine, 22.5% NDF, 10.2% ADF; finisher: 11.9 MJ DE/kg, 0.63% lysine, 23.5% NDF, 11.2% ADF). Two floor treatments were also investigated: 1) CF, partially slatted concrete flooring and 2) SF, straw bedding as flooring (~15 cm thick). At 24 weeks of age pigs were slaughtered at a commercial abattoir and 24 hours after slaughter one side of the carcass, 12 pigs/treatment, was collected and analyzed for fat and lean content (dual energy X-ray absorptiometry). Data were analyzed by two-way analysis of variance (Genstat v8).

Pigs appeared to compensate for the energy dilution of the SD diet by increasing voluntary food intake (VFI), however there was no effect on feed conversion efficiency (FCE) ( $P > 0.10$ ). Increased gut-fill for SD fed pigs may partly explain results for FCE as pigs without access to straw had the lowest LW and pigs with straw bedding and fed the SD diet were the heaviest. Live weight was intermediate for pigs in the SD-CF and CD-SF groups. Nonetheless, pigs fed the SD diet and/or housed on bedding had higher CW and similar dressing percentage compared to pigs without access to straw indicating that actual gain was higher. It is probable that the thermal effect of bedding contributed to higher LW by reducing the pigs' energy demand for thermoregulation and sparing more energy for growth. There were no differences in P2 backfat depth ( $P > 0.100$ ) and total carcass composition. However, there was a trend ( $P < 0.10$ ) for the interaction between diet and floor to alter fat distribution in the belly and ham primals. These results suggest that the presence of straw may contribute to the growth and carcass differences observed between C and DL pigs.

**Table 1. Growth and carcass characteristics of pigs raised in different housing treatments**

	SD-CF	SD-SF	CD-CF	CD-SF	SEM <sup>1</sup>	P-value		
						Diet	Floor	D*F
VFI (kg/day)	2.53 <sup>b</sup>	2.60 <sup>b</sup>	2.38 <sup>a</sup>	2.35 <sup>a</sup>	0.055	0.003	0.697	0.963
LW (kg) at 24 weeks of age	115.1 <sup>b</sup>	119.0 <sup>c</sup>	110.4 <sup>a</sup>	114.4 <sup>b</sup>	1.59	0.005	0.023	0.963
Carcass weight (kg)	78.5	81.5	76.4	78.4	1.45	0.085	0.084	0.762
Dressing %	68.2	68.5	69.2	68.8	0.58	0.288	0.970	0.529
Fat % side	18.2	19.6	20.8	19.0	1.10	0.499	0.962	0.138
Fat % shoulder	14.0	14.1	15.7	14.3	0.62	0.214	0.462	0.169
Fat% loin	23.8	25.8	26.0	24.3	1.43	0.700	0.979	0.211
Fat% belly	29.5	32.7	33.7	31.6	1.60	0.635	0.430	0.099
Fat% ham	13.9	14.7	16.3	14.5	0.85	0.330	0.786	0.097

<sup>1</sup>SEM = pooled standard error of mean.

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## Using statistical modelling to improve the precision of image analysis based weight estimation

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Recent studies overseas (Wang *et al.*, 2006) and in Australia (Kollis *et al.*, 2007) revealed that live weight of pigs could be estimated using image analysis (IA) techniques. These techniques involve the automatic recognition and measurement (extraction) of the physical dimensions of the pigs that are then correlated with live weight of pigs. However, the measurement error of IA based systems is greater than the traditional weight scales (Kollis *et al.*, 2007). It was hypothesized that by using multi-factorial statistical modelling techniques (that rely on multiple explanatory variables) the predictive power of IA based weight estimation could be improved. Thus, the aim of this preliminary study was to assess the likely increase in predictive power that could be achieved with statistical modelling of the information extracted from the pig images.

An automated weighing scale (Survey Scale, Osborne Inc USA) and image capture system described by Finch *et al.* (2006) were used in a fully slatted and tunnel ventilated grower/finisher building to collect the weight data and corresponding images of 77 male Large White pigs. The methodology used for feature extraction (area, length and width of pigs) has been described in details previously (Finch *et al.*, 2006; Kollis *et al.*, 2007). Using the extracted information, models (using single and multiple explanatory factors) were developed using Proc GLM (SAS Inc v6.0) and the resulting predictive power of the models was compared (Table 1).

**Table 1. Comparison of models using different explanatory variables**

Model based on:	Length	Maximum width	Area	Multi-Factorial
Model terms	I, L, L <sup>2</sup>	I, MW	I, A, A <sup>2</sup>	I, MW, A, A <sup>2</sup>
R <sup>2</sup> (%)	36	55	71	76
Residual SD* (kg)	3.54	2.93	2.36	2.19
95% CI** (kg)	13.86	11.47	9.26	8.57

\* Standard deviation; \*\* Confidence interval; I=intercept; L=length; MW=maximum width; A=area.

Among the single variable models, the model based on area measurement produced the best results, while the model based exclusively on the length measurement featured poorly. However, the multi-factorial model, which combined the maximum width of pigs and area measurements produced the best results. The model with multiple explanatory factors accounted for more variation and hence model accuracy increased as the residual variation and confidence intervals were reduced. However, the precision of weight estimation needs to be improved further. It is envisaged that by including more biologically meaningful measurements in the predictive models, the precision of weight estimation could rival traditional measurements. Future work is also planned to estimate dressing and retail yields of pig carcasses using similar statistical modelling approach.

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## Differences in colostrum and milk composition between gilts and sows

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Colostrum ingestion within the first 12 hours post-partum is vital for the establishment of normal patterns of growth and development in all neonate mammals. Colostrum contains specific peptides, proteins and immunoglobulins that are essential for immune protection and for proper development of the absorptive capacity of the gastrointestinal tract. A key concern of pig growers is that gilt progeny do not grow as efficiently as sow progeny and therefore represent a potential drain on production efficiency. To investigate this we undertook a preliminary survey of colostrum and milk composition of sows and gilts to determine if the inferior growth performance of gilt progeny was associated with an inferior milk and/or colostrum composition.

Colostrum samples were collected from primiparous gilts (n=5) and multiparous sows (n=5) from a single farrowing (7 February 2007). All animals were Large White x Landrace pigs in a commercial farrowing shed at Corowa NSW (QAF Meat Industries Pty Ltd). Farrowing was assisted by administering 1.5 units of oxytocin. Using manual palpation of the rear teats, 5 mm of colostrum was collected within one hour of placental presentation and 5 mm of milk was collected at day three (D3) of lactation. Samples were immediately put on ice and within 30 minutes of collection were frozen at minus 20°C overnight, then stored at minus 80°C until assayed. Protein concentrations were analyzed using a BCA™ Protein Assay Kit. Lactose was measured with a lactose/D-galactose kit (Arrow Scientific Pty Ltd) measurements obtained from spectrophotometer readings at 340 nm. Fat was analyzed by haematocrit. All analyses were duplicated and analyzed statistically by two sample T-tests (Genstat).

**Table 1. Protein, lactose and fat composition in sow and gilt colostrum (about one hour post-partum) and milk (day three of lactation). Data presented as mean±SEM**

	Protein mg/ml	Lactose mg/ml	Fat (%)
Gilt colostrum	261.3± 9.3 <sup>a</sup>	2.88+0.24	4.6+1.2 <sup>a</sup>
Sow colostrum	213.4±27.7 <sup>a</sup>	2.97+0.14	8.2+1.2 <sup>b</sup>
Gilt milk D3	144.8±27.7 <sup>b</sup>	2.94+0.29	8.8+0.4 <sup>b</sup>
Sow milk D3	116.30±3.1 <sup>b</sup>	2.97+0.13	8.4+1.4 <sup>b</sup>

Mean values in the same column with different superscripts are significantly different (P<0.01).

The protein concentration in gilt and sow colostrum was significantly higher (P<0.01) than that in milk collected at D3 with the protein values for milk higher than those reported previously (Attwood and Hartmann, 1993). However, there was no statistical difference in the protein concentration of gilt and sow colostrum or milk. In contrast, sow colostrum contained proportionally more (P<0.03) fat than gilt colostrum although this difference was not sustained through to day three. Sow colostrum may have a higher fat supply than gilt colostrum due to differences in energy partitioning between milk fat and tissue growth in the two age classes. Lactose concentrations remained relatively unchanged between colostrum and milk and gilt and sows. The superior performance of sow progeny may be related to the availability of more energy substrate early in development.

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## Providing finisher pigs with a choice of cooling system does not improve performance or reduce carcass variation

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When the ambient temperature reaches the upper critical level of 31°C pigs must be provided with supplemental cooling to ensure that growth performance and welfare are not affected (Giles and Black, 1991). Pigs have a limited capacity to increase heat loss to remain within their zone of thermal comfort. Furthermore, pigs within a group situation show individual behavioural responses when given a choice to control their micro-climates (Morrison and Smits, 2005). The aim of this experiment was to investigate the use of standard shed spray cooling, wallows and animal-activated spray cooling systems on growth performance, carcass characteristics and pig respiration rates.

Thirty pens of 38 entire males (0.84 m<sup>2</sup>/pig) were allocated to either: 1) a control-standard shed spray-cooling system (activated when temperature exceeded 26°C, turned on for five minutes and off for 15 minutes); 2) wallows, a 2.4 x 1.5 x 0.30 m structure in the pen filled with water; or 3) animal-activated spray cooling, a shower head activated by infrared motion sensor. The pigs were housed in their respective treatments from 16 to 22 weeks of age. Measures of respiration rates were conducted on 80 pigs per treatment at 21 weeks of age. Their average daily temperature was 29°C. Data were analyzed by analysis of variance with the pen as the experimental unit.

Growth performance, carcass weight and backfat and variation within the pen were not improved ( $P>0.05$ ) when pigs were provided with a choice of animal-activated cooling systems (Table 1). The pigs in the control treatment had significantly higher ( $P<0.05$ ) respiration rates than the other two cooling treatments, which indicates that they had adapted to the heat stress. These data indicate that the wallow and animal-activated spray cooling systems may have enabled the pigs to control their microclimates more effectively.

**Table 1. Average growth, carcass characteristics and respiration rates of pigs housed in pens with standard shed cooling (control), wallows or animal-activated spray cooling systems**

	Control	Wallow	Animal-activated spray cooling	SEM
Start weight (kg)	56.1	57.7	56.8	0.75
Rate of gain 16-22 weeks (kg/day) <sup>1</sup>	0.921	0.895	0.885	0.01
Carcass weight (kg)	72.0	73.1	72.3	0.68
Carcass P2 (mm) <sup>2</sup>	9.3	9.3	9.2	0.08
Respiration rate (breaths/min)	42.9a	35.3b	35.9b	1.42
Coefficient of variation between carcass weight	14.5	13.0	13.3	-
Coefficient of variation between carcass P2 backfat	22.0	21.3	23.0	-

<sup>a,b</sup>Within rows, means with different superscripts are significantly different ( $P<0.05$ ); <sup>1</sup>rate of gain adjusted for standardized start weight; <sup>2</sup>carcass P2 adjusted for standardized carcass weight.

The financial support from Australian Pork Limited and the technical assistance from the management and staff at QAF Meat Industries Pty Ltd are gratefully acknowledged.

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## The effects of organic mineral supplementation on sow productivity: an overview

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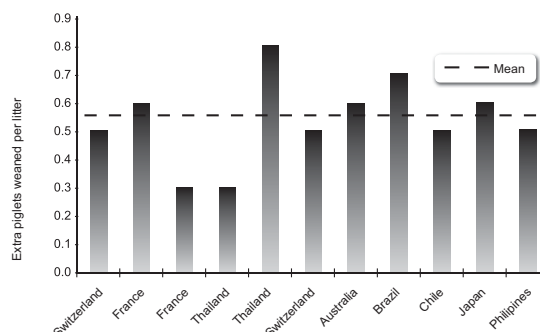
Close Consultancy, Wokingham, Berkshire RG41 2RS UK.

Trace minerals play a fundamental role in ensuring good sow reproductive performance, not only in terms of piglets weaned per litter or per year, but also in lifetime performance. However, it is questionable whether current trace mineral recommendations, such as those of the NRC (1998), meet the needs of the modern hyperprolific sow. There is also concern about the source of minerals and it is generally accepted that organic minerals, with their greater bioavailability and bioactivity, better meet the needs of the modern sow. Indeed, Fehse and Close (2000) reported that the supplementation of a sow diet with a special combination of organic minerals (Bioplex Sow-Pak™: Se, Cr, Fe, Cu, Zn and Mn; Alltech Biotechnology Pty Ltd) resulted in an extra 0.5 piglets weaned per litter for sows weaning 26.5 piglets per year.

Since this original study, a number of commercial and field trials have been conducted in which Bioplex Sow-Pak™ (with or without Cr) partially replaced or was added on top of the standard inorganic mineral supplement in the diet of the sow during both gestation and lactation. Performance was measured over a single parity or multiple parities for sows of different breeds in several countries throughout the world. The number of animals in the trials varied between 100 and >1,000 (Table 1).

**Table 1. The effect of organic mineral supplementation on litter size (mean±SE)**

	Total born	Born alive	Weaned	Mortality (%)
Control	10.98 ±1.5	10.03 ±1.27	9.30 ±0.9	7.3
+ Sow-Pak	11.55 ±1.4	10.68 ±1.29	9.83 ±0.9	7.9



**Figure 1.** Extra piglets weaned per litter for studies conducted with supplementation of sow diets with Sow-Pak™ in different countries

Supplementing Bioplex Sow-Pak™ to the diet of the sow during gestation and lactation increased the number of piglets born, born alive and weaned ( $p>0.05$ ). There was a small decrease in the number of stillborn piglets and a small increase in pre-weaning mortality. Litter size at weaning was increased by  $0.53\pm0.15$  ( $n=11$ ) ( $p>0.05$ ) piglets, with the response varying between 0.3 and 0.8 extra piglets per litter (Figure 1). Assuming 2.3 litters per sow per year, this improvement equates to an extra 1.2 weaned piglets per sow per year.

Litter size had no effect on the number of additional piglets weaned, with 0.49, 0.58 and 0.51 extra piglets for litter sizes of <9, 9-10 and >10 piglets, respectively.

The supplementation of the diet of the modern sow with organic minerals improved sow productivity and was also cost-effective, with a calculated ROI value of 4.5:1.

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## Injectable progesterone does not reduce mid to late pregnancy failure of sows during seasonal infertility

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Reproductive failure is a significant cause of culling sows. Pregnancy failure due to irregular returns to oestrus and non-pregnant sows at expected farrowing can account for 14% of non-age based culling of sows (Hughes and Smits, 2002). Social stresses are related to increased rate of pregnancy failure (Peltoniemi *et al.*, 2000) and may be mediated through a decline in plasma progesterone (P4). Low P4 may occur during seasonal infertility or when sows are relocated or mixed in groups after pregnancy diagnosis. Sows may benefit from exogenous P4 during these stressful periods. Lopez-Gatius *et al.* (2004) reduced pregnancy loss by supplementing lactating dairy cattle with P4. In an earlier experiment, we observed a trend for improved farrowing rate when sows received one intramuscular (i.m.) injection of 125 mg P4 on day 42 postcoitum (Johnston *et al.*, 2005). These results required verification with a statistically valid sample size.

We hypothesized that a single injection of P4 on day 34 post-mating, when sows are mixed in group pens, would increase the proportion of sows remaining pregnant to full term when subjected to the stresses associated with mixed social groups and environmental conditions during seasonal infertility. Large White x Landrace F1 sows (n=522) were selected randomly from matings during late summer (week two through to week eight) in a commercial production unit in Victoria. Sows were mated artificially and housed in individual stalls from weaning through 34+1.5 days post-mating. On day 34 post-mating, after positive diagnosis of pregnancy by ultrasound, sows were moved from stalls to group pens of 17-18 sows per pen (1.4 m<sup>2</sup>/sow). Treated sows received one i.m. injection of 125 mg P4 (Jurox Pty Ltd progesterone) in the neck while control sows received no injection. Sows from each treatment were housed together in pens from day 34 post-mating until farrowing or until the sow failed to farrow. Categorical data were analyzed using Chi square. Treatment effects on litter size were analyzed using analysis of variance with sow parity (range=0-8) used as a covariate. Sow was considered the experimental unit.

Progesterone injection did not affect the incidence of pregnancy failure after week five of gestation in this experiment (Table 1). Overall, 12.6% of sows confirmed pregnant at five weeks of gestation failed to maintain pregnancy and typified seasonal infertility on this farm. There were no interactions between P4 treatment and sow parity or week of mating for pregnancy loss. Litter size at farrowing was not affected by P4 treatment. Distribution of litter sizes at subsequent farrowing (range: 3-18 live pigs) was not affected by P4 treatment ( $\chi^2 = 3.94$ ;  $P < 0.42$ ).

We conclude that one i.m. injection of P4 at the time of moving sows from stalls to group pens at week five of gestation does not reduce pregnancy loss during a time of seasonal infertility.

**Table 1. Effect of exogenous P<sub>4</sub> on pregnancy loss and litter size**

Trait	Control	P4	Pooled SE	Significance
Sows allocated at 5 weeks	264	258		
Farrowed	233	223		$\chi^2 = 0.39$ ; ( $P < 0.53$ )
Pregnancy failed	31	35		
Litter size at farrowing:				
Born live	11.5	11.5	0.36	$P < 0.36$
Stillborn	0.48	0.48	0.03	$P < 0.36$
Mummies	0.43	0.43	0.04	$P < 0.97$

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## Tag design of high frequency and ultra high frequency radio frequency identification for pig tagging

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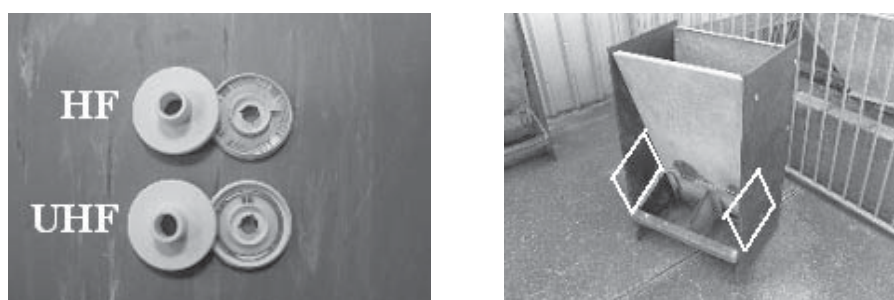
Pig identification is mandated by the Australian Livestock Disease Control Act (1994) to prevent disease spreading and to maintain consumer confidence in the quality of Australian pork. Radio frequency identification (RFID) offers automated identification. Current RFID technology used to identify cattle and sheep is based on the low frequency (LF) system. This LF system is based on ISO 11785 and does not offer anti-collision capability. As small piglets may be in the RF field at the same time, tagging systems for piglets would require anti-collision sensitivity. This paper presents the use of high frequency (HF; 13.56MHz) and ultra-high frequency (UHF; 920 – 926MHz) RFID tags for pig tagging in Australia. The RFID tags designed and presented in this paper can be attached to the pig's ear, offer anti-collision capability and are passive and reusable.

An RFID pig tag must be tamper and waterproof to protect it from the harsh environment. The HF and UHF RFID tag antenna designs presented here are based on Leong (2007) but modified to fit in the encapsulation casing (Figure 1). Texas Instruments Inc HF Tag-it™ chips and UHF C1G2 straps were attached to the newly designed antennas during the making of the HF and UHF RFID tags. Thirty HF and UHF RFID tags were fabricated for each laboratory testing and the average read range in free space and on human hands (to mimic pig's ears) determined (Table 1). The performance of the HF tag was limited by the size of the reader antenna while the performance of the UHF tag was reduced by half when attached to the human hand.

**Table 1. Average read range for the designed HF and UHF RFID tags**

Read Range	HF RFID Tag (m)	UHF RFID Tag (m)
Free Space	0.34	0.80
Placed on Hand	0.32	0.40

Field-testing of the pig ID system is planned on a commercial piggery in Victoria in August 2007 to determine how frequently pigs approach a feeder to obtain food. The pig feeder to be used in the trial is shown in Figure 1. The two reader antennas on the feeder face each other and are separated by a distance of 0.6 m. Pig ID will only be recorded when pigs place their heads into the feeder bin. Data recorded during the commercial trial will include date, time, antenna ID and the unique ID of the tag attached on the pigs' ears.



**Figure 1.** HF tag and UHF tag before encapsulation (left); Pig feeder showing position of the reader antennas (right)

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## Prediction of whole-body chemical composition using computed tomography

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Although computed tomography (CT) has been used in live pigs and carcasses as an alternative to physical carcass dissection, few studies have measured the association between chemical composition in the whole body and tissue mass measured using CT imaging. Kolstad (2001) demonstrated that it is possible to use CT imaging in live pigs to record muscle and fat mass in the whole body including the viscera. The objective of this study was to test the hypothesis that muscle and fat mass recorded in live pigs using CT imaging is closely associated with chemical protein and fat content in the whole body.

Forty-five male, female and castrate male pigs (hybrid, mainly Large White x Landrace) were allocated at 10 weeks of age and  $30.8 \pm 4.8$  (mean  $\pm$  SD) kg live weight to individual pens in four rooms maintained at 22°C. The pigs were fed *ad libitum* a commercial, pelleted diet adequate in digestible energy with free amino acids in excess of requirements. Water was provided using nipple drinkers. Nine pigs (three males, three females and three castrate males) were anaesthetized intravenously with a mixture of xylazine (1 mg/kg) and ketamine (10 mg/kg) at close to each of five live weights (30, 60, 90, 120 and 150 kg). Cross-sectional images were collected along the whole body at 10 mm intervals using a Picker PQ-2000 spiral CT scanner. During recovery, pigs were given intravenous yohimbine (5-10 mg). On the day after CT scanning each pig was sedated with intravenous barbiturate and then exsanguinated, eviscerated and sawn down the dorsal midline to record side weight. Intestinal contents were removed to record visceral weight. The left side (including the head, skin and hair) and viscera were frozen separately. Each side and viscera were minced, mixed, sampled (0.5 kg), freeze-dried and analyzed for nitrogen and fat content. The CT cross-sectional images for each animal were analyzed for muscle and fat volume (including viscera) using a Voxel Q workstation. Muscle and fat weight were calculated from volume and density measurements based on the Hounsfield unit range (Hounsfield, 1980). The interaction between pig sex and CT tissue weight was significantly different for chemical fat only (see Table 1). Hence the relationship between chemical protein and CT tissue weight was analyzed independently of animal sex using linear regression.

**Table 1. Wet<sup>1</sup> chemical protein and fat weight in the whole body as a function of CT muscle and fat weights (including viscera) in 45 pigs from 30 to 150 kg live weight**

Dependent variable	Independent variable	Intercept (SE)	Slope (SE)	R <sup>2</sup>	RSD
Chemical protein <sup>2</sup> (kg)	CT muscle (kg)	5.98 (1.458)	0.26 (0.024)	0.749	3.432
Chemical fat (kg)	CT fat (kg) - male	4.03 (1.640)	1.21 <sup>a</sup> (0.102)	0.936	3.714
	- female	6.72 (1.672)	0.94 <sup>b</sup> (0.080)		
	- castrate	2.59 (1.702)	1.25 <sup>a</sup> (0.076)		

<sup>1</sup>Chemical components corrected for sample moisture content; <sup>2</sup>Nitrogen x 6.25; <sup>a,b</sup>Slopes sharing the same superscript are not significantly different.

This research supports the hypothesis that CT imaging of muscle and fat content in live pigs is closely associated with chemical protein and fat content in the whole body. Computed tomography accounted for 74.9% of the variation in chemical protein and 93.6 % of the variation in chemical fat in the whole body. The inherent sampling difficulty associated with mincing of heavy carcasses is a likely source of error when measuring carcass protein and fat content. The relationships developed in this study will allow the prediction of chemical composition from CT assessment of muscle and fat content in individual animals scanned serially from 30 to 150 kg live weight.

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## Sow behaviour and piglet overlay in farrowing pens

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More piglet deaths occur in farrowing pens than crates due to overlying by the sow, particularly when sows descend from standing to lying (Marchant *et al.*, 2000). In this study we examined the relationship between sow behaviour in Werribee farrowing pens (WFP) constructed with internal features to encourage changes in sow posture and the incidence of piglet deaths from overlying in those pens.

The study involved 24 Large White x Landrace sows over six replicates in time. Sows ranged from parity 1-3 and had not previously farrowed in pens. Four WFPs were constructed so that the design of the 'nest' area of the WFPs could be changed in a 2x2 factorial arrangement by altering the shape of the piglet creep area (triangle vs. rectangle) and the angle of the sloping panels (10° vs. 40° from vertical). Within replicates there were four 'nest' designs and between replicates the positions of the 'nest' design treatments were randomized across the four pens. Sow behaviour was recorded using low-light cameras and time-lapse video during 72 hours commencing from the birth of the first piglet in each litter. Piglet production and mortality data were also collected. The incidence of any piglet dying from overlay within a litter was related to behaviour measurements and production parameters, together with factors for replicate, creep shape and panel angle treatments using logistic regression models with Bernoulli errors. Models were compared using Chi-square change in deviance tests. The most parsimonious model was chosen as having, on a logistic scale, a linear response to the angular transformation of the percentage of occasions sows were against a sloping panel when they descended from standing to lying (%Panel Use; P=0.011). Production parameters, replicate and treatment factors did not modify this relationship (P>0.1). Confidence intervals for predicted probability of an overlay in a litter, at a given %Panel Use, were calculated using a standard asymptotic normal approximation on the logistic scale followed by back-transformation. The range in piglets born live per litter was 4-16 (median 11) and overlays ranged from 0-5 (median 1). Sows descended from standing to lying between 8-59 times (median 25.5) and %Panel Use ranged from 35.6-100% (median 92.8%). High panel use behaviour minimized the incidence of any overlying deaths (Table 1).

**Table 1. Sow behaviour during descent from standing and piglet deaths from overlay**

<b>Percentage Panel Use</b>	<b>50</b>	<b>60</b>	<b>70</b>	<b>80</b>	<b>90</b>	<b>100</b>
Predicted probability of overlay in a litter	0.92	0.88	0.81	0.70	0.54	0.19
95% Lower Confidence Limit	0.51	0.48	0.46	0.41	0.31	0.04
95% Upper Confidence Limit	0.99	0.98	0.95	0.89	0.75	0.54

This result reinforces the importance of designing farrowing pens that encourage the use of panels by sows when they descend from standing to a lying posture. The Werribee farrowing pen has sloping panels in the 'nest' area to assist this type of behaviour (Cronin *et al.*, 1996).

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## Performance and gastric ulcers in growing-finishing pigs fed alfalfa hay meal or a coarse-milled pelleted diet

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Danish producers using pelleted feed and experiencing problems with gastric ulcers in their herd are sometimes advised to use diets containing alfalfa hay meal or diets that have greater mean particle size. However the effects of such interventions have not fully been investigated. These experiments investigated the effect of alfalfa hay meal and coarser milling in pelleted diets on growth performance and occurrence of gastric ulcers in growing-finishing pigs.

In Experiment 1, the outcome of dietary inclusion of 10% alfalfa hay meal in a pelleted diet (2 mm pellets) on performance and gastric ulceration was investigated in one herd. There were 10 pigs per pen and 12 pens per treatment. In Experiment 2, the effect of coarser milling in pelleted diets (3.5 mm pellets) was studied in two herds. There were 15 to 16 pigs per pen and 41 replicates in herd A and 34 replicates in herd B. In all studies the pigs were on trial from about 30 kg until slaughter at around 100 kg. The main ingredients in the diets in both experiments were wheat, barley and soybean meal. Stomachs were collected at the abattoir and lesions in the pars oesophagea were scored on a scale from 0-10, 0 being normal and 10 having severe gastric ulceration (Christensen, 1998). The influence of dietary treatment was analyzed univariately in a normal linear model using the GLM procedure in SAS (SAS Inc v.9.13). Stomach scores are categorical data and were further analyzed using the GENMOD procedure.

No significant effect of alfalfa hay meal was detected in Experiment 1 (Table 1). In Experiment 2, there was a significant interaction between treatment and herd, probably because the particle size distribution differed between herds. In Herd A, degree of milling did not significantly affect performance; however the number of pigs with more severe gastric lesions was higher among pigs fed the fine pellets. In Herd B, FCR was significantly poorer in pigs fed the more coarse diet. This was supported by a significantly higher starch content in the faecal dry matter of pigs fed the coarse pellets in both Herd A and Herd B (data not shown), suggesting that the pigs were not able to fully use the nutrients in these diets.

**Table 1. Effect of dietary treatment on performance and stomach ulceration score**

Experiment	n	Particle size, (<1/1-2/2-3/>3) (mm)	<sup>1</sup> ADFI (kg/day)	<sup>2</sup> ADG (g/day)	<sup>3</sup> FCR (kg/kg)	Ulcer score (n)	Ulcer score > 5 (%)
Exp. 1, control	12	-	2.25	964	2.33	4.7 (n=93)	54
Exp. 1, 10% alfalfa hay meal	12	-	2.23	955	2.33	4.1 (n=95)	47
SEM			0.023	5.6	0.023	0.22	
Exp. 2, Herd A, fine pellets	41	56/37/7/0	2.26	832	2.72	2.5 <sup>a</sup> (n=253)	14.6 <sup>a</sup>
Exp. 2, Herd A, coarse pellets	41	38/34/28/0	2.23	821	2.71	1.6 <sup>b</sup> (n=248)	8.1 <sup>b</sup>
SEM			0.012	5.0	0.015	0.10	
Exp. 2, Herd B, fine pellets	34	79/18/2/1	1.86	766	2.43 <sup>a</sup>	2.6 (n=141)	16.3
Exp. 2, Herd B, coarse pellets	34	52/35/12/0	1.89	760	2.49 <sup>b</sup>	2.3 (n=130)	13.1
SEM			0.012	6.02	0.014	0.15	

<sup>1</sup>ADFI: average daily feed intake; <sup>2</sup>ADG: average daily gain, <sup>3</sup>FCR: feed conversion ratio. <sup>a,b</sup>columns within an experiment with different letter subscript are significantly different (P<0.05).

It was concluded that dietary inclusion of 10% alfalfa hay meal does not affect performance or gastric health. Coarser milling of pelleted diets may compromise FCR but can reduce the prevalence of more severe gastric ulcers in some herds.

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## Effect of dam parity on transmission of proliferative enteropathy

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Proliferative enteropathy (PE) is an endemic disease responsible for substantial production losses in pigs. The causative agent, *Lawsonia intracellularis*, is transmitted via contact with infected faeces. Experimental challenge studies indicate that passive immunity derived maternally protects the progeny of seropositive dams, either preventing or reducing disease in suckling piglets (Poza *et al.*, 2000; Barna and Bilkei, 2003). Although *L. intracellularis* infection is predominantly detected in pigs after weaning, dams (particularly gilts) may be a source of *L. intracellularis* infection to their suckling piglets (McOrist and Gebhart, 1999; Bronsvort *et al.*, 2001). This study aimed to determine whether the dam was an important source of *L. intracellularis* infection for her progeny and whether dam parity had an impact on faecal excretion of *L. intracellularis*.

Gilts and sows (parity 2-5) from two commercial herds were tested for serum IgG antibodies to *L. intracellularis* five weeks before farrowing. Seropositive and seronegative dams (30 gilts and 30 sows of each serostatus) were randomly selected across the two herds. Herd 1 sows/gilts were housed in groups of at least 64 pigs in ecosheds and Herd 2 sows/gilts were housed in individual stalls throughout gestation. Medication programs preventing *L. intracellularis* infection were not used during gestation or lactation. Strict biosecurity was enforced in the two farrowing sheds throughout the trial and personnel entering farrowing crates were required to use single-use boot covers to reduce the potential for *L. intracellularis* contamination from the environment. Faecal samples were collected from the dams at 7, 14, and 21 days after farrowing for detection of *L. intracellularis* by polymerase chain reaction (PCR). DNA was extracted from the pooled faeces of each sow and amplified by a *L. intracellularis*-specific PCR. This assay can detect 103 *L. intracellularis*/g of faeces, which is 100-fold more sensitive than that required to detect sub-clinically infected pigs (Collins *et al.*, 2000). Sera collected from at least one pig per litter at four and eight weeks of age was tested for IgG antibodies to *L. intracellularis*.

*L. intracellularis* DNA was not amplified from any pooled faecal sample of any sows or gilts. DNA extraction controls and PCR assay controls all produced the expected results. Serum IgG antibodies to *L. intracellularis* were not detected in pigs from either seropositive or seronegative gilts or sows at four or eight weeks of age. There was no evidence of *L. intracellularis* excretion in the faeces of seronegative or seropositive gilts/sows at any time during lactation. Likewise, there was no serological evidence of *L. intracellularis* infection in their progeny at weaning and four weeks later. Dams do not appear to be an important source of *L. intracellularis* to their piglets during lactation.

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## Impact of pneumonia on activity in grower gilts

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Changes in physiological activity due to disease need to be accounted for when evaluating the nutritional requirements of pigs. The aim of this experiment was to measure changes in activity in response to experimental pneumonia due to the common pathogens *Mycoplasma hyopneumoniae* (*Mhp*) and *Pasteurella multocida* (*Pm*).

Gilts were assigned at 10 weeks of age ( $24 \pm 4$  kg) to four treatment groups of 16-17 pigs, of which two were challenged one week later with *Mhp* by intratracheal inoculation. At 14 weeks of age, one of these groups and a further group was challenged intratracheally with *Pm*; the final group was unchallenged controls. The groups were housed in separate rooms of a controlled environment facility, in individual pens, with temperature maintained in a range of 21-25°C and fed *ad libitum* on an antibiotic-free commercial pellet diet. Activity was monitored remotely using continuous time lapse VHS video surveillance equipment capturing 16-17 periods of activity per hour and 23-24 hours per tape, rotating continuously between four treatment rooms. As a period of <60 seconds was available at each time point to score activity, two operators each recorded four of eight designated pens/pigs per room. Activity of each pig was recorded as either: 1) sleeping/lying with head down; 2) resting/lying with head up; 3) standing or walking; or 4) eating. Activity was quantified approximately every three days from 12 days before *Pm* challenge until six weeks after *Pm* challenge, over one hour in the morning (0730-0830 hours) and afternoon (1430-1530 hours), totalling 32 hours per pig. The proportion of time spent among the four activities was calculated for the 32 individual pigs at each inspection. Data from each activity was fitted with a linear mixed model of fixed (treatment+day+interaction) and random (animal+error) effects. Treatment means were compared using Fisher's F protected least significant difference test. Day variation was assumed independent as there was no continuity between consecutive days. Residual maximum likelihood (REML) estimation was used to estimate all parameters.

The respiratory pathogens had significant effects on sleeping, lying and eating activity (Table 1). While *Mhp* infection alone had little impact on activity, *Pm* and combined *Mhp* and *Pm* infection resulted in a significantly greater frequency of sleeping and less eating activity than controls. Combined *Mhp* and *Pm* infection in particular was associated with significantly more time spent in sedentary (sleeping and lying) activities. The reduced activity associated with these infections has implications in quantifying physiological effects of pneumonia for the AUSPIG model.

**Table 1. Treatment means of time spent (%) in four activities following challenge with *Mycoplasma hyopneumoniae* (*Mhp*), *Pasteurella multocida* (*Pm*) and combined challenge**

Treatment	1) Sleeping	2) Lying	3) Standing	4) Eating
Control	51.7 <sup>b</sup>	10.2 <sup>ab</sup>	16.2	21.8 <sup>a</sup>
<i>Mhp</i>	46.2 <sup>b</sup>	13.3 <sup>a</sup>	18.8	21.7 <sup>a</sup>
<i>Pm</i>	61.3 <sup>a</sup>	11.6 <sup>a</sup>	13.4	13.6 <sup>b</sup>
<i>Mhp</i> + <i>Pm</i>	70.0 <sup>a</sup>	6.5 <sup>b</sup>	13.1	10.5 <sup>b</sup>
LSD 5%	9.3	3.8	5.8	5.3
Fixed terms (df)	F-probabilities			
Treatment (3)	<0.001	0.007	NS	<0.001
Day (31)	<0.001	<0.001	<0.001	<0.001
Interaction (93)	<0.001	<0.001	<0.001	<0.001

<sup>ab</sup> indicate significant treatment effects.

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## Do enterotoxigenic *Escherichia coli* strains isolated from Australian pigs carry transferable antimicrobial resistance genes of public health significance?

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Antibiotic use in piggeries to control bacterial pathogens with variable resistance patterns, such as enterotoxigenic *Escherichia coli* (ETEC), could potentially drive the development of antimicrobial resistance of significance to public health, especially if the antibiotics also have a high importance rating in human medicine. In Australia, antimicrobial use in food producing animals is highly regulated with many drugs not registered for use including fluoroquinolones and clavulanic acid potentiated  $\beta$ -lactams. In this study we hypothesized that Australian ETEC isolates will contain few resistance genes of public health significance. However, the aminoglycoside apramycin may select for antimicrobial resistance genes that give cross-resistance to gentamicin. In addition, off-label use of the third generation cephalosporin ceftiofur to treat multi-drug resistant (MDR) ETEC may drive selection of plasmid-mediated  $\beta$ -lactamase genes that are becoming prevalent in hospital-adapted Gram negative pathogens. Therefore, it is important to determine the transferable resistance genes present in Australian ETEC isolates and whether they are located within mobile genetic elements such as integrons and plasmids.

ETEC (n=117) isolated from diseased piglets were collected from five states of Australia (1998-2006). We have previously reported their resistance profiles to 13 different antimicrobial agents, including enrofloxacin, ceftiofur, florfenicol, apramycin, neomycin and gentamicin (Smith *et al.*, 2005). Polymerase Chain Reaction (PCR) primers (n=28) targeting resistance genes for each major class of antimicrobial were designed and applied.

No specific resistance genes to ceftiofur (blaCMY) or florfenicol (florR) were detected in the collection. Resistance to antimicrobials used historically, such as streptomycin, tetracycline and ampicillin was high. Resistance to apramycin and neomycin was moderate and corresponding resistance genes aac(3)-IV and apha(3)-I were identified in 34.1% and 32.4% of isolates, respectively. Of the 59.8% of isolates confirmed to carry a class I integron, 47% produced an amplicon using PCR primers targeting the variable gene cassette region. A total of five different sized gene-cassettes were sequenced and identified (Table 1), but none contained resistance genes that could be regarded as significant to public health. Australian porcine ETEC have acquired a large collection of potentially transferable resistance genes, but these tend to reflect antimicrobial agents used historically. No resistance genes of public health significance imparting specific resistance to recently introduced drugs (ceftiofur and florfenicol) have yet been detected. However, whether this is also the case for commensal *E. coli* isolates is yet to be determined.

**Table 1. ETEC isolates carrying class 1 integrons and identified gene-cassette inserts**

Integron amplicon size (base pairs)	Number of ETEC isolates (%)	Integron-associated resistance genes	Resistance to:
2000 bp	6 (5.1%)	<i>aadA2/dhfrXII</i>	Aminoglycosides Trimethoprim
1650 bp	1 (0.8%)	<i>aadA5/dhfrXVII</i>	Aminoglycosides Trimethoprim
1100 bp	47 (40.2%)	<i>aadA1</i>	Aminoglycosides
650 bp	2 (1.6%)	<i>sat</i>	Streptothricin
500 bp	6 (5.1%)	<i>dhfrV</i>	Trimethoprim

This work is supported by Australian Pork Limited.

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## Use of computed tomography in live pigs to measure the impact of pneumonia on carcass composition

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Although previous investigations (Le Grand and Kobisch 1996) have studied the effect of pneumonia on live performance and carcass composition in growing pigs, there is a lack of information on real-time changes in lung lesion size, live performance and body composition. The objective of this study was to use computed tomography (CT) in live pigs to measure performance and changes in lung lesion size and body composition due to pneumonia.

Sixty-four hybrid (mainly Large White x Landrace) gilts from a high-health status herd free of *Mycoplasma pneumoniae* were allocated to individual pens at  $24 \pm 4$  kg (mean  $\pm$  SD) live weight, and *ad libitum* fed an antibiotic-free diet with adequate digestible energy. At 11 weeks of age, pigs in two rooms were challenged with *Mycoplasma hyopneumoniae* (*Mhp*). Three weeks later, pigs in one of these rooms, and pigs in a third room, were challenged with *Pasteurella multocida* (*Pm*). Pigs in the fourth room were unchallenged controls. Before *Pm* challenge and at 2.5-3 weeks (subacute phase) and 5-6 weeks (chronic phase) post-challenge, cross-sectional images were collected using a spiral CT scanner. From tissue and lung lesion volumes measured on a Voxel Q workstation, density measurements were used to calculate tissue weights and carcass composition. Data were analysed using analysis of variance and treatment differences tested by Fisher's F protected LSD test. Logarithmic transformation was used to stabilize variances for lung lesion data.

Alone, *Mhp* infection affected only 6% of lung volume by 2.5-3 weeks post-challenge and had no significant effect on either live performance or body composition compared to controls (Table 1). In contrast, combined infection with *Mhp* and *Pm* affected 16% of lung volume and significantly reduced feed intake ( $P < 0.001$ ) and body fatness in both sub-acute and chronic phases ( $P < 0.05$ ). The use of computed tomography to measure real-time changes in lung lesions and body composition in live pigs enables the effects of pneumonia to be predicted in models like AUSPIG.

**Table 1. Effect of pneumonia of varying severity due to *M. hyopneumoniae* (*Mhp*), *P. multocida* (*Pm*) or combined infection on average daily gain, feed intake and carcass fat and muscle. Time of measurement of means relates to weeks after *Pm* challenge and different superscripts indicate significant differences between treatments**

	Week	Control	Mhp	Pm	Mhp + Pm	LSD 5%	P
ADG (Kg/d)	-4 to 6	0.95 <sup>a</sup>	0.89 <sup>ab</sup>	0.81 <sup>b</sup>	0.74 <sup>c</sup>	0.072	< 0.001
Feed intake (Kg/d)	-2 to 0	1.70	1.84	1.91	1.76	0.02	NS
	0 to 4	2.15 <sup>a</sup>	1.93 <sup>ab</sup>	1.65 <sup>b</sup>	1.30 <sup>c</sup>	0.03	< 0.001
Log (lung lesion % + 0.1)	-1	-0.8863 <sup>b</sup>	0.1563 <sup>a</sup>	-0.8041 <sup>b</sup>	0.1147 <sup>a</sup>	0.357	< 0.001
	2.5-3	-0.9654 <sup>c</sup>	0.6670 <sup>b</sup>	0.4256 <sup>b</sup>	1.0979 <sup>a</sup>	0.345	< 0.001
	5-6	-0.3984 <sup>b</sup>	-0.3751 <sup>b</sup>	0.2902 <sup>a</sup>	0.6555 <sup>a</sup>	0.440	< 0.001
Carcass fat (% by wt)	-1	9.3 <sup>b</sup>	11.2 <sup>a</sup>	10.6 <sup>a</sup>	10.1 <sup>ab</sup>	1.255	0.027
	2.5-3	13.1 <sup>a</sup>	12.6 <sup>a</sup>	12.7 <sup>a</sup>	10.2 <sup>c</sup>	1.966	0.017
	5-6	15.1 <sup>b</sup>	14.3 <sup>ab</sup>	15.1 <sup>a</sup>	12.2 <sup>b</sup>	2.29	0.047
Carcass muscle (% by wt)	-1	64.9	63.5	63.5	64.4	1.35	NS
	2.5-3	62.6 <sup>b</sup>	63.0 <sup>b</sup>	62.5 <sup>b</sup>	64.9 <sup>a</sup>	1.77	0.029
	5-6	61.3	61.5	61.2	63.5	2.20	NS

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## Piglet growth after weaning is improved when a yeast extract is included in the diet of the lactating sow

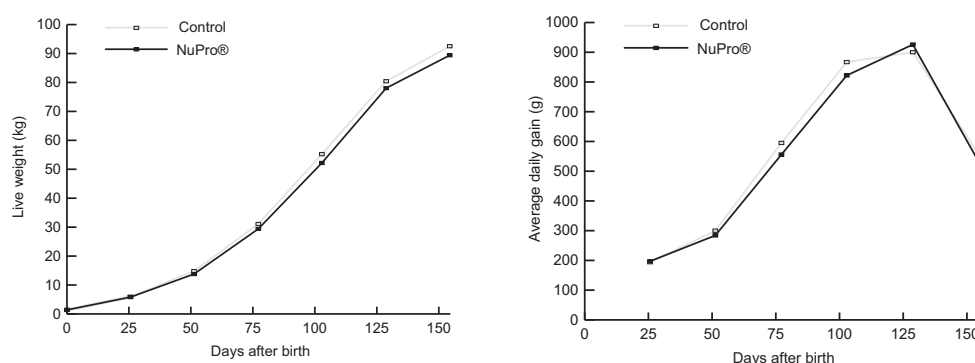
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The yeast extract NuPro<sup>®</sup> is derived from processing the cell contents of the *Saccharomyces cerevisiae* yeast. It is a source of high quality amino acids, glutamic acid and nucleotides (Halbrook *et al.*, 2004). The need for nucleotides is elevated during periods of rapid growth and stress and also in immuno-compromised animals. These factors are often present in piglets during the pre- and post-weaning period. While including NuPro<sup>®</sup> in weaner diets can improve the growth rate of piglets and reduce the incidence of diarrhoea post-weaning, no research has determined whether including NuPro<sup>®</sup> in the diet of lactating sows will have any benefit to the litter post-weaning. It was hypothesized that the post-weaning performance of pigs will be improved when the lactating sow diet contains 2% NuPro<sup>®</sup>.

Sixteen Large White x Landrace x Duroc sows and their progeny were fed either a control diet or a diet containing 2% NuPro<sup>®</sup> during lactation. The diets were formulated to contain the same level of energy and amino acids. Sows received the lactation diet from the time they entered the environmentally controlled farrowing room (about three days pre-farrowing) through to weaning. Piglets were cross-fostered within treatment during the first 48 hours to equalize litter size and average birth weight was the same between treatments. The piglets did not receive creep feed and were moved to a commercial grow-out unit at weaning (21 days of age). Piglet live weight and growth rates were determined at about four-weekly intervals from birth until slaughter. All data were analyzed by analysis of variance.

There were no significant differences in piglet weight at weaning between the control and NuPro<sup>®</sup> diets (5.53 kg and 5.42 kg, respectively) (Figure 1). The lack of a difference in growth rate at weaning may have been because NuPro<sup>®</sup> was included at a relatively low level and only fed during lactation. However, from day 49 to 77 after birth, pigs from sows on the NuPro<sup>®</sup> diet grew faster than those from sows fed the control diet (595 vs. 556 g/day, respectively,  $P < 0.05$ ). The difference in liveweight was maintained until slaughter ( $P < 0.05$ ). This indicates that there is a long term benefit to progeny of including NuPro<sup>®</sup> in the lactating sow diet. Further research is required to determine why the post-weaning performance of pigs was improved even though there was no significant difference in weight at weaning.



**Figure 1.** Increase in liveweight and average daily gain from birth to slaughter of pigs reared by sows fed either a control (□) or NuPro<sup>®</sup> (■) diet during lactation (\*= $P < 0.05$ )

Supported in part by Alltech Biotechnology Pty Ltd.

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## What determines teat position within a litter?

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Teat position may be affected by birthweight and fostering but the impact of these may differ between gilt and sow-reared litters. Piglets grow faster when feeding from anterior teats than posterior teats in sow-reared but not gilt-reared litters. Competition for anterior teats is also greater in sow-reared litters than those litters reared on gilts (Fraser and Thompson, 1986). The experimental hypothesis was that heavier, non-fostered piglets would be found on anterior teats, but only in older parity sow litters.

The study population was 160 gilts, 160 parity 2-5 sows and their litters (3218 piglets). The dams were randomly selected from five farrowing sheds on a commercial farm and randomly allocated to pens, blocked by shed and row, to minimize environmental influence. Individual piglets were identified with numbered eartags. Piglets were fostered once within 36 hours of birth to standardize litter sizes to 10 piglets. The teat position for each piglet was recorded three times during the first two weeks of lactation with the day of recording varying with staff availability. Piglets were weaned at 26 days of age.

An ordinal logistic regression was conducted using Genstat (VSN International Ltd). Teat position was recorded as 1 (first anterior teat pair) to 6+ (last posterior teat pairs). The teat position varied for 53% of piglets for whom an average teat position was calculated. Birthweight, dam parity, gender and fostering (Yes/No) were included as fixed effects in the model. The fitted model  $P(Y \leq k) = \{1 + \exp[-(\hat{\theta}_k + 0.4179 \times BW - 0.391 \times XFOST)]\}^{-1}$  generated the coefficients  $\hat{\theta}_1 = -2.296$ ,  $\hat{\theta}_2 = -1.291$ ,  $\hat{\theta}_3 = -0.502$ ,  $\hat{\theta}_4 = -0.21$  and  $\hat{\theta}_5 = 1.10$ .  $P(Y \leq k)$  was the cumulative probability of a teat position of  $k$  or lower,  $BW$  was the birthweight, and  $XFOST$  was an indicator variable for cross-fostering (yes/no). The probability that piglets would be found on particular teats was the difference between consecutive cumulative probabilities.

Dam parity ( $P=0.966$ ) and gender ( $P=0.705$ ) did not influence piglet teat position and were eliminated from the model. Random effects could not be included in the analysis due to model limitations. The minimal fostering resulted in 6.2% of piglets being fostered. Fostering ( $P=0.002$ ) and birthweight ( $P<0.001$ ) were significant predictors of piglet teat position. As the birthweight of piglets increased, they were more likely to be found on an anterior teat than a posterior teat (Table 1). Fostered piglets were more likely to be found on posterior teats than on anterior teats, after adjusting for birthweight.

**Table 1. The probability that a piglet of a particular birthweight will be found on a particular teat pair; anterior (1) to posterior (6) adjusted for fostering**

Birthweight (kg)	Teat 1	Teat 2	Teat 3	Teat 4	Teat 5	Teat 6
0.6	0.11	0.14	0.18	0.18	0.18	0.21
1.6	0.16	0.18	0.19	0.17	0.15	0.15
2.2	0.20	0.20	0.19	0.15	0.13	0.12
3.0	0.26	0.23	0.19	0.13	0.10	0.09

Our findings indicate that heavier piglets are more likely to be found on anterior teats, regardless of dam parity. It appears that anterior teats are claimed early in the establishment of the teat order as the fostered piglets were more likely to be found on the posterior teats, regardless of birthweight. The growth advantage previously found for piglets with an anterior teat position (Fraser and Jones, 1975) tends to indicate that preferential fostering of heavier birthweight piglets may assist in reducing within-litter weight variation.

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## Particle size and form of the diet influence production and gastric health in growing-finishing pigs

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Under Danish conditions, pelleted diets increase performance in growing-finishing pigs but compromise gastric health compared with coarsely-ground meal-feed (Hansen, 2004). However, in these studies the particle size of the meal-feed was coarser than commercial practice, and consequently it is not known if pelleted feed increases performance compared with meal-feed or if differences were attributable to particle size effects. The aim of this study was to investigate the effect of feed grinding and pelleting on performance and gastric health in growing-finishing pigs.

The experiment comprised a 2x2 factorial design with the factors being grinding (feed ground through a 3.5 mm screen versus feed ground through a 2 mm screen) and form of the diet (pelleted versus meal-feed). All diets were based on wheat, barley and soybean meal and differed only in processing and milling. A total of 456 pigs in 11 randomized complete blocks (10 or 11 pigs per pen) were used from 29±3.1 kg until slaughter at 103±6.1 kg. Lean meat percentage was measured at the slaughter line using a KC21 classification centre. Lesions in the pars oesophagea were scored on a scale from 0-10, 0 being normal and 10 having severe gastric ulceration (Christensen, 1998). The influence of dietary treatment was analyzed univariately in a normal linear model using the GLM procedure in SAS (SAS Inc v.9.13). In addition, stomach scores were analyzed using the GENMOD procedure.

Pelleting significantly improved average daily gain (ADG) as well as feed conversion ratio (FCR) but lowered lean meat percentage and resulted in more lesions in the pars oesophagea (Table 1). Meal-feed ground to pass a 2 mm screen resulted in reduced average daily feed intake (ADFI) and hence a lower ADG. In general, finer grinding increased gastric lesions (P<0.001).

**Table 1. Performance results and gastric health for growing-finishing pigs fed different processed diets**

Grinding								
Particle size, mm (<1/1-2/2-3/>3)	3.5mm screen (47/45/8/0)		2.0mm screen (82/18/0/0)		SEM	P-value		
Form	Pelleted	Meal	Pelleted	Meal		Grinding	Form	Grinding x Form
ADFI <sup>1</sup> , kg/day	2.47 <sup>a</sup>	2.40 <sup>a</sup>	2.42 <sup>a</sup>	2.19 <sup>b</sup>	0.024	-	-	0.004
ADG <sup>2</sup> , g/day	897 <sup>a</sup>	823 <sup>b</sup>	902 <sup>a</sup>	771 <sup>c</sup>	11.1	-	-	0.03
FCR <sup>3</sup> , kg/kg	2.75	2.92	2.67	2.85	0.020	N.S.	< 0.001	N.S.
LMP <sup>4</sup> , %	59.2	59.9	59.3	60.3	0.15	N.S.	0.004	N.S.
Ulcer score	1.7	0.2	3.7	2.4	0.11	< 0.001	< 0.001	N.S.
Ulcer score > 5, %	2.1	1.0	21.9	6.7		< 0.001	< 0.001	N.S.

<sup>1</sup>ADFI: average daily feed intake; <sup>2</sup>ADG: average daily gain; <sup>3</sup>FCR: feed conversion ratio; <sup>4</sup>LMP: lean meat percentage. <sup>abc</sup>: values with different letter subscript are significantly different (P<0.05).

Feeding meal-feed ground to pass a 3.5 mm screen resulted in a poorer ADG and FCR than pelleted feed probably due to reduced digestibility. Finer grinding of meal-feed is no solution as this compromises average daily feed intake (ADFI) and ADG. The best performance is therefore achieved with feed that is pelleted, but the number of pigs with gastric lesions will increase. Further research is needed to identify the optimal particle size distribution in meal-feed to optimize both production indices and gastric ulcerations.

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## Feeding a low protein amino-acid supplemented diet after weaning reduces incidence of post-weaning diarrhoea

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Post-weaning diarrhoea (PWD), a condition associated with proliferation of  $\beta$ -haemolytic strains of *Escherichia coli* in the small and large intestine, often occurs after weaning. Once attached to the small intestinal epithelium, these strains of *E. coli* can disrupt digestive and absorptive functions of the enterocytes by releasing both heat labile toxins (LT) and heat stable toxins (ST; variants STa and STb) that are responsible for hypersecretory diarrhoea (Pluske *et al.*, 2002). Numerous dietary strategies have been attempted to ameliorate the losses associated with PWD. Of these, feeding a lower-protein diet with supplementation of essential amino acids has been suggested because by-products of protein fermentation, such as ammonia and amines, are implicated in the aetiology of the condition (Aumaitre *et al.*, 1995). However feeding a lower-protein diet after weaning is associated with reductions in performance (Nyachoti *et al.*, 2006). In this study, we hypothesized that feeding a low protein diet for a short period of time after weaning would reduce PWD by reducing protein fermentation in the LI.

Seventy-two female pigs (Large White  $\times$  Landrace) aged 21 days and weighing  $5.9 \pm 0.12$  kg (mean  $\pm$  SEM) were randomly allocated to six treatments based on weaning weight. The treatments were: high protein diet (24% CP) for day 14 (HP14); low protein amino-acid supplemented diet (18% CP) for either day 14 (LP14) or day 7 (LP7). Half of the pigs (n=12) per treatment were infected (I) with 3 mL, 8 mL and 8 mL (107 CFU/mL) of *E. coli* (serotype O149; K91; K88) at 72, 96 and 120 hours after arrival, respectively. Diet LP was fortified with lysine, methionine, tryptophan, threonine, isoleucine and valine, based on proposed ideal amino acid patterns. An intermediate diet (20.5% CP) was fed to pigs at the conclusion of each treatment. None of the diets contained antimicrobial compounds. Rectal swabs were scored according to the number of positive streaked sections (0-5) on days zero, five, seven, 10 and 14. Plasma urea nitrogen (PUN) was measured on day seven and 14. Diarrhoea was recorded for the first 14 days. Repeated-measures analysis of variance (SPSS Inc v.14.0, SAS Inc) was used to analyze the results.

Infection increased *E. coli* shedding ( $P < 0.001$ ) and the DI ( $P < 0.001$ ) (Table 1). Piglets fed a low-protein diet both for seven and 14 days after weaning showed lower levels of PUN ( $P < 0.001$ ) and the DI ( $P < 0.001$ ) than piglets fed a high-protein diet. The effects of PL on the decreased PUN and DI were independent of *E. coli* infection. These results support our hypothesis that feeding a lower-protein, amino-acid supplemented diet for a shorter period of time after weaning reduces PWD by reducing the protein load entering the hindgut of the young pig.

**Table 1. Effect of a low protein diet on shedding of  $\beta$ -haemolytic *E. coli*, PUN and DI2.**

Infection	Non- infected			Infected			P-value			
	HP14	LP14	LP7	HP14	LP14	LP7	SEM	PL <sup>1</sup>	I	PL $\times$ I
<i>E. coli</i> score	0.3	0.1	0.1	0.6	0.6	0.5	0.11	0.373	<0.001	0.490
PUN	5.2 <sup>a</sup>	2.1 <sup>c</sup>	3.4 <sup>b</sup>	5.6	2.2	3.9	0.59	<0.001	0.467	0.944
DI2	19.8 <sup>a</sup>	8.5 <sup>c</sup>	9.6 <sup>b</sup>	44.2 <sup>a</sup>	31.6 <sup>b</sup>	21.4 <sup>c</sup>	3.54	<0.001	<0.001	0.154

<sup>1</sup>Protein level; <sup>2</sup>Diarrhoea index: Proportion of days with diarrhoea with respect to total days (14 days) on trial.

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## Manipulation of *ad libitum* feed intake in sows

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Fed individually, the fibre sources readily available in Australia have limited potential to facilitate *ad libitum* feeding of sows although there is some potential for combinations of fibre sources to be used (R.J. van Barneveld, pers. comm.). In this experiment we determined whether fibre sources combined with other ingredients known to influence feed intake could be used to facilitate *ad libitum* feeding of gestating sows.

Multiparous sows selected on day 56 of gestation were housed in individual pens and fed *ad libitum* for 14 days. All diets were formulated to contain 12 MJ digestible energy (DE)/kg and 0.4 g available lysine/MJ DE and were offered in gas bottle feeders. Each treatment was replicated four times using different groups of sows. Wheat straw was added as a bulking agent (0 vs. 15%) and guar gum as a source of soluble non-starch polysaccharide with high water holding capacity (0 vs. 0.5%). Also included was *Lupinus albus* (0 vs. 6.5%). Salt levels were manipulated (0.2% sodium chloride vs. 0.4% sodium chloride+0.3% potassium chloride). These ingredients were included alone or in combination in 16 diets to assess their effect on *ad libitum* feed intake, weight gain and backfat gain in sows (Table 1).

**Table 1. Mean *ad libitum* feed intake (kg/day) of gestating sows fed fibre and other ingredients to manipulate feed intake either alone or in combination**

Treatment	Mean feed intake (kg/day)	Weight gain (kg/day)	Backfat gain (mm)
Control	7.77 <sup>a</sup>	2.10	2.7
Salt	7.10 <sup>a</sup>	1.75	3.0
Salt+lupins+wheat straw + guar gum	6.09 <sup>b</sup>	1.50	2.2
Salt+lupins+wheat straw	5.91 <sup>b</sup>	1.43	4.1
Salt+lupins+guar gum	6.62 <sup>a</sup>	1.46	1.8
Salt+lupins	8.29 <sup>a</sup>	2.02	2.3
Salt+wheat straw+guar gum	5.94 <sup>b</sup>	1.93	2.6
Salt+wheat straw	6.85 <sup>a</sup>	1.86	2.1
Salt+guar gum	6.92 <sup>a</sup>	1.70	2.9
Lupins	7.05 <sup>a</sup>	1.80	3.6
Lupins+wheat straw+guar gum	5.40 <sup>b</sup>	1.29	2.3
Lupins+wheat straw	5.93 <sup>b</sup>	1.68	1.6
Lupins+guar gum	6.32 <sup>a</sup>	1.61	2.7
Wheat straw+guar gum	6.07 <sup>b</sup>	1.79	2.1
Wheat straw	5.71 <sup>b</sup>	1.36	3.0
Guar gum	5.68 <sup>b</sup>	1.43	2.8
Standard error of means	0.479	0.286	0.07

<sup>a,b</sup>Means in a column with different superscripts differ significantly (P<0.05).

Significant differences (P<0.05) in feed intake were detected across the feed treatments with wheat straw and guar gum the most effective at reducing *ad libitum* intake. However, no significant change in weight gain or backfat gain was detected across the treatments. Despite a structured approach to the control of *ad libitum* feed intake in sows, no treatment was effective at limiting intake to an acceptable level during lactation with apparent DE intakes at least 1.5-2.0 times higher than the desired level of 30-35 MJ DE per day. There appears to be limited options for the *ad libitum* feeding of group-housed sows in Australian production systems.

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## Particle size of *Lupinus angustifolius* is associated with energy and protein digestibility in growing pigs

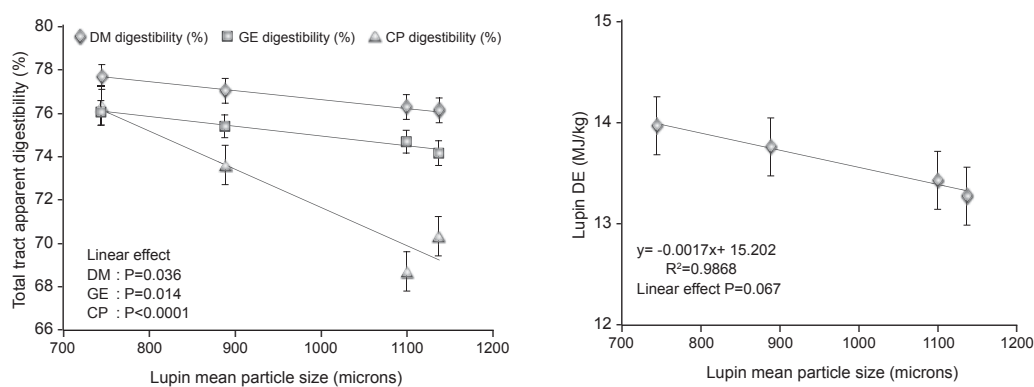
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Lupins contain high levels of non-starch polysaccharides (NSP) both in hulls and kernels, suggesting complex associations between digestible nutrients and these structural NSP in kernels. Therefore, the degree of mechanical grinding could influence digestibility of dietary components. Hammer-milling lupins through a finer screen (3 vs. 5 mm) increased total tract digestibility (TTD) of energy by 5% and digestible energy (DE) content by 1 MJ/kg (Wigan *et al.*, 1995). However, associations between TTD of nutrients and the lupin particle size have not been investigated in detail. The aim of this study was to establish relationships between particle size of lupins and TTAD of dietary components in growing pigs.

Sixty-three (Large White x Landrace), initial weight 63.5 kg  $\pm$  0.95) individually-housed male pigs were randomly allocated to a 2x4 factorial experiment with the respective factors being lupin variety (Mandelup and Tanjil) and particle size of lupins (744, 888, 1099 and 1136  $\mu$ m). A wheat control diet was used to determine the DE contents of the basal diet. All diets except the wheat control diet contained 574 g wheat, 350 g test lupins, titanium dioxide as a digestibility marker and other additives. Canola oil (930 g/kg) was used to prevent possible segregation. Wheat was ground using the same screen (6 mm) across treatments but lupins were hammer-milled through different sized screens (2, 4, 6 and 8 mm) to achieve the desired particle sizes. Pigs were fed their respective diets at three times maintenance [ $3 \times (0.458 \text{ MJ DE} \times \text{body weight}^{0.75}) / \text{diet DE}$ ] for 10 days and faecal samples were collected over the final three consecutive days to determine digestibility of dietary components. The GLM procedure of SPSS (SPSS Inc) was used for statistical evaluation. There were no variety effect on digestibility and hence data were pooled for particle size effects.

Increasing particle size of lupins from a mean particle size of 744  $\mu$ m to 1136  $\mu$ m linearly decreased total tract apparent digestibility of dry matter (DM), gross energy (GE) and crude protein (CP) of the test diets, and the DE content of lupins (Figure 1). However the extent of decline was greater for CP digestibility than other dietary components such as DM and GE. The results suggest that fine grinding of lupins is essential for adequate utilization of CP in pigs.



**Figure 1.** Effects of lupin particle size on total tract apparent digestibilities of dietary components and digestible energy contents of lupins (*Lupinus angustifolius*)

Study funded by the WA Agricultural Produce Commission: Pork Producers' Committee.

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## Yeast extract reduces histological indices of inflammation in the small intestine of weaned piglets

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Feed additives are sometimes used in diets after weaning to modulate the structure and function of the gastrointestinal tract. Bio-Mos® (Alltech Biotechnology Pty Ltd), a mannan oligosaccharide derived from the cell wall of *Saccharomyces cerevisiae*, has been shown to influence positively the performance of weaning pigs (Miguel *et al.*, 2004). However, the precise mechanism(s) for these effects has not been fully elucidated. This study tested whether Bio-Mos® included in diets for sows in gestation and lactation and then in a post-weaning diet would alter indices of inflammation in the small intestine of young pigs. This was presumed on the basis that Bio-Mos® has been suggested to influence pathogen colonization and/or localized immunity (Davis *et al.*, 2004).

Samples of jejunum and ileum of piglets were collected into phosphate-buffered formalin at weaning or 14 days after weaning from either: 1) sows fed Bio-Mos® throughout pregnancy and lactation (1 g/kg) and then piglets fed Bio-Mos® (3 g/kg) after weaning or 2) sows not fed Bio-Mos® throughout pregnancy and lactation and piglets not fed Bio-Mos® after weaning. Tissue samples were subsequently fixed in haematoxylin and eosin, and then processed using established histological procedures at Murdoch University for the subsequent enumeration of goblet cells, granulated mononuclear inflammatory cells (ICs) and non-granulated mononuclear ICs per 500 enterocytes. The statistical model included the main effects ( $\pm$ Bio-Mos®, time of euthanasia) and the interactions using JMP (SAS Inc v.6.1).

There were no differences ( $P>0.05$ ) in goblet cell numbers (Table 1). Pigs from sows fed Bio-Mos® had fewer granulytic ( $P=0.009$ ) and non-granulytic ( $P<0.001$ ) ICs than pigs not exposed to Bio-Mos®. Newly weaned pigs had fewer granulytic ( $P<0.001$ ) and non-granulytic ( $P<0.001$ ) ICs than pigs killed 14 days after weaning. These data suggest that the small intestine of piglets derived from sows fed Bio-Mos® in gestation and lactation, and then fed a diet containing Bio-Mos® for 14 days after weaning, was less challenged. The reduction in the number of inflammatory cells suggests a direct effect of Bio-Mos® on the small intestine, however the mechanism(s) whereby this effect occurred could not be ascertained from this work.

**Table 1. Least-squares main effect means for the number of goblet cells, granulytic inflammatory cells (ICs) and non-granulated (NG) inflammatory cells (ICs) (all expressed per 500 enterocytes)**

Cell type	Bio-Mos® (B)		Time (T)		RMSE <sup>1</sup>	Level of Significance		
	+	-	Wean <sup>3</sup>	After <sup>3</sup>		B	T	BxT
Goblet	54.2	53.3	58.7	48.8	30.37	NS	NS	NS
Granulated ICs	26.5	36.0	19.9	42.5	12.84	0.009	<0.001	NS
NG ICs	46.2	63.8	41.8	68.1	17.24	<0.001	<0.001	NS

<sup>1</sup>RMSE: root mean square error; <sup>2</sup>NS:  $P>0.1$ ; <sup>3</sup>Wean: pigs killed at weaning; After: pigs euthanased 14 days after weaning.

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## Performance of weaner pigs offered 32 different grain diets

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The nutritional quality of cereal grains used in pig production in Australia vary by up to 2 MJ/kg in digestible energy (DE) (Kopinski, 1997; Black, 2001). Cadogan *et al.* (1999) found highly significant differences in the feed intakes of young male pigs fed different wheat varieties. However there is little information about the variation in voluntary feed intake and growth of pigs fed other grain species and cultivars. In this experiment we evaluated the variation in pig growth performance fed 32 different diets from four different grain types (9 wheat, 7 barley, 11 sorghum and 5 triticale).

Complete weaner diets containing 65% of the test grains and highly digestible raw ingredients were prepared. The diets were formulated to contain, on average, 14 MJ DE/kg and an available lysine content of 0.85 g/MJ DE. The 32 grains were assigned to pigs using a random block design consisting of 640 (32 grains x 20 pigs) individual Large White weaner pigs (633 males and seven females) over 10 runs/block. The pigs weighed 6.5-8.5 kg and were placed in individual crates for 28 days. After a pre-treatment of five days on a common nursery diet, pigs were fed the test diets and water *ad libitum* for a further 21 days. Body weight and feed intake were measured on days 0, 14 and 21 after the initial five-day period. The data were analyzed using linear mixed model technology. The model included fixed terms for sex, batch, grain type and grain cultivar and random terms for run and block.

There were significant sex effects for total grain intake and daily intake (Table 1). The between- and within-grain type effects were significant for total grain intake, daily intake, feed conversion ratio (FCR) and daily gain. The differences between grain types were influenced by sorghum, which reduced pig performance compared to wheat, barley and triticale. In summary, there is evidence to suggest pig performance is influenced by grain type.

**Table 1. P-values of significant effects on intake and performance of weaner pigs**

Source	Total grain intake 0-21 d	Daily intake (kg/d) 14-21 d	FCR 14-21 d	Daily gain (kg/d) 14-21 d
Sex	0.003	0.015	NS	NS
Between grain types	<0.001	0.010	0.045	<0.001
Within grain types	0.033	0.008	0.019	0.010

Supported by the Qld Department of Primary Industries and Fisheries. The support of QAF Meat Industries Pty Ltd staff is acknowledged.

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## The relationships between sow water intake during gestation on reproductive and lactation performance

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Water intake may be critical during gestation as the fetuses are growing in a fluid environment and nutrients and excretory products are transported via blood and water. There is enormous variation in water intake between gestating sows (0.5 to 42 L/sow/day) (Cargill *et al.*, 1999), and it is unknown whether this variation in intake is responsible for variation in reproductive and lactation performance of sows. Therefore, the aim of this experiment was to examine relationships between sow daily water intake during gestation on reproduction and lactation performance and urinary tract health.

Two hundred and twenty Large White x Landrace sows were housed in gestation stalls from the day of mating until day 108 of gestation. The experiment was carried out in summer and winter. The sows were fed 2.5 kg/day of pelleted feed. Water use of individual sows was recorded via water meters supplying a nipple in an individual trough at a rate of 3.5 L/minute. Urine samples were collected from 70% of the sows in week one, eight and 15 of gestation. Samples were collected during feeding in the morning. The samples were tested for, bilirubin, blood, Ph, and leucocytes. The incidence of blood in the urine, high pH values and leucocytes can indicate urinary tract infections. Sows were provided with *ad libitum* feed once they had farrowed and the litters of piglets were weighed at birth and prior to weaning (average 23 days). The data for each experimental period (summer and winter) were analyzed by Pearson's correlation two-tailed analysis.

The average daily water intake was significantly higher ( $P < 0.5$ ) in summer than winter; 20 vs. 17.6 L, respectively (range of 4.5-41.3 L) (Table 1). There were no significant relationships ( $P > 0.05$ ) between daily water intake in gestation on maintenance of pregnancy, urinary health, litter birth weight, litter size, number of stillborn piglets, daily lactation feed intake and weaning weight of pigs. The sows in the current experiment were drinking enough to maintain urinary tract health and their water intake during gestation did not affect reproductive or lactation performance.

**Table 1. Pearson's correlation coefficients (P-value) between average daily water intake and farrowing rate, litter birth weight, litter size, number of stillborn piglets, daily lactation feed intake and weaning weight of piglets**

	Daily water intake (Summer)	Daily water intake (Winter)
Successful gestation (0=no; 1=yes)	-0.188 (0.06)	-0.134 (0.172)
Incidence of urinary tract infection (0=no; 1=yes)	0.073 (0.471)	0.076 (0.443)
Litter birth weight	0.046 (0.689)	-0.001 (0.995)
Total litter size	0.073 (0.525)	0.021 (0.846)
Number of stillborn piglets	0.125 (0.274)	-0.154 (0.377)
Daily lactation feed intake	0.205 (0.137)	0.215 (0.076)
Weaning weight of piglets	-0.046 (0.744)	-0.044 (0.736)

Financial support from Australian Pork Limited and technical assistance from management and staff at QAF Meat Industries Pty Ltd is acknowledged.

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## The interaction of organic acids with medium-chain fatty acids on growth performance of piglets raised under sub-optimal management conditions

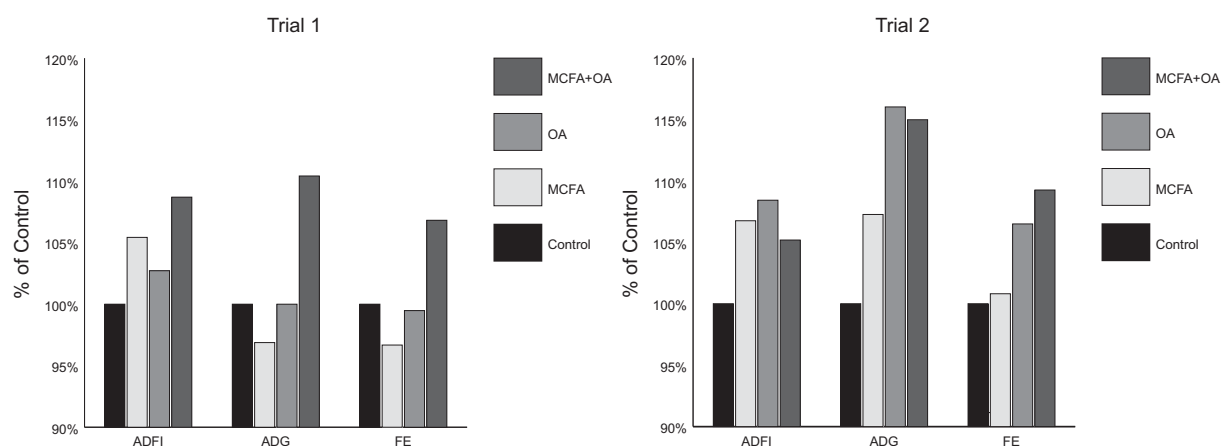
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Organic acids (OA) inhibit gram-negative bacteria by reducing pH and via direct bacteriostatic action of non-dissociated acid molecules (Partanen and Mroz, 1999). Recently medium-chain fatty acids (MCFA) have been shown to have strong bactericidal properties toward gram-positive bacteria (Dierick *et al.*, 2002). In this study, we hypothesized that MCFA and OA used together would improve antibacterial strength in a synergistic way and lead to improved growth performance of weaned piglets with compromised gut health.

Piglet feed was dosed with MCFA and/or commercial OA blends during the first two weeks after weaning to evaluate the synergistic antimicrobial effect of MCFA and OA on piglet production parameters. Trial 1 was performed on a farm with known poor husbandry conditions. Pens of nine piglets each (weaning weight  $6.5 \pm 0.08$  kg) were assigned to one of four treatments (30 replicates). Tested levels were 0.2% MCFA and 0.5% OA blend. Trial 2 was performed at the Nutreco Swine Research Centre where suboptimal conditions were created by not cleaning the room before the start of the trial and introducing sow manure, lowering ambient temperature settings and increasing dust concentration. Each treatment consisted of 18 pens with three piglets each (weaning weight  $9.1 \pm 1.32$  kg). Tested levels were 0.1% MCFA and 0.6% OA blend. Data were analyzed using the GLM procedure of SAS (SAS Inc) with batch (only Trial 1), gender and diet as independent variables.

In Trial 1, average daily feed intake (ADFI) tended to be improved in the MCFA and MCFA+OA treatments ( $P=0.082$ ) (Figure 1). In Trial 2, feed efficiency (FE) was improved when OA was used as the main effect ( $P=0.044$ ).



**Figure 1.** Performance parameters of piglets 1-14 days after weaning, in response to medium-chain fatty acids (MCFA) and/or organic acids in the feed (OA; control = 100%)

These *in vivo* data could not confirm the interactions between OA and MCFA previously observed *in vitro*. Still, the combination of OA and MCFA numerically offered the best weight gain (Trial 1) and FE (Trial 1 and 2) and warrants further investigation.

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## Benzoic acid as an alternative to prophylactic antibiotics in swine diets

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Consumer awareness of food safety issues is becoming increasingly important and has resulted in the ban of prophylactic antibiotic use in animal feeds in the European Union (EU). A similar ban has not been implemented in Asia Pacific, but interest in alternatives to antibiotic growth promoters has increased for several reasons: 1) export of animal-origin food products to the EU requires that the animals are grown without the use of dietary antibiotics and 2) some countries in the region are restricting the use of antibiotic growth promoters and producers are preparing for a possible full ban in the future.

The current study was designed to investigate the efficacy of VevoVital<sup>®</sup> (Benzoic acid) as a replacement for prophylactic antibiotics in piglet diets. VevoVital<sup>®</sup> is a commonly available dietary acidifier that is known to have strong antimicrobial effects in the stomach and small intestine of piglets (Knarreborg *et al.*, 2002). It was hypothesized that these antimicrobial properties would allow VevoVital<sup>®</sup> to be a substitute for dietary antibiotics.

The trial was done using two groups of 40 piglets (10 replicates, four pigs/replicate). The piglets were a cross between Landrace and Large White from the sows and Duroc and Pietrain from the boars. The treatment diets fed to the piglets contained either a standard antibiotic treatment in the control group (Table 1), or VevoVital<sup>®</sup> at 5 kg/MT with no antibiotics in the treatment group (Table 2).

**Table 1. Antibiotic treatment provided in control diet**

Diet type	Antibiotic treatment	Duration of treatment
Pre-starter	Tiamulin and trimethoprim sulfadiazine	30 days
Starter I	Florfenicol	15 days
Starter II	Norfloxacin and tylosin	15 days

**Table 2. Daily weight gain, feed consumption and feed efficiency in piglets fed either a control diet (with antibiotics) or a VevoVital<sup>®</sup> diet (without antibiotics)**

	Pre-starter (41-70 days of age)		Starter (71-100 days of age)	
	Control	VevoVital <sup>®</sup>	Control	VevoVital <sup>®</sup>
Weight Gain (g/day)	595.7	603.3	657.3	708.0
Feed Consumption (g/day)	963.3	955.7	1691.0	1646.7
Feed Efficiency	1.62	1.58	2.57	2.33

There were no significant differences between the control and treatment groups when statistical analysis was done using analysis of variance and significance was considered at  $P > 0.05$ . Numerically the group treated with VevoVital<sup>®</sup> tended to have a slightly higher weight gain and slightly lower feed consumption and feed efficiency. The results of this study suggest that VevoVital<sup>®</sup> can be used as a substitute for prophylactic antibiotics without affecting growth parameters in pre-starter and starter piglets.

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## Quality control results for artificial insemination boar semen

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Artificial insemination (AI) is the major driver of genetic improvement in the pig industry. However, the genetic potential of pig AI is only fully realized when sperm fertility is optimal. The main vehicle for the transmission of superior genetics is using extended chilled semen where success depends on insemination of adequate numbers of effective sperm. In turn, this depends upon initial semen assessment and processing as well as factors involved with transport, heat detection and insemination. King (2000) identified several constraints to the Australian pig AI industry, including superficial and unsophisticated assessment of quantitative and qualitative semen quality. In this preliminary study, pig semen submitted from 259 boar studs located in four states for routine monitoring, was subjected to stringent evaluation procedures with the objective of establishing industry benchmarks.

Routine monitoring of semen submitted by boar studs commenced in June 2006. Boar semen is usually submitted in one of two forms: 1) as insemination units in chilled client-ready packs or 2) as samples 'fixed' in phosphate buffered saline (PBS). Upon arrival at the laboratory, chilled semen was assessed for: temperature, motility (HTM-IVOS v.12; CASA), volume (adjusted weight), concentration (haemocytometer), morphology and percent intact acrosomes (PIA). 'Fixed' semen was evaluated for sperm morphology and PIA. For all samples, a total of 100 sperm were counted with differential interference phase contrast (DIC) microscopy at 1100X using established semen evaluation categories (Chenoweth *et al.*, 1994). A total of 250 samples representing submissions from six boar studs from four states over a period of 11 months were subjected to routine statistical analysis using Minitab (Minitab Inc v.12).

Means, standard deviations and minimum/maximum values were calculated for arrival temperature, sperm motility, concentration, percent normal sperm and percent intact acrosomes (Table 1).

**Table 1. Statistical analyses of survey data**

Semen Trait	Mean	SD	Min	Max
Arrival temp. (°C)	19.77	2.23	15	23.1
Sperm motility (%)	69.98	20.21	10	99
Sperm concentration (10 <sup>6</sup> /ml)	28.6	14.54	11.5	92
Normal sperm (%)	75.88	14.29	26	96
PIA* (%)	93.51	4.15	70	100

\*PIA - Percent intact acrosomes

Chilled boar semen is usually delivered as a dose of about 80-90 ml containing 2.5-3 billion 'normal' sperm, which necessitates sperm concentrations of about 40x10<sup>6</sup>/ml and good sperm motility (>70%) and morphology (>70%) (Althouse, 1995). In this study, sperm concentration, motility and morphology levels were considered to be adequate although variations in these traits were higher than necessary. In contrast, PIA levels (93.51±4.15%) were generally good and arrival temperatures (19.77±2.23°C) were within recommended guidelines (15-20°C). Further analyses are being done to identify the major causes of variations in both quantitative and qualitative semen assessments with the expectation that these can be reduced with improved procedures and monitoring.

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## Use of rapid visco analyser for feed grain evaluation

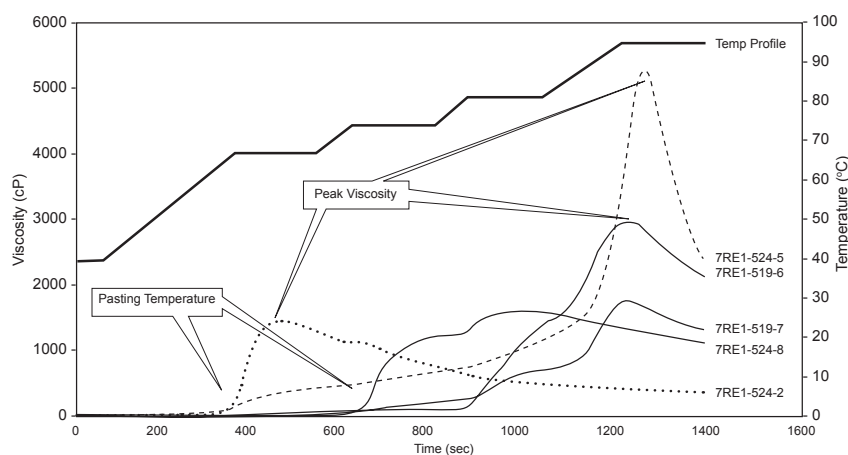
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The Rapid Visco™ Analyser (RVA) is commonly used to predict the pasting behaviour of baking-flours to enhance their blending and baking properties. RVA is likely to have application in stock feed manufacture as the pasting characteristics of individual grains directly affects the processing and handling properties of individual ration components and their feeding value. A better understanding of the pasting behaviour of stock feed grains would help to match feed preparation methods to the characteristics of individual feed grains, to the advantage of both stock feed users and producers.

Wheat, barley and sorghum samples were obtained from the Premium Grains for Livestock feed database. Samples were ground through a fine laboratory hammermill (0.8 mm screen) without sieving. Sub-samples (4 g, about 11.5% moisture) were mixed with 27 g of distilled water and tested (Newport Scientific RVA Model 4) using an extended temperature profile to provide greater separation in pasting curves (Blakeney and Booth, 2000).

Pasting curves for a selection of wheat, sorghum and barley grains showed clear differences within and between grain types in both pasting point and peak viscosity (Figure 1). Both the waxy wheat (7RE1-524-2) and barley (7RE1-524-5) gelatinized at a lower temperature (65°C). Peak viscosity for sample 7RE1-524-2 occurred sooner and at a lower temperature than for either sorghum or barley samples. A common pasting temperature (75°C) was exhibited by all sorghum samples and exceeded that of barley and wheat. Variations in the pasting curves suggest that the RVA technique could be extended to the evaluation of grains of interest to livestock feed manufacturers and pig producers.



**Figure 1.** Pasting curves for wheat (.....), barley (-----) and sorghum (—) grains obtained by Rapid Visco™ Analysis using the temperature profile (—) displayed

Supported by a Pig R&D Corporation Industry Placement Award.

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## Tuna fishmeal supplementation to pigs does not influence lipid oxidation of fresh pork

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Howe *et al.* (2002) demonstrated that including 15% PorcOmega™, a stabilized tuna fishmeal formulation, in the pig diets resulted in a substantial increase in long chain n-3 polyunsaturated fatty acids in pork products. However, omega-3 enriched pork may potentially be more prone to lipid oxidation and quality deterioration due to the increased content of unsaturated fatty acids. This study was done to determine whether supplementing 15% PorcOmega™ to pigs for 42 days before slaughter resulted in increased rates of lipid oxidation in pork loin steaks, mince and sausages following storage at 4°C for five days.

Twenty-four gilts, 14-15 weeks of age, were randomly selected, weighed and allocated one of two dietary treatments (isocaloric with similar protein) fed *ad libitum* for 42 days before slaughter. Control pigs were fed a standard wheat/barley based diet and the treatment diet was supplemented with 15% PorcOmega™ (Bartlett Grain Pty Ltd). Before slaughter, all pigs were individually tattooed for ease of identification in the boning room. Two days after slaughter, all pork samples were trimmed of visible fat and processed into three different cuts (steak, mince and sausage) and packaged into bags containing ~200 gram servings (day 0). Samples from each of the three cuts were allocated one of two storage treatments: 1) samples sealed and placed in -80°C degree freezer on day 0; or 2) samples stored at 4°C for five days (to simulate retail shelf life for fresh meat), then sealed and frozen at -80°C. Samples were analyzed for lipid oxidation using thiobarbituric acid reactive substances (TBARS) test (based on Witte *et al.*, 1970). Results were adjusted for 75% water in fresh meat and expressed as mg of malonaldehyde (MDA) equivalent per kg fresh muscle and analyzed using analysis of variance.

Including 15% PorcOmega™ into finisher pig diets did not influence ( $P>0.05$ ) lipid oxidation, as measured by TBARS, for pork loin steaks, mince or sausages (Table 1). Refrigerated storage of mince and steak for five days after boning resulted in increased TBARS values compared with those at day 0. The lower TBARS values obtained for sausages after five days of refrigerated storage indicates that pre-mix ingredients added to sausages during manufacture impeded further lipid oxidation. In conclusion, these results indicate that it is feasible to supply omega-3 enriched fresh pork products to a retail level of quality comparable, in terms of lipid oxidation, to that of current pork products.

**Table 1. Effect of dietary treatment (D; control or PorcOmega™ enriched) and storage time (S; 0 or 5 days after boning) on TBARS values of pork sausage, mince and loin steak**

Diet treatment	Control		PorcOmega™ enriched		SED (D*S)	P values		
	Day 0	Day 5	Day 0	Day 5		D	S	D*S
Storage time	Day 0	Day 5	Day 0	Day 5	(D*S)	D	S	D*S
Sausage	0.191	0.244	0.235	0.312	0.0488	0.069	0.11	0.94
Mince	0.172	0.835	0.134	0.941	0.1796	0.79	<0.001	0.58
Loin steak	0.120	1.397	0.074	1.412	0.1054	0.84	<0.001	0.68

Supported by an ARC Linkage grant in partnership with Bartlett Grain and Australian Pork Limited.

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## Evaluation of faba beans as an alternative legume for pigs

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The availability of faba beans in southern Australia is increasing but information on the performance of pigs fed the current varieties of faba beans is scarce. There is a significant negative correlation between *in vitro* digestibility of faba beans and the tannin content of the seed. The tannins are mostly located in the hulls and de-hulling is reported to increase protein digestibility and feed conversion efficiency. In this experiment we used *Vicia faba* minor with a protein content of 26% and a fat content of 4%. The hypothesis tested was that including up to 20% faba bean into the weaner diet would not impact on weaner performance.

Eighty Landrace x Large White male weaner pigs with a weaning weight of 7.8kg +/-1.5kg (mean±SD) at about 25 days of age were housed in individual cages for a five-day acclimatization period before being randomly assigned to one of four treatments stratified on start weight. Treatments were assigned as: 0% faba beans (0%); 10% faba beans (10%); 15% faba beans (15%) and 20% faba beans (20%). Diets were formulated to a standard specification with 14.8 MJ DE /kg and 0.89 g available lysine to digestible energy. Faba beans replaced a mixture of soybean meal and wheat in the diets. The dietary treatments were fed for 21 days and average daily gain (ADG), average daily intake (ADI) and feed conversion ratio (FCR) recorded at 7, 14 and 21 days. Performance data were analyzed using general linear model multi-variant approach, analysis of variance (SPSS v14).

**Table 1. Growth performance of weaner pigs fed faba beans at a range of dietary inclusion levels for 21 days**

Dietary inclusion level	Start weight (kg)	Final weight (kg)	Daily gain (kg/day)	Average daily intake (kg/day)	FCR (feed:wt. gain)
0%	8.01	16.12	0.39	0.45	1.16a
10%	8.01	16.11	0.39	0.45	1.17a
15%	8.01	16.25	0.39	0.44	1.14a
20%	8.01	16.01	0.38	0.47	1.24b
P=	1.00	0.99	0.99	0.82	0.03
Linear	0.99	0.95	0.95	0.58	0.15
Quadratic	1.00	0.98	0.97	0.67	0.07
SEM	0.10	0.23	0.01	0.01	0.01

<sup>abc</sup> Mean values with different superscripts within columns are significant (P<0.05).

Feed conversion efficiency was significantly reduced (P=0.032) with a trend for a quadratic response (P=0.067) when faba beans were included in the diet at 20%. We therefore reject the hypothesis and suggest that the current varieties of faba beans available in Australia will impact on pig performance. However, the data from this experiment suggest that faba beans can be included into the diets at 15% without negatively affecting growth performance. Further analysis on tannin levels may be warranted if availability of faba beans increases.

## Ingestion of nutrients from pasture by grazing European wild boar

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Meat production from the European wild boar (*Sus scrofa* L.) is increasing in Chile, using a grazing system supplemented by a concentrated diet. Knowing the amount of nutrients consumed by these animals through pasture would enable higher precision in the formulation of the concentrated diet. The objective of this study was to determine the consumption of dry matter (DM), gross energy (GE) and crude protein (CP) from pasture by European wild boar in a pastoral system.

Pastures containing *Lolium perenne* or *Plantago lanceolata* were established. Each pasture was divided by fencing into 19 strips (S1 to S19), with each strip divided into three areas of 1.4x6.3 m. Twelve purebred European wild boar of 18.8±0.8 kg (mean±SEM) were randomly grouped into pairs. On each day of the 19-day study, a pair of animals was placed into each of the three areas of one pasture strip of each pasture species from 0830 hours until 1630 hours, after which the animals entered a barn and had free access to a commercial diet for 60 minutes. On day one the animals grazed S1; on day two S2 and so on, with water always available. No pasture samples were collected during days 1-3 (period of adjustment). On days 4-19, the DM content was determined of pasture samples taken pre- and post-grazing from each grazed area, cut to ground level (area 0.25 m<sup>2</sup>). The DM content of the entire grazed area was calculated and the DM consumption per animal was determined (difference between the DM availability pre- and post-grazing, divided by two). Additional pasture samples from each strip cut to ground level (area 0.04 m<sup>2</sup>) were analyzed for DM, GE and CP to calculate the nutrient consumption from pasture. To increase the range of pasture intakes, the study was conducted in spring (high quality pasture) and again in summer (low quality pasture). Nutrient consumptions were compared between pasture species and seasons using analysis of variance. A correlation analysis was done between DM availability and consumption.

The consumption of nutrients from pasture by European Wild Boar was notably variable, with a higher consumption in spring than summer (Table 1). There was a significant correlation ( $r=0.59$ ,  $P<0.0001$ ) between DM availability and consumption. Assuming a similar energy and protein digestibility of *L. perenne* to that in the pig (energy digestibility 0.51, CP digestibility 0.60) (Lindberg and Andersson, 1998), it is estimated that the animals satisfied up to 30% of their DE and 22% of their CP requirements through pasture.

**Table 1. Mean daily consumption (+SEM)<sup>1</sup> of dry matter (DM), crude protein (CP) and gross energy (GE) from pasture by European wild boar during spring and summer**

Season	Pasture species	DM (g)	GE (Mcal)	CP (g)
Spring	<i>Lolium perenne</i>	418±72a	1.926±0.311a	95.4±14.2a
	<i>Plantago lanceolata</i>	550±86a	2.481±0.359a	115.4±14.8a
Summer	<i>Lolium perenne</i>	210±38b	0.962±0.167b	58.0±8.5b
	<i>Plantago lanceolata</i>	226±45b	1.055±0.198b	41.1±8.4b

<sup>1</sup>Means followed by different letter in a column were significantly different ( $P<0.05$ ),  $n=48$ .

This work was supported by the Chilean National Fund for Science and Technology.

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## Culture-independent detection of zoonotic bacterial pathogens in pig faeces

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Pigs are natural reservoirs of several zoonotic bacterial pathogens such as *Staphylococcus aureus* and methicillin resistant *S. aureus* (MRSA), *Clostridium difficile*, *Escherichia coli* O157:H7 and numerous *Salmonella* serovars including *Typhimurium*. As residents in the gastrointestinal tract, these pathogens can gain entry to the food chain by way of faecal contamination of pig meat and hide during abattoir processing.

Hazard Analysis and Critical Control Point protocols have been put in place to reduce the risk of microbial contamination in the food chain from farm to fork. Despite this, detection of food-borne bacterial pathogens (FBPs) remains imperative as they pose critical health risks and cause substantial economic losses. Current methods for the detection and identification of FBPs rely greatly on culture-dependent protocols followed by pathogen identification using standard microbiological methods. These methods are laborious, expensive and selective for only single pathogens.

To achieve culture-independent detection of more than one FBP, we propose the development of a multiplex polymerase chain reaction (PCR) assay to test the hypothesis that multiplex PCR is a feasible approach for the specific and rapid detection of *S. aureus*, MRSA, *C. difficile*, *E. coli* O157:H7 and *Salmonella* serovars. This approach is based on the ability of the technology to detect the unique carriage of several virulence genes by each pathogen. Fourteen such genes were selected and incorporated into multiplex PCR assays, including the toxin A and B (*tcdA* and *tcdB*) genes found in *C. difficile* and the methicillin resistance (*mecA*) gene carried by *S. aureus*.

However, a prerequisite to multiplex PCR is the validation of individual primer sets and optimization of the specific PCR conditions for each gene via uniplex assays. To achieve this, DNA was extracted from several different isolates of each of the four FBPs. The assay conditions were optimized, while the 14 primer pairs were validated and cross-checked against the alternate three FBPs to ensure specificity. This effectively led to the amplification of the target genes.

The results show that the genes associated with each pathogen are detectable by uniplex PCR and do not occur non-specifically (Table 1). The achievement of these uniplex assays will pave the way for future development of the multiplex PCR assay. This multiplex will be subsequently applied to the screening of meat and faecal samples obtained from pigs at an abattoir to evaluate the prevalence of each of the four targeted pathogens.

**Table 1. Positive (+) and negative (–) carriage of genes specific to four species of food-borne human pathogenic bacteria commonly found to contaminate pork**

		Species Specific Genes													
		<i>S. aureus</i>			<i>C. difficile</i>					O157:H7				<i>Salmonella</i> spp.	
Organism	No. Isolates	<i>mecA</i>	<i>spa</i>	<i>coa</i>	<i>tpi</i>	<i>cdtA</i>	<i>cdtB</i>	<i>tcdA</i>	<i>tcdB</i>	<i>uspA</i>	<i>rfbE</i>	<i>uidA</i>	<i>flicH7</i>	<i>invA</i>	<i>fimY</i>
<i>S. aureus</i>	50	+	+	-	-	-	-	-	-	-	-	-	-	-	-
<i>C. difficile</i>	10	-	-	-	+	+	+	+	+	-	-	-	-	-	-
<i>E. coli</i> o157:H7	30	-	-	-	-	-	-	-	-	+	+	+	+	-	-
<i>Salmonella</i> spp.	30	-	-	-	-	-	-	-	-	-	-	-	-	+	+

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## Antimicrobial susceptibility of recent Australian isolates of *Brachyspira hyodysenteriae*

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Swine dysentery (SD) is an important disease in Australia, causing considerable economic loss through reduced growth rates in grower/finisher pigs and control costs. Swine dysentery is characterized by mucohaemorrhagic colitis, resulting from infection with the anaerobic intestinal spirochaete *Brachyspira hyodysenteriae*. The diseases can be controlled, but worldwide there is concern about reduced susceptibility of many strains to the commonly available antimicrobials. Furthermore, antimicrobials are being withdrawn due to fears of transmission of resistance to human pathogenic microorganisms, or the presence of potentially toxic residues. The aim of this study was to examine the susceptibility of recent Australian *B. hyodysenteriae* isolates to commonly available antimicrobial agents.

Isolates (n=125) of *B. hyodysenteriae* recovered from diagnostic samples received at Murdoch University from farms across Australia between 2002-2007 were tested. Spirochaetes were confirmed as *B. hyodysenteriae* on the basis of their phenotypic properties and amplification in a specific polymerase chain reaction amplifying a portion of the nicotinamide adenine dinucleotide (NADH) oxidase gene (La *et al.*, 2003). All isolates were tested against tiamulin hydrogen fumarate, lincomycin hydrochloride, dimetridazole and tylosin tartrate. In addition, 36 isolates from 2006-2007 were tested against monensin, olaquinox, ampicillin and tetracycline hydrochloride. Serial dilutions of the drugs were made in Trypticase Soy agar with 5% ovine blood, and isolates (105 cells) were drop inoculated in duplicate at each dilution. Growth was assessed by the presence of haemolysis around the inoculum after five days anaerobic incubation. The minimum inhibitory concentration was recorded, and the isolates categorized as susceptible, intermediate or resistant to each antimicrobial (Rønne and Szancer, 1990).

Summary results for the four main antimicrobials tested are shown (Table 1). In addition, the 36 isolates from 2006-2007 were either susceptible (75%) or intermediate (25%) to monensin, resistant (100%) to olaquinox, susceptible (91%), intermediate (6%) or resistant (3%) to tetracycline, and either susceptible (97%) or resistant (1%) to ampicillin.

**Table 1. Antimicrobial susceptibility of 125 Australian isolates of *B. hyodysenteriae***

Antimicrobial	Susceptible	Intermediate	Resistant
Tiamulin	72 (58%)	42 (34%)	11 (9%)
Lincomycin	32 (26%)	81 (65%)	4 (3%)
Dimetridazole	119/119 (100%)	-	-
Tylosin	1 (1%)	1 (1%)	123 (98%)

Antimicrobial resistance patterns were variable. The presence of tiamulin resistant isolates was of concern, as none were found in a survey of pre-2002 isolates (Karlsson *et al.*, 2002). Unfortunately dimetridazole, the drug to which all the isolates were susceptible, is being withdrawn for use in the treatment of SD in Australia. Most of the recent isolates tested were still susceptible to monensin. To reduce current reliance on antimicrobials, the Australian pig industry needs to consider developing alternative methods to help control SD.

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# 3

## Energy utilization

# Net energy system for pigs

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## Introduction

The importance of an accurate energy evaluation system for the formulation of swine diets was introduced to the audience of the Australasian Pig Science Association conference 16 years ago (Noblet and Henry, 1991). Together with the digestible energy (DE) and metabolizable energy (ME) systems, the net energy (NE) system is one of the three most common energy systems used globally to evaluate energy in feedstuffs and complete feeds that are fed to swine. Of these, the NE system has been marketed as the most accurate energy system (Noblet, 2006b), although opinions differ (Susenbeth, 2006). Organisations within Australia, Canada, parts of the EU, and the USA have been struggling with a decision about the introduction and full adoption of the developed NE system, as reflected by a large array of reviews, presentations, or dedicated symposia directed to the NE system in swine nutrition within the last few years (e.g. de Lange and Birkett, 2005; Noblet, 2006a). Historically, most of these countries have been lacking a central authority that determines preferred methods for feed quality evaluation (such as the Centraal Veevoeder Bureau (CVB) in The Netherlands), leaving it up to individual efforts or personal preferences to start initiatives for the adoption of NE systems.

Although NE was likely defined by scientists such as Blaxter or Kleiber and was introduced into swine nutrition by Schiemann *et al.* (1972), French and Dutch scientists and industry stakeholders have been laborious in their efforts to adopt and fine-tune the NE system for swine (Blok, 2006; Noblet, 2006b). Their conclusions have been published in tables with the nutritional composition of feedstuffs (CVB, 2003; Sauvant *et al.*, 2004). In The Netherlands, more than one generation of swine nutritionists has experience with the NE system, and does not question its usefulness relative to the DE and ME systems. Rather, they focus on further improvements of equations to predict the NE values for feedstuffs (Blok, 2006) or identification of limitations and their potential solutions (de Lange and Birkett, 2005).

The NE system has gathered more interest recently in English-speaking countries because of the appearance of a competitive disadvantage by using the DE and ME systems for feed quality evaluation and feed formulation. In the western Canadian feed industry, the emergence of the independent feed consultant (i.e. a feed formulator not working for the commercial feed industry) resulted eventually in the exploration of the NE system to reduce feed costs. The use of the NE system turned into a local competitive advantage resulting in the rapid adoption of the NE system by part of the swine industry.

The advantages of the NE system and the amount of new research required to adopt the NE system have sometimes been overstated. The advantages of the NE versus the DE/ME system are not related to improvements in growth performance or feed efficiency (Frantz *et al.*, 2004). Instead advantages of the NE system are related to: 1) ensuring consistent growth performance and likely carcass quality while making alterations in the macro-nutrient composition and thus NE content of feeds (Cadogan *et al.*, 2005); 2) managing the risk of inclusion of alternative feedstuffs and co-products into swine diets (Smits and Sijtsma, 2007), and 3) reductions in feed costs per kg of feed or lean gain (Payne and Zijlstra, 2007). Energy is the main component of feed costs for swine (i.e. the greatest cost-pressure in swine feed is against energy content), and an accurate system to evaluate energy quality will thus play a role in managing feed costs (Zijlstra *et al.*, 2001; Noblet, 2006b). However, in addition to a focus on the energy system used for diet formulation, large opportunities remain to be fulfilled practically to better manage dietary energy content (Patience *et al.*, 2005) and further reduce feed costs per unit of gain.

In this summary, differences among the main energy evaluation systems will be explained. Possible advantages of the NE system relative to the DE and ME systems for feed formulation, feedstuff selection, and reduced feed costs will be pointed out. Critical steps to adopt the NE system will be listed to allow the audience to explore the NE system within their organization. Derivations of the DE, ME, and NE systems exist, but are not included.

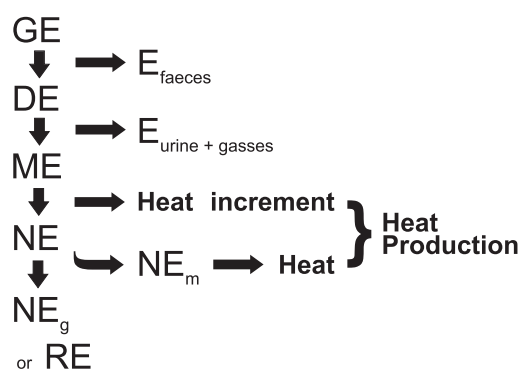
## Energy evaluation systems

Proper feed quality evaluation for energy and amino acids is critical for the effective use of feedstuffs in swine diets. Feed quality evaluation for nutrients will determine first and foremost if a predictable growth performance and carcass quality can be achieved, but can also be linked to other important components of pork production such as nutrient excretion and intestinal health. For energy, the DE, ME, and NE systems are the evaluation systems used



most commonly throughout the world. The DE and ME systems remain popular in North America (NRC, 1998), Australia, and Asia, in part because it is relatively straightforward to determine the DE and ME contents in feeds and feedstuffs. A main disadvantage of the DE and ME system is that they predict animal performance poorly (de Lange and Birkett, 2005).

The DE content of feedstuffs can be measured via the collection of faeces and analyses of gross energy in feed and faeces (Figure 1). Energy digestibility appears to be the most important factor describing changes in energy utilization among feeds (Noblet *et al.*, 1994), indicating the need for laboratory measures that can predict energy digestibility accurately among feedstuffs (Boisen, 2000) and within feedstuffs (Zijlstra, 2006). Energy digestibility averaged 81% for 61 swine feeds, but ranged from 65 to 95%, and a higher variation will be observed among feedstuffs. Energy digestibility is drastically affected by the maturity or body weight of the pig, with sows achieving on average a higher digestibility for energy (85.2 vs. 82.1%) and the macronutrients fat (37.1 vs. 31.6%) and fibre (as NDF, 64.4 vs. 56.3%) than grower pigs (Le Goff and Noblet, 2001), indicating the need for a separate energy evaluation between these two swine categories.



**Figure 1.** Components of feed quality evaluation for energy for grower-finisher pigs. Abbreviations: DE, digestible energy; E, energy; GE, gross energy; ME, metabolizable energy; NE, net energy; NE<sub>g</sub>, net energy for gain; NE<sub>m</sub>, net energy for maintenance; RE, retained energy

For the measurement of ME content, the collection of urine and its analyses for energy content is added, and the small amount of energy lost via gasses (~1%) is generally ignored. The difference between ME and DE is a less important factor describing changes in energy utilization due to its low variability (Noblet *et al.*, 1994). The ME:DE ratio averaged 96% for 61 swine feeds with a narrow range from 94 to 98%. This difference is a largely reflection of the protein content of the feedstuffs, because the urinary energy excretion is largely nitrogenous compounds. High protein feedstuffs thus get a slightly lower ranking in the ME than DE system (Table 1).

Net energy is defined as ME minus the heat increment (Figure 1), or in the case of grower pigs, NE equals retained energy plus the fasting heat production. The heat increment includes the energy costs associated with the metabolic utilization of ME, including the energy cost of ingestion, digestion, and some physical activity. The heat increment is largest for the macronutrients fibre due to enteric fermentation and protein due to energy required for the catabolism of excess absorbed amino acids and excretion of nitrogen. The NE system therefore takes differences in metabolic utilization of ME among nutrients into consideration (Noblet, 2006b). The heat increment (i.e. the difference between ME and NE, also known as 1-k), appears to be an important factor describing changes in energy utilization (Noblet *et al.*, 1994). Efficiencies of utilization of ME for NE (k, %) averaged 74% for 61 swine feeds with a range from 69 to 77%. Among the three main factors impacting energy utilization, energy digestibility, the ME:DE ratio, and k, energy digestibility had a 30% unit range, k an 8% unit range, and the ME:DE ratio a 4% unit range among 61 feeds (Noblet *et al.*, 1994). Any feed quality evaluation system for energy should thus start with a description of energy digestibility.

**Table 1. Relative DE, ME and NE values of selected feedstuffs<sup>1</sup>**

Feedstuff	DE	ME	NE	NE/ME
Animal fat	243	252	300	90
Corn	103	105	112	80
Wheat	101	102	106	78
Barley	94	94	96	77
Reference diet	100	100	100	75
Pea	101	100	98	73
Soybean (full-fat)	116	113	108	72
Wheat bran	68	67	63	71
Distiller's Dried Grains	82	80	71	67
Soybean meal	107	102	82	60
Canola meal	84	81	64	60

<sup>1</sup>Source: Adapted Sauvant *et al.* (2004). Within each system, values are expressed as percentages of the energy value of a reference diet containing 68% wheat, 16% soybean meal, 2.5% fat, 5% wheat bran, 5% peas and 4% vitamins and minerals. NE/ME is the ratio between NE and ME.

As a result of considering efficiency of metabolic utilization, feedstuffs with a high starch content and especially with a high fat content receive a higher relative ranking in the NE system (Table 1), where higher fibre and high protein feedstuffs lose ranking. In other words, the “true” energy of fat and starch sources are underestimated and of protein and fibrous feeds are overestimated in the DE and ME systems. If this assumption is correct, less variation in animal performance should be observed for diet formulated to equal NE than for diets formulated to equal DE and ME, following a change in macronutrient content (Noblet, 2007).

For the absolute values for NE content, refer to Sauvant *et al.* (2004) or CVB (2003); the NE content correlates well between the two data sets. Do not refer to NRC (1998) for NE values; the NRC values do not correlate well with either the French or Dutch system. The absolute NE values are systematically higher in Sauvant *et al.* (2004) than CVB (2003), which seems to be related to a higher estimate of NE required for maintenance in the French NE system. The difference indicates that NE values for feedstuffs should be compared to other NE values within the same data set.

Using measured GE, DE, ME, and NE values for feedstuffs, the energy contributions for each of the four macronutrients can be calculated (Noblet and van Milgen, 2004) (Table 2). The calculated trends for macronutrients confirm the relationships established for feedstuff categories, although some of the absolute values in the table appear less clear. Following these calculations, the value of the macronutrients protein and fibre is severely deducted in the NE system, relative to the DE and ME systems, whereas fat and starch increase their relative importance.

The process to obtain actual NE measurements among major feedstuffs using indirect calorimetry in swine was an enormous undertaking. The sample set of 61 feeds analyzed by Noblet and colleagues with climate respiration and coinciding digestibility experiments in France is therefore unique. These feeds varied widely in macronutrient and feedstuff composition; for example, crude protein ranged from 8 to 24%, ether extract from 0.3 to 8.6%, and starch from 23 to 64% (Noblet *et al.*, 1994). The feeds were tested in 21-day experimental periods for nitrogen balance (day 11 to 18), heat production at high intake (day 15 to 18) and at low intake (day 20). These measurements for heat production at high and low intake (both above maintenance intake) are conducted to predict heat production at zero energy intake, presumably equivalent to fasting heat production. Subsequently, the NE content for each diet was calculated for adding the heat production at zero intake plus retained energy at the high intake level ( $NE = FHP + RE$ ). Considerable debate exists as to whether this is the correct approach to measure fasting heat production, because of its impact on the absolute NE value of these feeds (de Lange and Birkett, 2005); this debate should not affect the ranking of feedstuffs, but is part of the explanation for the difference in absolute NE values between the French and Dutch databases.

**Table 2. Calculated contributions calculated as GE, DE, ME and NE of the four dietary macronutrients in grower pigs (kJ/g)<sup>1</sup>**

Nutrient	GE	DE	ME	NE
Crude protein	22.6	22.5	19.7	11.8
Fat	38.8	31.8	32.2	28.9
Starch	17.5	18.3	18.2	14.8
Sugars	16.7	16.1	15.9	11.5
Residue	18.6	0.5	0.5	-0.9

<sup>1</sup>Source: Noblet and van Milgen (2004). Values were derived from recalculations of data from measurements were conducted on 61 diets fed to 45-kg pigs and coefficients are obtained from multiple linear regression equations (without intercept). Residue is the difference between organic matter and the sum of CP, fat, starch, and sugars; residue might be considered equivalent to fibre.

Unlike routine DE and ME measurements of feedstuffs, continued NE measurements for the sole sake of routine quality evaluation of feedstuffs are unlikely, mostly due to costs of experimentation and apparent lack of need. For practical application of the NE system, equations to predict NE content were therefore developed that link analyses of the total or digestible macronutrient contents in feeds to the measured NE content of feeds (Table 3). The French calorimetry experiments were specifically set up to allow the development of prediction equations via multiple regression analyses (Noblet, 1994). This unique sample set has also been used by the Dutch to further fine-tune their equations to predict NE content (Blok, 2006).

**Table 3. French and Dutch equations for predicting NE in swine feeds (NEg; MJ/kg dry matter; composition: g/kg DM)<sup>1</sup>**

Equation	RSD,%
French <sup>2</sup>	
NEg2 = 0.0121 x DCP + 0.0350 x DEE + 0.0143 x ST + 0.0119 x SU + 0.0086 x DRes	2.4
NEg4 = 0.703 x DE - 0.0041 x CP + 0.0066 x EE - 0.0041 x CF + 0.0020 x ST	1.7
NEg7 = 0.730 x ME - 0.0028 x CP + 0.0055 x EE - 0.0041 x CF + 0.0015 x ST	1.6
NEg9 = 11.69 + 0.0174 x EE + 0.0034 x ST - 0.0296 x Ash - 0.0225 x CF	3.4
Dutch <sup>3</sup>	
NE03 = (10.8xDCP + 36.1xDDEE + 13.5xStarch-Am-e + 12.2xSug-e + 9.5xFCH)/1000	
NE06 = (11.70xDCP + 35.74xDDEE-h + 14.14xStarch-Am-e + 12.75xSug-e + 9.74xFCH)/1000	

<sup>1</sup>CF: Crude Fibre, CP: crude protein, EE: ether extract, ST: starch, DCP: digestible CP, DEE: digestible EE, DRes: digestible residue (i.e. difference between digestible organic matter and other digestible nutrients), FCH = fermentable carbohydrates = fermentable starch and sugars + digestible NSP = digestible organic residue).

<sup>2</sup>From Noblet (2006) with original data from Noblet *et al.* (1994; 2004).

<sup>3</sup>From Blok (2006), with the equations used in 2003 and proposed for 2006 listed.

In summary, various equations have been developed by Noblet and colleagues and by CVB from different combinations of macronutrient analyses, digestible nutrient analyses, and *in vivo* DE and ME measurements (Table 3). The RSD for the equations indicate that the prediction based on macronutrient content was the least accurate, but the advantage of this prediction is that it can be completed solely with laboratory analyses and animal work is not required. The RSD based on digestible nutrient profile was intermediate in accuracy whereas equations based on either DE or ME together with macronutrients were the most robust. The equations can also be used to predict the NE content of a range of samples within a feedstuff, in other words, nutrient digestibility can be predicted from nutrient composition, and NE content can be predicted from digestible nutrient profiles. The latter procedure forms the basis for the Dutch NE system.

Correct determination of NE values of feedstuff samples depends on several factors: 1) prediction equations used, 2) correct determination of total tract digestibility of energy and nutrients, 3) correct laboratory protocol, and 4) correct execution of chosen laboratory protocols. Although the critical prediction equations were derived from one common data set, the use of the different prediction equations results in a different predicted NE value. The differences are sometimes large, and an average NE for the NE predicted by the four using French prediction equations (Table 3) has been suggested (Degussa, 2006). Correct execution of nutrient digestibility studies plays an essential role in reaching accurate NE values for feedstuffs via the use of prediction equations (Jansman, 2006). The identified procedure should be used, because results can differ drastically among starch analyses (Blok, 2006), ether

extract analyses (Blok, 2006), and fibre analyses (Udén *et al.*, 2005). Finally, the correct laboratory procedures should be executed properly, because large differences have been observed among laboratory results for the same analyses (Cromwell *et al.*, 2003; Clowes *et al.*, 2004).

In summary, the NE system is based on indirect calorimetry experimentation of pigs fed an array of feeds, followed by the development of equations to predict NE of feeds and feedstuffs based on macronutrient content, preferably in combination with nutrient digestibility analyses. Net energy NE values for additional feedstuffs are determined subsequently using digestibility analyses and equations to predict NE content in swine.

### Advantages of the NE system

The NE system is basically set up as a system based on the evaluation of macronutrients (Blok, 2006; Noblet, 2006b). In the NE system, starch and fat receive more credit while the protein and fibre receive less credit for their energy-yielding potential than in the DE and ME systems. The NE system can be considered a critical step forward, but perhaps not the final step toward a nutrient based system to evaluate feedstuffs. Its advantages include:

#### *Changes in dietary macronutrients*

Changes in energy value of feeds are related to changes in energy-yielding substrates. Energy systems should predict performance measured as daily gain or energy retention of pigs following changes in dietary energy content measured as DE, ME or NE content. The system related changes in macronutrients best to changes in energy retention provides the best prediction and is most valuable to maintain growth performance. Of the four macronutrients (protein, fat, fibre and starch), specific changes in a macronutrient that likely have been studied most in swine is a reduction in crude protein content. Changes in macronutrient content have also been studied specifically to test whether the DE, ME, or NE system best predicted subsequent growth performance. Obviously, changes in the macronutrient content of feeds are accomplished by changing the feedstuff composition of feeds.

#### *Protein*

Changes in dietary crude protein content have been studied for the last two decades in an attempt to reduce nitrogen excretion by swine (e.g. Gatel and Grosjean, 1992). Reductions in nitrogen excretion could be achieved easily via a reduction in dietary crude protein content while balancing for digestible amino acids, across an array of nitrogen balance studies (e.g. Kerr and Easter, 1995; Zervas and Zijlstra, 2002). However, in diets formulated to equivalent DE or ME content, reducing the dietary crude protein content tended to reduce growth performance or increase carcass fat content (Kerr and Easter, 1995; Tuitoek *et al.*, 1997). Excess absorbed dietary amino acids that are not used for protein deposition will be catabolized in the liver, and excess nitrogen of these amino acids will be excreted in the urine as urea; these processes require energy. The increased carcass fat in pigs fed low-crude protein, AA-supplemented diets formulated to equal DE content may be partially due to more dietary energy being available for fat synthesis as a result of reduced energy expenditure for catabolism of excess dietary protein (Kerr *et al.*, 2003). However, in pigs fed diets with a reduced crude protein content that were balanced for equal NE content, differences in growth performance, feed efficiency, and carcass traits were not observed (Dourmad *et al.*, 1993; Kerr and Easter, 1995; Canh *et al.*, 1998; Le Bellego *et al.*, 2001, 2002; Patience *et al.*, 2003). The NE system therefore properly accounts for the energy that is not used anymore to support the excretion of excess nitrogen and can be used instead to support protein or lipid deposition.

#### *Fat*

Specific changes to swine diets to manage heat stress have been considered for grower-finisher pigs. Swine producers can ameliorate the negative effects of heat stress on late-finishing growth rate by supplementing diets with high levels of fat (Spencer *et al.*, 2005). Digestion of fat can dramatically lower the heat increment (i.e. the thermal effect of feeding) (Forbes and Swift, 1944) and thereby reduce the impact of environmental heat on energy intake. The impact of the DE, ME and NE systems in predicting the energy cost of gain was evaluated in growing-finishing pigs fed diets containing different levels of dietary fat in two experiments (Wu *et al.*, 2007). Diets were based on wheat, corn and soybean meal and supplemented with 0, 1.75, 3.50 and 5.25% tallow. For the combined grower and finisher periods, the DE and ME costs of gain decreased linearly, whereas the NE cost of gain did not decrease. The NE system can predict the performance of growing-finishing pigs more precisely for diets differing in fat content than the DE and ME systems (Wu *et al.*, 2007).

### *Fibre.*

Changes in fibre content have been implemented for various reasons, including pork quality standards. For example, fattening pigs in Italy are fed diets containing 8 to 10% wheat bran to reduce growth rate to 0.6 kg/day to meet Parma Ham quality guidelines. In pig fed these high fibre diets among four periods, retained energy measured via indirect calorimetry had the smaller difference than ME and DE intake versus pigs fed a control diet, indicating that NE content predicted energy retention better than ME and DE content (Galassi *et al.*, 2005). The inclusion of fibre in the diet may affect the NE content by reducing energy required for maintenance via reduced physical activity (Schrama *et al.*, 1998); however, this effect is not consistent among fibre sources (Rijnen *et al.*, 2003b). Fermentable fibre sources such as sugar beet pulp silage reduced physical activity and therefore NE required for maintenance (Rijnen *et al.*, 2003a), whereas less fermentable fibre sources such as solvent-extracted coconut meal and soybean hulls only show a marginal effect of reduced physical activity (Rijnen *et al.*, 2003b). Differences in fibre fermentation characteristics are not accounted for in any of the equations to predict NE content.

In summary, the energy costs of growth or the daily energy requirement are independent on diet composition when expressed on a NE basis (Noblet, 2006). The energy cost of growth is decreased on a DE or ME basis when dietary crude protein content is decreased or fat content is increased. Further proof is provided that the DE and ME system overestimate energy value of protein and underestimate the energy value of fat.

### **Inclusion of alternative feedstuffs**

In Canada and the USA, the inclusion of co-products has been considered advantageous during periods of price increases for feed grains or main protein sources. Few countries have a logistical system in place that relies on co-products such as countries in the EU (FEFAC, 2005). However, some countries such as The Netherlands have historically been heavily dependent on the NE system for accurate ranking of feedstuffs based on their energy values in feed tables (CVB, 2003). The difference in approach to energy evaluation among continents is reflected in research outputs. The inclusion of new co-products such as corn and wheat dried distiller's grain plus solubles is regularly tested in North America by feeding diets formulated with incremental concentrations of the test feedstuff to an equivalent DE or ME content. Not surprisingly, this approach frequently results in reduced growth performance (e.g. Roth-Maier *et al.*, 2004; Friesen *et al.*, 2006; Thacker, 2006; Whitney *et al.*, 2006; Widyaratne and Zijlstra, 2007). Subsequently, the test feedstuff is blamed in studies that observed a reduced growth performance, as opposed to the feed quality evaluation system used to analyse dietary energy and amino acid content.

In Europe, getting an accurate prediction of the NE content of alternative feedstuffs is considered important (Smits and Sijtsma, 2007) to attempt assurance of equivalent growth performance following the introduction of alternative feedstuffs or co-products. However, European validation studies with alternative feedstuffs in swine diets formulated to an equal NE content are either rarely conducted or published in the scientific literature. As a rare example, incremental increasing levels of dietary content of canola meal up to 18% in diets formulated for grower finisher pigs to equal NE and digestible amino acids did not change growth performance in a French study (Albar *et al.*, 2001). In North America, formulating diets to equal NE content might also result in less performance difference being observed following the introduction of alternative feedstuffs, such as the zero-tannin faba bean (Zijlstra *et al.*, 2004; Gunawardena *et al.*, 2007; Young *et al.*, 2007). Controlled comparisons of the introduction of alternative feedstuffs into swine diets formulated using the DE/ME system and NE system are not part of the scientific literature. Feed quality evaluation for energy, however, likely plays a role in the successful introduction of new feedstuffs.

### **Feed costs**

Using a more accurate system of feed quality evaluation for energy relative to the DE and ME systems means that starch and fat will receive additional credit and protein and fibre will receive less credit. Diets formulated using the NE system are typically lower in crude protein content than those formulated using the DE or ME system (Payne and Zijlstra, 2007) (Table 4), because heat lost during catabolism and excretion of excess nitrogen is considered in the NE system. By maintaining the identical Lys content in the diets, continued use of the ideal protein concepts, and accounting for standardized ileal digestible amino acids in feedstuffs, dietary content of other essential amino acids are maintained. These changes mean that more expensive protein feedstuffs will be partially replaced by cereal grains and synthetic amino acids, with the current prices for these three feedstuff categories. The difference in calculating the energy-yield for the macronutrients also means that the calculated ME content of the diets generally declines. The content of fat sources might increase slightly as well, but because these feedstuffs tend to be more expensive, an increase in dietary inclusion is less likely than for cereal grains.

**Table 4. Example diets formulated using NE compared with ME<sup>1</sup>**

Item	25 to 50 kg bodyweight		50 to 75 kg body weight	
	ME	NE	ME	NE
<b>Ingredient, %</b>				
Wheat	22.81	62.88	42.78	82.98
Corn	31.16	-	31.53	-
Field peas	20.00	16.72	-	-
Soybean meal (48%)	17.64	11.72	17.59	8.74
Tallow	5.00	5.00	5.00	5.00
Biolys (50.7% Lys)	0.37	0.64	0.50	0.83
L-Threonine	0.08	0.14	0.07	0.16
DL-Methionine	0.10	0.10	0.01	0.04
Other <sup>2</sup>	2.84	2.80	2.52	2.49
<b>Calculated content</b>				
ME, MJ/kg	14.11	14.03	14.19	14.03
NE, MJ/kg	10.76	10.76	10.97	10.97
Crude protein, %	18.5	17.8	17.0	15.7
SID Lys, %	1.00	1.00	0.88	0.88
SID Thr, %	0.62	0.62	0.55	0.55
SID Met+Cys	0.57	0.57	0.50	0.50
SID Trp, %	0.17	0.17	0.17	0.16
Cost, CDN\$/MT	162.35	158.67	159.74	155.00

<sup>1</sup>Source: Payne and Zijlstra (2007).

<sup>2</sup>Includes macro- and micro-minerals and vitamins.

Example diet formulations and pricing scenario differ across the globe, but in the North American example (Table 4), feed costs per MT of feed were reduced following the use of the NE system instead of the ME system for feed formulation (Payne and Zijlstra, 2007). Negative effects on animal growth performance are not expected following these changes in feed formulation; hence, feed costs per unit of lean grain or per pig are expected to decline slightly (Patience, 2005; Payne, 2006). The use of the NE system instead of the DE or ME system will also mean that dietary crude protein will decline while content of essential amino acids will be maintained and therefore nitrogen excretion will decline, which is an indirect benefit of the adoption of the NE system. For each %-unit reduction in dietary crude protein content, a 10% reduction in nitrogen excretion and ammonia emissions can be expected (Canh *et al.*, 1998).

### Shortcomings of the NE system

An original intent of feed quality evaluation was to relate feedstuffs to nutrient requirements of animals and thereby reach a predictable growth performance. The DE and ME systems fall short of the NE system in predicting energy retention, growth performance, and carcass quality (Noblet, 2007; Cadogan *et al.*, 2005). The NE system might thus be superior to the DE and ME system to formulate diets for grower-finisher pigs, and is a worthwhile energy system to pursue for practical feed formulation. However, in contrast to the positive aspect of the NE system, the ME system has been described as a better energy system to describe the potential energy available for other requirements such as pregnancy, lactation, physical activity, etc. (Susenbeth, 2006). Still, the NE system is clearly progressing in feed quality evaluation from the DE and ME system by further describing the interaction between the animal and feed on the road towards achieving a predictable growth performance, but it might not be the final stop, because, for example, effects of kinetics of nutrient absorptions and impact of feed intake are ignored in the current NE system.

Most studies comparing the energy systems have been conducted with restricted-fed pigs housed in calorimetry chambers (Noblet *et al.*, 1994), whereas validation studies with pigs with free access to feed are scarce. Only the latter would allow the pigs to express their feed intake and other behaviours and nutrients to express their potential, and would allow a full assessment of the potential of the NE system to predict feed efficiency and growth performance. For

example, fibre included in feeds clearly affects the NE content of feeds via changes in efficiency of ME utilization to support growth, but might also reduce physical activity (Schrama *et al.*, 1998) and reduce voluntary feed intake (e.g. Skiba *et al.*, 2006). The NE system does not account for such effects, which may help explain why an accurate prediction of growth performance in pigs with free access to feed might be difficult to reach with the current NE system.

Shortcomings of the DE and ME system from an energetics and nutrient flow perspective have been described (de Lange and Birkett, 2005). These deficiencies include lack of accuracy for maintenance requirements, poor prediction of performance, and lack of describing the relationships between animal and diet. The NE system partially deals with these concerns, in particular the relationship between animal and diet (de Lange and Birkett, 2005); however, limitations remain in the NE system for the estimation of the maintenance requirement and reflection in variations in the metabolic use of energy for the different functions such as lipid retention and ATP production (Birkett and de Lange, 2001).

Mathematical models may overcome the shortcoming of the current energy systems (de Lange and Birkett, 2005) via inclusion of additional swine production components, and additional variables such as feed intake; however, the models appear removed from routine application in feed mills by swine nutritionists. Additional software will be required for technology adaptation, whereas the NE system can be adopted within the existing software.

### Adoption of NE system

Following a decision to pursue the NE system for feed formulation, an action plan for implementation needs to be developed (Payne and Zijlstra, 2007). This action plan should focus on feedstuff composition and formulated diet energy content (Table 5). For feedstuffs, the focus will be on the development or update of a database and, in particular, the macronutrient composition of the important feedstuffs. The two most-widely used NE systems are solidly based on the macro-nutrient composition of feedstuffs (CVB, 2003; Sauvant *et al.*, 2004), also indicating that specific laboratory analyses of macronutrients (Blok, 2006) and variation in laboratory results (Clowes *et al.*, 2004) should be part of the evaluation. Reformulating the diets for NE content as suggested in Table 4, and a direct recalculation of diet energy content could be used ( $NE = 0.74 \times ME$  or  $NE = 0.71 \times DE$ ; Noblet, 2006b).

**Table 5. Suggested action plan for implementing NE<sup>1</sup>**

Step	Activity
1	Identify energy-yielding raw materials that will be used in grower-finish diet formulations
2	Collect required raw material samples for pre-determined length of time
3	Analyze raw materials for their nutrient content. These include but are not limited to: dry matter, crude protein, ether extract, crude fibre, acid and neutral detergent fibre, starch, sugar
4	Calculate DE, ME, and NE values for raw materials based on raw material analyses using currently available NE prediction equations
5	Compare calculated DE, ME, and NE values for raw materials with values currently being used in formulation software
6	Update nutrient matrices for energy-yielding raw materials in diet formulation software
7	Insert NE as a nutrient in grow-finish diets, and then reformulate all diets using current energy system (DE or ME)
8	Based on calculated NE from reformulated diets, remove former energy restrictions (on DE or ME) and place new nutrient restrictions on NE
9	Re-optimize all diets to balance on their NE content

<sup>1</sup>Source: Payne and Zijlstra (2007)

The NE system could be simply implemented without a critical evaluation of the ingredient database. However, ingredients vary regionally in their macronutrient profile (Fairbairn *et al.*, 1999; Zijlstra *et al.*, 1999) and thus NE content. Therefore, a critical evaluation of feedstuffs is recommended for risk management, and should preferably be linked with an evaluation of rapid feed quality to allow flexible feed formulation. Following the switch to the NE system, the DE or ME content of the diet could initially be monitored for greater assurance. Feed costs, growth performance and carcass characteristics should also be monitored to ensure the switch is implemented successfully.

## Conclusions

A practical NE system has been developed in France and The Netherlands based on a decade of solid scientific research. The NE system is being applied in specific regions in the world, and appears to be superior to the DE and ME systems in terms of growth performance, greater flexibility in feedstuff use and reduced feed costs. The advantage of the NE system seems to be based on a more accurate assessment of the energy yield of the macronutrients (protein, starch, fibre and fat) than the DE and ME systems can provide. However, the NE system might not provide the most accurate prediction possible for growth performance. Adopting the NE system is relatively easy, and it is being adopted locally in western Canada via feed consultants. Some limitations remain within the NE system and these should be addressed via other means, such as modelling.

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## Carcass growth is influenced by dietary factors in addition to net energy

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Cadogan *et al.* (2005) showed that carcass growth and carcass characteristics were more strongly related to net energy (NE) than digestible energy (DE). There are concerns, however, that the influence of NE changes when diets are formulated from different raw ingredients, and that NE is not additive when diets are formulated in linear programs.

To test the hypothesis that diets formulated on NE produce more consistent growth performance and carcass characteristics, 200 entire males were allocated to 54 treatments at  $54.7 \pm 3.9$  kg live weight. Dietary NE was kept constant at 10.3 MJ/kg. The four diets consisted of different bases, from high starch, to high fibre and fat, and compared different cereal based diets. The diets were formulated to an available lysine of 0.07 g/MJ NE (0.54 g/MJ DE), and other essential aminos were balanced to the ideal protein ratio. All diets were supplemented with phytase and xylanase to hydrolyze anti-nutritional factors that restrict nutrient intake. Pigs were housed in group pens of 10 pens and offered diets *ad libitum* for 42 days. Average daily gain (ADG), average daily intake (ADI) and feed conversion ratio (FCR) were measured weekly. On day 43, pigs were killed and carcass weight (CWT), dressing percentage (D%) and P2 backfat recorded.

**Table 1. Effects of differing raw materials and DE on growth and carcass parameters of male pigs offered diets formulated to 10.3 MJ/kg NE**

Diet	Low protein, wheat based	High NDF <sup>1</sup> , wheat, canola	Sorghum, soybean	Mixed cereal, high protein	SEM	Significance <sup>2</sup>
DE	13.9	14.0	14.1	14.2		
ADG	808b	894a	872ab	803b	4.36	0.030
ADI	2.117	2.075	2.240	2.088	0.033	0.319
FCR	2.62b	2.34a	2.57ab	2.60a	0.040	0.036
CWT	69.6	71.4	71.2	68.4	0.512	0.134
D%	77.3a	76.9ab	76.8ab	76.1b	0.189	0.019
P2	10.5	10.4	10.7	9.9	0.293	0.311
Leg Fat	11.6	11.9	12.2	11.1	0.252	0.316

<sup>1</sup>NDF = Neutral Detergent Fibre, <sup>2</sup>\*P<0.05, \*\*P<0.01, \*\*\*P<0.001. <sup>abc</sup>Means with different superscripts are significantly different (P<0.05).

Diet type had a significant influence on ADG, FCR and D % (P<0.05) however, there was no difference in ADI, CWT and P2. The diet containing high neutral detergent fibre (NDF), wheat and canola produced the highest ADG, FCR and carcass weight, and was significantly better than the low protein and the high protein diets (P<0.05). There was a direct relationship between the growth and carcass measurements and NE. However, a negative relationship of DE ( $R^2 = -0.24$ ;  $P = 0.032$ ) and D%. The crude protein level also produced a negative trend on D% ( $R^2 = -0.18$ ;  $P = 0.06$ ). The present study suggests that NE is the superior measure of energy for formulating pig diets. However, dietary factors other than energy can also influence efficient live weight gain and carcass growth performance.

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## Supplemental dietary fat increases growth performance of grower and finisher pigs

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Supplemental fat is routinely included in young pig diets but often removed with increasing weight to match the perceived declining need for high energy as pigs grow and thus to prevent excessive fat deposition during finishing. However, recent results suggest that early positive effects on growth may be lost if fat is removed from subsequent diets and that, for reasons yet to be established, older pigs may be more responsive to supplemental fat than younger pigs (Campbell, 2005). This project was designed to characterize the effects of added fat on performance, carcass quality and the possible mechanisms of why the pig responds like it does to added fat. A total of 288 pigs (144 boars and 144 gilts; 30 kg) in pens of 12 were offered diets containing 0%, 4% and 8% added fat for five weeks followed by the diets containing 0% or 4% added fat for the subsequent five weeks. The diets were formulated to contain 13.5, 14.3 and 15.2 MJ DE /kg, respectively. The amino acid:DE ratio was maintained constant in all diets. Live weight and feed intake were measured weekly. Pen growth performance and carcass data were analyzed by analysis of variance.

During the first 35 days the growth rate of both sexes increased linearly with increasing dietary fat content while feed intake and feed conversion were not significantly affected by the dietary treatments (data not shown). Dietary fat supplementation in the initial five weeks tended to reduce daily gain over the second period but had no effect on carcass weight or P2 fat thickness at 10 weeks (Table 1). In contrast, dietary fat supplementation during the second or both periods increased growth rate, carcass weight, P2 fat thickness and tended to improve feed efficiency.

**Table 1. The effect of changing dietary fat levels on feed:gain, carcass weight and P2 backfat of gilts and boars fed either no dietary fat (0%) or diets with either 4% or 8% fat for an initial period of 34 days, then changed to either 0% or 4% dietary fat from days 35 to 70**

Dietary Fat day 0-34	Treatment Effects				Sex Effect	
	0% fat		4 or 8% fat		Gilt	Boar
Dietary Fat day 35-70	0% fat	4% fat	0% fat	4% fat		
Feed:Gain	3.3	2.9	3.3	3.2	3.3	3.1
Carcass weight (kg)	66.8 <sup>c</sup>	73.3 <sup>d</sup>	68 <sup>c</sup>	70.8 <sup>d</sup>	69.7	69.8
P2 (mm)	10.2 <sup>c</sup>	11.6 <sup>d</sup>	10.7 <sup>c</sup>	11.5 <sup>d</sup>	11.1 <sup>a</sup>	10.8 <sup>b</sup>

<sup>a,b</sup> across rows indicate significant main effect of sex (P<0.05)

<sup>c,d</sup> across rows indicate significant main dietary effect day 35-70 (P<0.05)

These data tend to confirm those of Campbell (2005) and suggest adding fat to the diets offered to pigs in one period adversely affects growth rate in subsequent periods, and has little or no effect on final carcass weight if fat is removed in the second period. The data also demonstrate the positive effects of fat and or higher energy diets in the finisher period on carcass weight, and suggest 14.3 MJ DE/kg may be the most cost effective energy level to use for pigs between 60 and 100 kg. In conclusion, dietary fat supplementation from 30 kg can increase growth performance and carcass weight without any detrimental effects on carcass P2 once the effect of carcass weight is taken into account. There is no evidence to suggest that removing fat from the diet is worthwhile once it is already present. Importantly, feeding fat in the finisher period results in sustained growth promotion and, more importantly, increased carcass weight.

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## Bedding material consumption by growing pigs depresses overall diet energy digestibility

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Deep litter housing systems are widely used by the Australian pig industry, but consumption of deep litter material (straw, rice hulls etc.) by the housed pigs makes it difficult to develop accurate diets to meet pig requirements and hence optimize the nutrition of these animals (van Barneveld *et al.*, 2003). In particular, consumption of bedding material may reduce overall nutrient and energy digestibility as well as diluting total digestible energy intake. The aim of this experiment was to define the influence of graded levels of bedding material intake on the ileal and faecal digestibility of gross energy in growing pigs.

Ileal and faecal gross energy digestibility was determined using seven male pigs (commercial genotype; 35 kg live weight) fitted with simple T-piece cannulas 15 cm anterior to the ileo-caecal valve that were fed each of seven diets in a 7x7 cross over design. A basal diet was formulated to contain 14.3 MJ digestible energy (DE) and 0.69 g available lysine:MJ DE. Celite was included in the diet as an acid-insoluble ash marker. Rice hulls and barley straw were added to the basal diet at levels of 5, 10 or 15% respectively based on upper levels of bedding material intake reported by van Barneveld *et al.* (2003). All diets were cold press pelleted before feeding. Following a seven-day recovery period after surgery, pigs were fed the diets for five days followed by two days of digesta and faeces collection over eight hours. Digesta was collected directly onto ice before bulking and freezing. Faeces were collected as voided, bulked and frozen. Data were subjected to an analysis of variance and means were separated by least significant difference ( $P < 0.05$ ).

Consumption of rice hulls and barley straw by growing pigs at levels of 5% had no significant effect ( $P > 0.05$ ) on the ileal digestibility of gross energy, but energy digestibility was significantly depressed at consumption levels above 10% (Table 1). A significant depression in faecal gross energy digestibility was observed for both rice hulls and barley straw at all inclusion levels. Barley straw consumption above 10% had a greater influence on energy digestion in the small intestine whereas rice hull consumption induced a greater depression in gross energy digestion across the whole tract. As well as diluting total digestible energy intake, bedding material depresses energy digestibility as consumption increases and the type of bedding material has varying influences on the site of energy digestion.

**Table 1. Ileal and faecal digestibility of gross energy in diets containing 0, 5, 10 or 15% rice hulls or barley straw**

Treatment	Bedding material inclusion (%)				SEM	P-value
	0	5	10	15		
<i>Ileal</i>						
Rice hulls	73.6 <sup>a</sup>	70.1 <sup>ab</sup>	68.8 <sup>b</sup>	63.76 <sup>c</sup>	2.13	$P < 0.01$
Barley straw	73.6 <sup>a</sup>	70.6 <sup>a</sup>	63.4 <sup>b</sup>	57.2 <sup>b</sup>	3.40	$P < 0.001$
<i>Faecal</i>						
Rice hulls	87.8 <sup>a</sup>	83.0 <sup>b</sup>	80.1 <sup>c</sup>	73.9 <sup>d</sup>	0.63	$P < 0.001$
Barley straw	87.8 <sup>a</sup>	84.7 <sup>b</sup>	79.4 <sup>c</sup>	77.6 <sup>d</sup>	1.44	$P < 0.001$

<sup>a,b,c,d</sup>Means in a column with different superscripts differ significantly ( $P < 0.05$ ).

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## Bedding material consumption by growing pigs alters the digestible lysine:digestible energy ratio of the diet

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The consumption of graded levels of rice hulls and barley straw by growing pigs housed in deep litter systems has the potential to reduce the ileal and faecal digestibility of gross energy significantly if consumption exceeds 5% (van Barneveld *et al.*, 2007). While increased intake may be able to compensate for a reduction in energy digestibility, this is not the case if bedding material consumption depresses the digestion of other nutrients and alters the overall digestible lysine:digestible energy ratio of the diet. The aim of this experiment was to define the influence of graded levels of bedding material intake on the ileal digestibility of lysine in growing pigs and the resulting digestible lysine:digestible energy ratio of the entire diet.

Ileal lysine digestibility was determined using seven male pigs (commercial genotype; 35 kg live weight) fitted with simple T-piece cannulas 15 cm anterior to the ileo-caecal valve that were fed each of seven diets in a 7x7 cross over design. A basal diet was formulated to contain 14.3 MJ digestible energy (DE) and 0.69 g available lysine: MJ DE. Celite was included in the diet as an acid-insoluble ash marker. Rice hulls and barley straw were added to the basal diet at levels of 5, 10 or 15% respectively, with the inclusion levels based on upper bedding material intake levels reported by van Barneveld *et al.* (2003). All diets were cold press pelleted before feeding. Following a seven-day surgery recovery period, pigs were fed the diets for five days followed by two days of digesta and faeces collection over eight hours. Digesta was collected directly onto ice before bulking and freezing. Data were subjected to an analysis of variance and means were separated by least significant difference ( $P < 0.05$ ). Ileal digestible lysine:digestible energy ratios were calculated using energy digestibility measurements reported by van Barneveld *et al.* (2007).

Consumption of rice hulls at more than 5% significantly depressed ( $P < 0.05$ ) ileal lysine digestibility whereas consumption of barley straw did not exert an influence until consumption reached 10% of the total diet (Table 1). While ileal and faecal energy digestibility is also depressed by bedding material consumption (van Barneveld *et al.*, 2007), the reduction in lysine digestibility relative to energy digestibility is sufficient to also depress the dietary digestible lysine:digestible energy ratio. This will influence the overall efficiency of production in deep litter systems and needs to be accommodated when formulating diets.

**Table 1. Ileal digestibility of lysine (%) in diets containing 0, 5, 10 or 15% rice hulls or barley straw**

Treatment	Bedding material inclusion (%)				SEM	P-value
	0	5	10	15		
Rice hulls	94.8a	92.5b	91.7b	88.9c	0.89	$P < 0.001$
Barley straw	94.8a	95.1a	92.8b	93.1b	0.71	$P < 0.009$
Ileal digestible lysine (g):digestible energy(MJ)						
Rice hulls	0.73	0.64	0.67	0.66		
Barley straw	0.73	0.68	0.57	0.62		

<sup>a,b,c</sup>Means in a column with different superscripts differ significantly ( $P < 0.05$ ).

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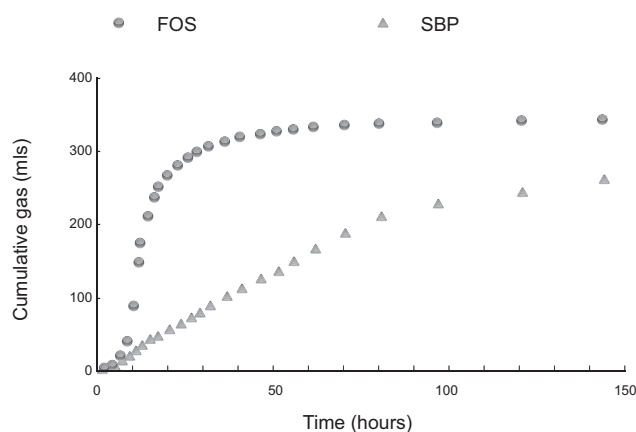
## Use of cumulative gas production to assess feed ingredients for gastro-intestinal fermentability

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Fermentation in the monogastric gastrointestinal tract (GIT) can have a positive effect on the health of the GIT and the animal (Williams *et al.*, 2001). Fermentable carbohydrates are considered promising candidates for stimulating beneficial microflora and potentially reducing the need for anti-microbial agents. These compounds are not absorbed in the upper GIT or hydrolyzed by mammalian digestive enzymes.

The cumulative gas production technique (Williams *et al.*, 2005) is an *in vitro* measure of a fermentation end-product. Such measurements indicate the kinetics of fermentation. Knowledge of the kinetics of pig feed ingredients can help identify the approximate location of the fermentation process, if the transit time in the GIT is taken into account. For example, a rapidly fermented ingredient is more likely to be fermented in the small intestine. Conversely, if an ingredient is only slowly fermented it is likely it will not be fermented efficiently in the GIT and another ingredient should be chosen to stimulate beneficial microflora. For example, sugarbeet pulp (SBP) is more slowly fermented than fructo-oligosaccharides (FOS), which are known to be indigestible and are therefore not detected at the terminal ileum (Houdijk, 1998).



**Figure 1.** Cumulative gas production of sugarbeet pulp (SBP) and fructo-oligosaccharide (FOS)

Knowing the kinetics and end-products of a range of potential feed ingredients makes it possible to design diets that will stimulate appropriate fermentation and reduce the need for anti-microbial agents in feeds.

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## Ungelatinized undigested starch in pig digesta

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The Premium Grain for Livestock Program (PGLP) showed that the ileal and faecal digestible energy (DE) content of cereal grains for pigs ranged, respectively, from 6–16 and 12–17 MJ/kg dry matter (Black, 2006). The ileal: faecal DE ratio (IFR) also varied from 0.6–0.9, which suggests the flow of starch from the ileum may be substantial and varies between grain samples. An experiment was designed to measure the amount and form of starch appearing at the ileum and faeces of pigs fed cereal grains which varied in IFR. The null hypothesis was that starch from cereal grains is not completely digested in the small intestines of pigs, and that it varies between grain samples.

Ileal digesta was collected within PGLP from five pigs each fed diets containing one of three barley and two sorghum samples. The design was a randomized complete block with a balanced allocation of animals to blocks. The data were analyzed using linear mixed models with the statistical package ASReml (VSN International Ltd). The model included fixed effects for grain type and variety and random effects for animal and block. Total starch was determined using the dimethyl sulphoxide,  $\alpha$ -amylase and amyloglucosidase method. Starch morphology was also examined under light microscopy with polarising units.

A significant difference in the starch content was found between the grains ( $P < 0.001$ ). The predicted total starch content of ileal digesta is shown in Figure 1 with the corresponding range in IFR for each pig. The average starch content of ileal digesta ranged from 4–20% for the barley, and from 13–18% for sorghum (predicted mean  $\pm$  SE is shown for five pigs). Microscopy (Figure 2) using polarized light showed bright 'Maltese cross' patterns that are characteristic of crystalline starch granules. All ileal samples contained starch granules (Figure 2A). Faecal samples also contained starch granules (Figure 2B).

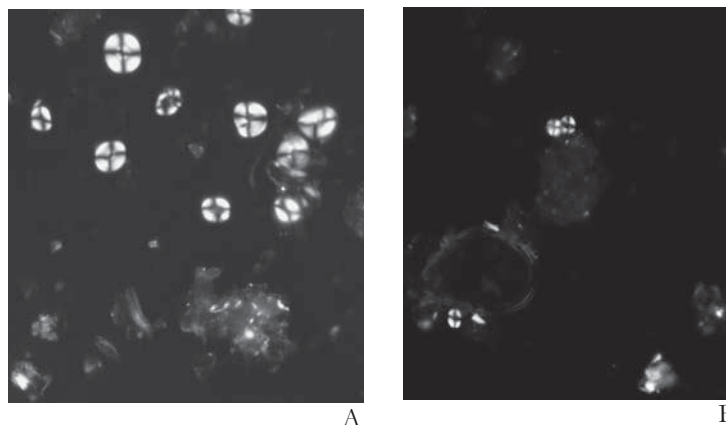
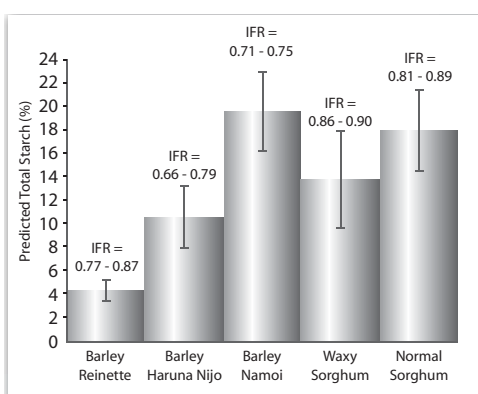


Figure 1. Starch contents of the ileal samples

Figure 2. Birefringent granules in ileal (A) and faecal (B) samples from Namoi

The results confirm that ileal digesta from pigs fed cereal grains contain significant amounts of starch that varies with the individual grain sample consumed. The presence of crystalline starch in both ileal and faecal material shows that some starch was not gelatinized during the cold-pelleting process or by digestion and fermentation in the gastrointestinal tract. Gelatinization is reported to improve digestibility of starch (Sun *et al.*, 2006). Feed processes that increase starch gelatinization may raise digestibility and the DE content of grains for pigs.

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# 4

## Nutrition methodology and effluent management

## Use of lithium chloride to measure feed intake of individual pigs housed in groups – effect of inclusion level on *ad libitum* feed intake and plasma lithium concentration

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Measuring the feed intake of individual pigs housed in groups is a difficult task. Capacity to achieve this would facilitate measurement of variation in feed intake of pigs within and between pens - enabling assessment of feed access, stocking density influences and other factors potentially promoting variability in pig performance. Lithium chloride has been used to estimate supplement intake in grazing ruminants (Kahn, 1994) and has high potential as a marker given it is cheap, stable, easily analyzed and not present in normal pig feed ingredients. The objective of this experiment was to establish the optimum inclusion rate of lithium chloride in grower pig diets that would not compromise *ad libitum* feed intake while allowing accurate measurement of lithium levels in collected blood, to assess its potential for use in feed intake measurements.

Twenty-five male pigs (hybrid, mainly Large White x Landrace) housed in individual pens were allocated to one of five dietary treatments based on a randomized block design (blocked by live weight) at eight weeks of age and  $26.4 \pm 1.2$  (mean  $\pm$  SD) kg live weight. Treatments consisted of the same basal diet for six days (13.9 MJ digestible energy (DE)/kg; 0.68 g available lysine/MJ DE) but varying lithium chloride concentration (0.2, 0.4, 0.6, 0.8 and 1.0 g/kg, respectively) on day seven. Daily voluntary feed intake (VFI) was measured during the 24 hours after experimental diets were presented and for the prior six days. The mean VFI's over the six days were used as covariates in VFI analysis during the lithium chloride inclusion period, allowing each pig to be its own control. Blood was collected from the pigs by jugular venapuncture, while restrained with a snout snare, 24 hours after feed was offered and plasma lithium concentration was assessed (Symbio Alliance Pty Ltd).

Before the feeding of experimental diets, there was no significant difference in mean VFI between treatments groups ( $P=0.669$ ) (Table 1). Mean VFI during the 24 hours when lithium chloride was fed, ranged from 1.21 kg (1.0 g/kg lithium chloride) to 1.44 kg (0.2 g/kg lithium chloride) but these differences were not significant ( $P=0.732$ ). Plasma lithium concentrations ranged from 0.114 (0.2 g/kg lithium chloride) to 0.634 mmol/L (1.0 g/kg lithium chloride). Lithium concentrations followed a linear relationship with increased inclusion rate as per the equation: Plasma [Li] =  $0.5615 * \text{Inclusion Rate} - 0.0235$  ( $R^2=0.86$ ).

**Table 1. Influence of dietary lithium chloride inclusion at 0.2, 0.4, 0.6, 0.8 or 1.0 g/kg, on voluntary feed intake (VFI (SE); kg/day) and subsequent plasma lithium concentration (mmol/L) of growing pigs**

Treatment	Mean VFI before LiCl inclusion, mean (SE)	VFI during LiCl inclusion, mean (SE)	Plasma lithium concentration mean (SE)
0.2 g/kg	1.22 (0.06)	1.44 (0.08)	0.114 (0.018)
0.4 g/kg	1.37 (0.12)	1.27 (0.16)	0.223 (0.019)
0.6 g/kg	1.36 (0.10)	1.40 (0.17)	0.356 (0.044)
0.8 g/kg	1.41 (0.11)	1.33 (0.12)	0.421 (0.022)
1.0 g/kg	1.38 (0.07)	1.21 (0.10)	0.634 (0.041)

While the lowest VFI was recorded for the highest lithium chloride inclusion level, intermediate treatments did not show a linear effect. These results support the hypothesis that the inclusion of lithium chloride at these levels as a feed intake marker will not compromise the VFI of the animal while delivering measurable concentrations of plasma lithium. In this study blood collection was done 24 hours after feed was offered however, before lithium chloride can be used to assess feed intake, the time of blood sampling after ingestion that best reflects VFI must be determined.

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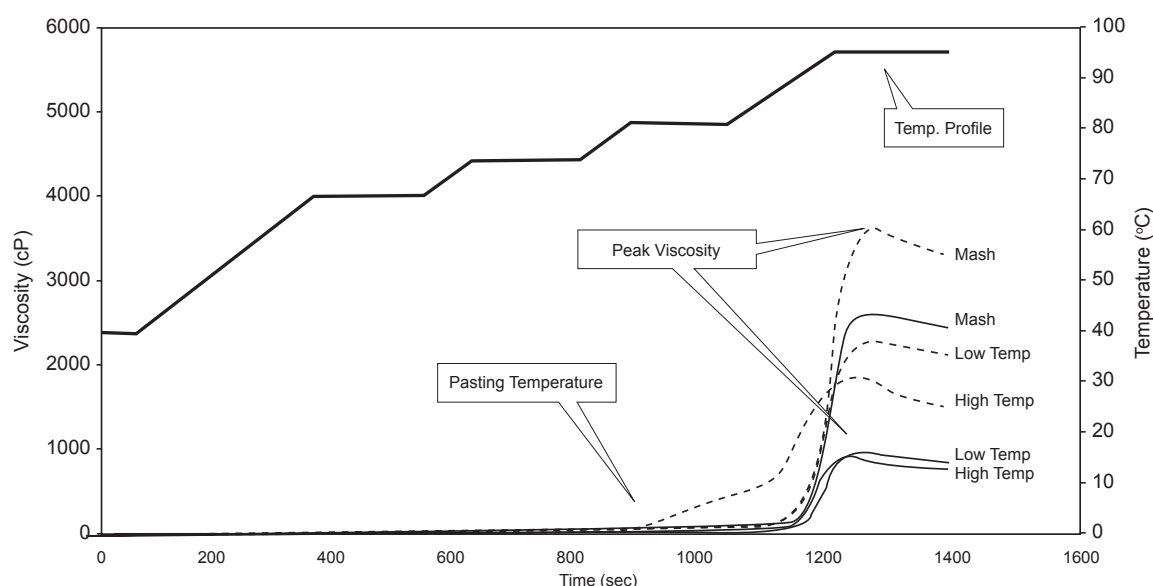
## Use of the rapid visco analyser to evaluate finished feeds

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The Rapid Visco Analyser (RVA) is widely used in the flour industry for comparison of the gelatinization and retrogradation behaviours of starch-based materials. While it does not measure a discrete chemical entity, this technique is responsive to interactions between components in the sample being tested and the concentration and type of starch being analyzed (Blakeney and Booth, 2000). This study examined how changes in non-starch feed components may affect the pasting properties of sorghum-based finished feeds and the potential application of the technique in stock feed manufacture.

Sorghum samples were obtained both prior to and after heat treatment in a commercial feed mill using a mixer/conditioner and expander system to provide: High, 75-85°C; Low, 50-70°C; and Mash (no heat) treatments. Sorghum grain was also used as an inclusion in weaner pig diets (75% grain, 25% protein and vitamin/mineral premix) prepared using the same heat-treatment profiles. Finely ground sub-samples (4 g, 11.2% moisture (Grain Only) and 10.1% moisture (Whole Diet)) were mixed with 27 g of distilled water and tested (Newport Scientific RVA Model 4) using an extended temperature profile as suggested by Blakeney and Booth (2000).



**Figure 1.** Pasting curves for sorghum (-----) and sorghum based weaner diets (—) obtained by Rapid Visco Analysis using the temperature profile (—) displayed

Pasting curves for sorghum grain showed a progressive reduction in peak viscosity with increased processing temperature consistent with suspected changes in the gelatinization of the sample. The pasting curves also show the peak viscosity for the finished feed was less than that of any of the grain samples. This suggests the RVA technique is sensitive to dilution of the grain component with other ingredients and is suited to the further characterization of processed grain-based diets.

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## Symposium – effluent management

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Over the past decade in Australia, there has been a vast increase in community interest and awareness of issues such as environmental protection. Consumers are also demanding that their food meets strict safety standards and that it is produced in a 'clean, green' manner without adversely affecting the environment or breaching animal welfare considerations. Large national food retailers have responded to consumer demands by initiating various quality assurance standards.

The urbanization of Australian society has eroded the political influence of rural industries. Social tensions have arisen as urban encroachment has resulted in people from an urban background moving into areas traditionally used for commercial primary production. In many cases, the newcomers do not earn their primary income from the land and often have vastly different values and lifestyle expectations in comparison to traditional primary producers. The media have sometimes portrayed primary producers and rural industries as being irresponsible in the way they manage environmental and other issues such as animal welfare and food safety.

These factors have resulted in environmental and food safety issues moving from a government policy and legislative backwater, to a far more prominent position. State governments have responded to the changing social and political climate by introducing new and/or revised legislation such as the Environmental Protection Act 1994 that was implemented in the Queensland pig industry in 1996. The object of this legislation is to protect Queensland's environment while allowing for 'ecologically sustainable development' (ESD) that is defined as 'development that improves the total quality of life, both now and in the future, in a way that maintains the ecological processes on which life depends'.

The introduction of this legislation resulted in the implementation of more rigorous assessment and compliance requirements for new and existing piggeries. Piggery development approvals are now accompanied by a comprehensive suite of prescriptive and performance-based design, construction and operational conditions which seek to achieve ESD by minimizing the risk of adverse impacts on the environment and community amenity. While these conditions are generally based on the best available scientific knowledge, because of a lack of relevant Australian research data in many areas of piggery environmental management, some conditions may be based on limited (and sometimes outdated) overseas research which may not always be relevant for Australian production systems and climatic conditions.

The intensive livestock industries have responded to these challenges by providing funding support for targeted research addressing key environmental issues facing the industry. This symposium provides three examples of how industry and government have addressed important issues facing the pig industry. Firstly, using scientific research to evaluate the risk posed to human and animal health by the range of pathogens that are likely to be present in pig waste products; secondly, the quest for better, more cost effective ways to manage effluent, resulting in reduced pond sizes, easier de-sludging and better utilization of nutrient resources; and thirdly, exploring the scope for trialling and implementing new and innovative management practices within the highly regulated Australian pig industry.

In addressing these important issues, the authors have highlighted the results of previous and current research undertaken primarily by the Queensland Department of Primary Industries and Fisheries with funding support from industry, through Australian Pork Limited (APL).

# Food-borne pathogens and pig waste products - the spoke in the wheel?

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## Abstract

Food-borne pathogens are present in normal healthy pigs and thus are also present in pig wastes and by-products. The presence of these pathogens can be viewed negatively (i.e. 'a spoke in the wheel') or as simply another issue that requires the adoption of appropriate guidelines and management procedures. A key component in the development of appropriate, effective guidelines and management practices is a solid basis of knowledge on which pathogens are present as well as the levels of these pathogens. This paper reviews Australian Pork Limited (APL) funded projects carried out in our laboratories that have provided a solid base of Australian data for the pig industry. These data will ensure that pathogens are not 'a spoke in the wheel' but rather an issue - like many others that confront the industry - that can be managed to ensure that there is no unacceptable risk to either public health or the environment.

## Introduction

By-products produced during normal pig production are a valuable resource. As an example, pig effluent (as well as other composted products) is commonly used for agricultural purposes due to the fact that they are high in valuable nutrients. However, in addition to the nutrient rich material contained in wastes, by-products such as effluent, solids and composted material can be a source of food-borne pathogens. These pathogens are normal inhabitants of the pig gastro-intestinal tract and are commonly excreted along with their faeces. These pathogens have been a source of public concern, especially due to their possible link within the food chain.

Are these pathogens the figurative 'spoke in the wheel' of recycling and re-use of valuable nutrients or are they a factor which - while normally present - can be controlled and managed with suitable guidelines?

In a series of industry funded research projects, we have attempted to provide the knowledge and information to ensure that this issue can be effectively managed using Australian data and avoid the problem of a 'pathogenic spoke in the wheel'. This paper will review the broad issues and provide highlights of our research outcomes.

## What are the food-borne pathogens of potential concern?

Based on a comprehensive literature review, we have identified the primary organisms of concern as *Salmonella* spp. and *Campylobacter* spp. In addition, the presence of *E. coli* can be used as an indicator of the truly pathogenic organisms such as *Salmonella* spp. (Blackall *et al.*, 2000). In more recent work, we have specifically looked at the issue of *Arcobacter* spp. - recently recognized as an emerging food-borne pathogen (Chinivasagam *et al.*, in press).

There have also been theoretical concerns raised in terms of Transmissible Spongiform Encephalopathies (TSEs) or prion diseases. A group of infectious neurodegenerative diseases, which cause brain and nerve damage in humans and animals, TSEs occur in many countries, but not Australia or New Zealand (Gavier-Widen *et al.*, 2005). As part of the national program to prevent the entry of TSE agents into the food chain, Australian authorities have ruled that meat and bone meal ingredients cannot be fed as part of ruminant diets. As pigs are resistant to TSE agents (Wells *et al.*, 2003), meat and bone meal are still permitted in pig diets. Hence, there is a need for consideration of how pig by-products (which may contain meat and bone meal) and composts of pig carcasses are used in the environment, particularly in agricultural production systems where cattle are present. In a current industry-funded project, we are developing guidelines to ensure that the pig industry has practices in place to ensure that this theoretical risk is appropriately assessed and managed.

## What are the current processes used to reduce pathogen levels in pig by-products?

In most instances, liquid effluent is stored on site and undergoes simple treatment processes aimed at managing nutrients and pathogen transfer to the environment. Pathogen reduction occurs by the simple methods of settling and other natural microbial interactions conducive to the elimination or reduction in the levels of these pathogens. Solid wastes, such as screened solids, used bedding (litter) and carcasses, commonly undergo a composting process

on site. Composting has a potential to reduce pathogen levels due to the temperatures achieved within the piles as well as the microbial interactions occurring during the various stages of composting. In all these processes there is the potential for the pathogens to transfer to the environment (e.g. via run-off during large rainfall events), a situation which requires effective management procedures. Similarly, there needs to be effective procedures in place to control potential vectors such as birds and wild animals.

### The Australian context

An understanding of the presence of food-borne pathogens in piggery waste and by-products and the level of these pathogens is of vital importance. Knowledge of the levels of the different pathogens allows an understanding of the efficacy of the different treatment processes as well as assisting in quantifying the possible risks during storage and re-use of different waste and by-products. Further, even though not a current risk, the potential risks due to the possible contribution of TSEs - such as bovine spongiform encephalopathy (BSE) - can also be put in to perspective via 'risk assessment processes'.

Piggery environmental issues are subject to both state and national guidelines which are designed to incorporate processes to ensure that adverse events (in terms of human health as well as environmental health) are minimized if not eliminated. A key issue with respect to the development of workable guidelines for the Australian pig industry is to ensure that such guidelines, wherever possible, should be supported by national data relevant to prevailing local conditions. In the absence of such local data, it is possible that international data, which may not reflect national conditions, would be used.

Since 2000 we have been responsible for the delivery of data on food-borne pathogen levels in various pig waste and by-products. The outcomes from these studies support the development of workable guidelines that ensure safe and sustainable re-use applications.

### Pathogens and pig effluent

In our initial work, we examined 13 final stage effluent ponds from piggeries in south-east Queensland to gain an understanding of the levels of key target pathogens and indicator organisms (Chinivasagam *et al.*, 2004). We found that the 13 final effluent ponds contained an average of  $1.2 \times 10^5$  colony-forming units (CFU) per 100 ml of thermotolerant coliforms and  $1.03 \times 10^5$  CFU per 100 ml of *E. coli*. The level of *Campylobacter* varied from none detectable to a maximum of 930 'most probable number' (MPN) per 100 ml. *Salmonella* was detected in the final ponds of only four of the 13 piggeries and then only at a low level (highest level being 51 MPN per 100 ml). No rotavirus and no *Erysipelothrix rhusiopathiae* were detected. The average log<sub>10</sub> reductions across the ponding systems to the final irrigation pond were 1.77 for thermotolerant coliforms, 1.71 for *E. coli* and 1.4 for *Campylobacter*. To put it into context, this represents reductions of approximately 95% in these organisms across these pond systems. Thus this study has provided baseline knowledge on the levels of indicator organisms and selected pathogens in typical Australian piggery effluent.

In seven Queensland piggery effluent ponds, we found that *Arcobacter* spp. levels varied from  $6.5 \times 10^5$  to  $1.1 \times 10^8$  MPN per 100 ml (Chinivasagam *et al.*, in press). As well, in freshly irrigated soils, the *Arcobacter* spp. level varied from  $9.5 \times 10^2$  to  $2.8 \times 10^4$  MPN per g. We identified three species within a representative set of eighty-three isolates - 35% of these isolates being *Arcobacter butzleri*, 49% *A. cryaerophilus* and 16% *A. cibarius*. As *A. butzleri* is generally regarded as being pathogenic for humans, our work has extended our understanding of the risks associated with piggery effluent. This research is a world first both for providing data on the levels of *Arcobacter* spp. in effluent and also for identifying *A. cibarius* in pigs; the organism had only previously been associated with chickens. Overall, *Arcobacter* spp. were isolated from piggery effluent at levels suggestive of good survival in the effluent pond.

### Pathogens and aerosols

In addition to being a source of concern in waste and by-products, pathogens are also a concern in terms of their possible presence in aerosols in areas surrounding pig production facilities. We have completed an industry funded study aimed at understanding the impact of aerosols created when treated effluent is used to flush the gutters in pig sheds (Chinivasagam and Blackall, 2005). The levels of total bacteria and *E. coli* in air inside three piggeries, during normal pig activities, were  $2.2 \times 10^5$  and 21 colony forming units (CFU) per m<sup>3</sup> of air respectively. These levels of organisms are consistent with those previously reported in other studies. We found that flushing with ponded effluent had no marked or consistent effect on the airborne bacteria or *E. coli* levels. It would seem unlikely that any flushing with recycled effluent has a dominant influence on levels of airborne bacteria and *E. coli* in pig sheds.

**Pathogens and composting**

In our current industry funded work, we are studying the levels of food-borne pathogens in a variety of compost types - including 1) pig carcasses; 2) screened solids; and 3) effluent incorporated into a carbon source such as cotton trash. No results are yet available from this work.

**Conclusions**

Our studies - undertaken with industry funding - have provided much needed baseline data on the types and levels of pathogens present in pig wastes and piggery by-products. The data from our earlier studies have already been used in framing national environmental guidelines.

In our view, food-borne pathogens are not 'a spoke in the wheel'. There is no doubt that food-borne pathogens can be present in the raw waste product. An understanding of the types of pathogens and their levels will allow the development and implementation of appropriate management guidelines and practices. Just as the nutrients present in pig waste have to be managed, so do the pathogens.

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# The big pond debate

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## Abstract

Over recent decades, Australian piggeries have commonly employed anaerobic ponds to treat effluent to a standard suitable for recycling for shed flushing purposes and for irrigation onto nearby agricultural land. Anaerobic ponds are generally sized according to the Rational Design Standard (RDS) developed by Barth (1985), resulting in large ponds, which can be expensive to construct, occupy large land areas, and are difficult and expensive to de-sludge, potentially disrupting the whole piggery operation. Limited anecdotal and scientific evidence suggests that anaerobic ponds that are undersized according to the RDS, operate satisfactorily, without excessive odour emission, impaired biological function or high rates of solids accumulation. Based on these observations, this paper questions the validity of rigidly applying the principles of the RDS and presents a number of alternate design approaches resulting in smaller, more highly loaded ponds that are easier and cheaper to construct and manage. Based on limited data of pond odour emission, it is suggested that higher pond loading rates may reduce overall odour emission by decreasing the pond volume and surface area. Other management options that could be implemented to reduce pond volumes include permeable pond covers, various solids separation methods, and bio-digesters with impermeable covers, used in conjunction with biofilters and/or systems designed for biogas recovery. To ensure that new effluent management options are accepted by regulatory authorities, it is important for researchers to address both industry and regulator concerns and uncertainties regarding new technology, and to demonstrate, beyond reasonable doubt, that new technologies do not increase the risk of adverse impacts on the environment or community amenity. Further development of raw research outcomes to produce relatively simple, practical guidelines and implementation tools also increases the potential for acceptance and implementation of new technology by regulators and industry.

## Introduction

Over recent decades, Australian piggeries have commonly used anaerobic ponds to treat effluent discharged from conventional pig production sheds. These ponds have several advantages over higher technology treatment options including relatively low construction costs and moderate ongoing management requirements. The treated effluent is generally considered suitable for recycling for shed flushing purposes and for irrigation onto crop or pasture. Most piggeries are located in rural areas where agricultural crops are grown either on the piggery property or on nearby properties. Consequently, it makes sense to use the valuable water and nutrient resources in the treated effluent as an organic fertiliser and soil amendment to promote crop growth, rather than treating the effluent to a higher standard (for example, by removing nutrients) for discharge/disposal into the environment. In this way, the nutrients in the effluent are effectively recycled into crops that may be harvested for use as pig feed or for other purposes. This practice also reduces the requirement for synthetic fertilisers.

## Rational design standard

Throughout Australia, the current design standards used for sizing anaerobic treatment ponds are generally based on the Rational Design Standard (RDS) developed by Barth (1985) in the United States. This standard appears to have been developed primarily to limit odour emission from anaerobic ponds, based on the principle that higher organic loading rates result in higher odour emissions. Anaerobic pond loading rates are generally expressed in terms of the mass of volatile solids (VS) discharged into the pond on a daily basis, per cubic metre of pond volume available for effluent treatment. In Australia, the RDS generally results in relatively large anaerobic treatment pond volumes (Table 1), ranging from 7.7 m<sup>3</sup>/standard pig unit (SPU) for cool climates to 6.0 m<sup>3</sup>/SPU for hot climatic localities (Tucker *et al.*, 2004).



**Table 1. Anaerobic pond volumes and dimensions determined using the RDS, for a range of Australian climates and de-sludging intervals**

Climate	De-sludging interval (years)	Pond storage volume <sup>1</sup> (m <sup>3</sup> )	Pond side length at top <sup>2</sup> (m)	Pond side length at base <sup>2</sup> (m)	Pond volume per SPU <sup>1</sup> (m <sup>3</sup> /SPU)
Cool (k=0.60)	1	22,306	78.9	53.9	4.5
	2	24,079	81.5	56.5	4.8
	5	29,397	88.8	63.8	5.9
	10	38,259	99.7	74.7	7.7
Warm (k=0.80)	1	17,173	70.7	45.7	3.4
	2	18,946	73.6	48.6	3.8
	5	24,263	81.8	56.8	4.9
	10	33,126	93.6	68.6	6.6
Hot (k=1.00)	1	14,093	65.1	40.1	2.8
	2	15,865	68.4	43.4	3.2
	5	21,183	77.2	52.2	4.2
	10	30,046	89.7	64.7	6.0

<sup>1</sup>Pond volumes based on 5000 SPU piggery (equivalent to 500 sows farrow to finish); <sup>2</sup>pond dimensions based on square shape with storage depth of 5 m and 1:2.5 (vertical to horizontal) batters on all four sides.

It is acknowledged that anaerobic ponds designed in accordance with this standard generally function effectively, with relatively low to moderate ongoing odour emissions, over their design lifetime, which generally ranges from 2-10 years. However, once the sludge build-up on the bottom of the pond starts to encroach on the design pond treatment volume, pond function may be adversely affected. This may increase odour emission and the effluent may become unsuitable for use in flushing sheds and for irrigation onto agricultural land, due to higher total solids concentrations. At this time, producers are faced with the major problem of determining how to de-sludge a relatively large pond while keeping the piggery operating.

### Pond de-sludging

Methods commonly used for de-sludging anaerobic ponds include:

- Long reach excavator;
- Vacuum tanker and mechanical stirrer;
- Solids handling pump;
- Drag line;
- Breach pond bank and retain sludge in a shallow, bunded drying bed; and
- Abandon pond and construct a new one.

These de-sludging methods are generally costly, disruptive to normal piggery operations and not always successful in removing a large proportion of the accumulated sludge. Contractors with suitable equipment are relatively uncommon in many parts of Australia and often difficult to source when required.

Excavators generally have a maximum reach of approximately 18 metres. This is clearly insufficient for the pond dimensions shown in Table 1 for a moderate sized piggery (500 sows, farrow to finish). Furthermore, the slurry removed from the pond is generally too wet for immediate spreading onto land. Consequently, it must be either transported to a suitable drying bed or placed directly into a bunded drying area near the pond, within reach of the excavator arm.

Vacuum tankers seem to work reasonably well in relatively small ponds where a mechanical agitator, generally a tractor power take off (PTO) driven propeller, is used to resuspend all the solids with the liquid in at least part of the pond. The resulting slurry is pumped into a tanker which may be self propelled or towed behind a tractor. The tanker is used to transport and spread the sludge on a suitable area of cultivation or pasture, negating the need for double handling of the sludge. However, if sufficient suitable agricultural land is not available in close vicinity to the pond site, the tanker turnaround time increases, resulting in high costs, as most contractors are paid on an hourly rate.

Some producers have hired large solids handling pumps, similar to those used in the mining industry, to pump sludge from effluent ponds. They generally need to be suspended on an excavator arm or a floating pontoon so that they can be moved around the pond; otherwise they only effectively remove solids in a relatively small, coned shaped area around the pump. This de-sludging method also requires the availability of a bunded sludge drying bed or some form of irrigation system capable of spreading the sludge over a sufficient area of agricultural land. A north Queensland-based pond de-sludging contractor uses a custom made 'big gun' irrigator to apply pond sludge to land, by delivery through 250 m x 100 mm diameter lay-flat hose.

While drag line contractors are relatively rare, one such contractor was offering pond de-sludging services in south-east Queensland over recent years, primarily servicing the beef feedlot industry. This method of pond de-sludging also requires a suitable bunded area for the storage/drying of the wet slurry removed from the pond.

Pond breaching is generally only feasible for ponds constructed on sloping land where the base of the pond is at or above the ground level on the lower side of the pond. A suitable bunded area for collection and storage/drying of the sludge is also required below the pond site. Major earthworks are required to breach and reinstate the pond embankment.

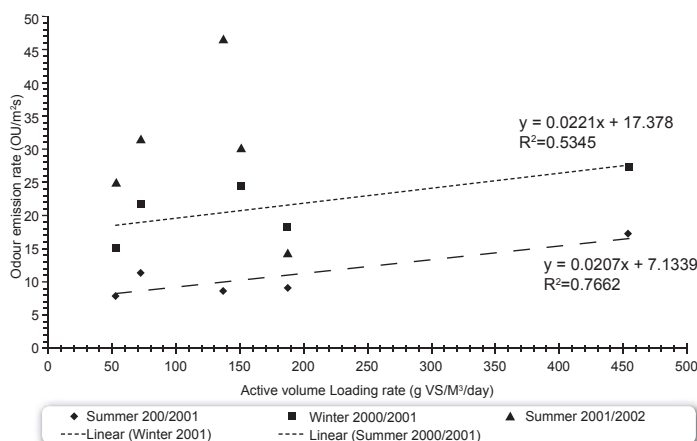
Abandoning a sludged-up effluent pond may be an option at sites where there is sufficient land to construct a new pond. However, this would result in significant additional earthworks costs. It is likely that the abandoned pond site would eventually require remediation and the removal of dried solids to minimize the risk of any subsequent environmental harm.

### Alternative effluent system design strategies

Limited anecdotal and scientific evidence suggests that piggeries with anaerobic ponds that are undersized according to the RDS do not emit excessive odour or exhibit impaired pond biological function resulting in high rates of solids accumulation when compared to ponds designed based on the RDS. Furthermore, treated effluent from undersized ponds does not appear to be unsuitable for shed flushing or irrigation onto agricultural land.

As a consequence of these observations, this paper questions the validity of rigidly applying the principles of the RDS when designing ponds for the anaerobic treatment of piggery wastes in Australia. It is suggested that a number of alternate design approaches may be followed, resulting in smaller ponds that are easier and cheaper to construct and manage, particularly if they require clay or synthetic lining to minimize the risk of groundwater contamination. In addition, odour emissions from these ponds may be similar to, or even lower than, those generated by ponds designed using the RDS.

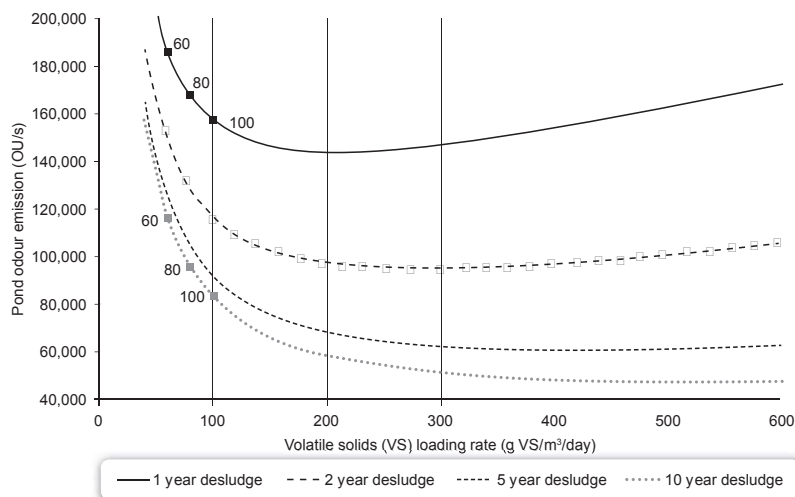
Using dynamic olfactometry, Hudson *et al.* (2004) measured odour emissions from anaerobic ponds at five Queensland piggeries over three seasons. The active volume loading rates in these ponds ranged from 53-454 g volatile solids (VS)/m<sup>3</sup>/day, or up to 5.5 times the loading rates recommended by the RDS. The measured odour emission rates per square metre of pond surface area are plotted against the active volume loading rates in Figure 1. Trend lines for two of the three seasons (Summer 2000/2001 and Winter 2001) clearly show similar linear increases in odour emission with increasing loading rates. Data for the third season (Summer 2001/2002) did not show any clear trends. The highest odour emission rate increase occurred during Winter 2001. Based on the trend line equation for this season, a 350% increase in loading rate produced a 39% increase in odour emission. It was also apparent that seasonal variations in odour emission were at least as great as those due to VS loading rate.



**Figure 1.** Relationship between piggery effluent pond odour emission and volatile solids (VS) loading rate, based on data reported by Hudson *et al.*, 2004

Based on the trend line equation for Winter 2001 (Figure 1), total pond odour emissions were determined for a range of active volume VS loading rates, along with corresponding pond volumes and dimensions. For these scenarios, it was assumed that the ponds were square in shape, with total depths of five metres and 1:2.5 (vertical:horizontal) batters, to enable the calculation of pond surface areas and total odour emission rates.

Pond dimensions and odour emissions were calculated for four common pond de-sludging intervals, ranging from 1-10 years. The resulting total pond odour emission rates are plotted against loading rates in Figure 2.



**Figure 2.** Pond odour emissions calculated using the Winter 2001 trend line equation from Figure 1 plotted against pond active volume loading rate, for a range of pond de-sludging intervals. The pond depths have been assumed to be 5 m with 1 vertical: 2.5 horizontal batters

Figure 2 indicates that the total pond odour emission initially decreases as the loading rate increases above the normal RDS design rates of 60, 80 and 100 g VS/m<sup>3</sup>/day, for cool, warm and hot climates, respectively. However, the odour emissions increase as the loading rates increase above approximately 220 and 300 g VS/m<sup>3</sup>/day for ponds having 10 and five-year de-sludging intervals, respectively. There is minimal or no increase in odour emission for the one and two year de-sludging interval ponds because of the smaller overall pond volumes and surface areas emitting odour.

Figure 2 suggests that overall pond odour emission could actually be decreased by designing ponds with significantly higher loading rates, even for the longer (five-and 10-year) de-sludging intervals. The resulting reduction in pond volume and surface area more than compensates for the relatively small increase in odour emission per square metre at the higher loading rates. Other advantages of the resulting smaller ponds include less expensive construction and easier (but perhaps more frequent) de-sludging.

It must be acknowledged that the relationship between VS loading rate and odour emission has not currently been clearly established. The relationships presented in Figure 1 are based on limited data collected over three seasons and the data from one of these seasons did not provide any clear trends. Nevertheless, anecdotal evidence supports the concept of smaller, more highly loaded ponds.

### Permeable pond covers

Hudson *et al.* (2007) found that a polypropylene and shade cloth cover reduced piggery effluent pond odour emission rates by 50% compared to an uncovered pond. The nature of the odour emitted from the covered pond surface was also found to be quite different, further reducing the overall impact of the pond odour emissions. This suggests that by using a permeable pond cover, the loading rate could be increased substantially without increasing the total pond odour emission. The increased loading rate would reduce the required pond storage volume, resulting in a smaller surface area and reduced overall pond odour emission.

A permeable pond cover could be used as further 'insurance' against higher odour emissions from ponds loaded at rates higher than those determined using the RDS, as discussed in the previous section.

### Solids separation

Watts *et al.* (2002) investigated various methods for separating solids from piggery effluent streams, amongst them the use of gravity run-down screens, vibrating screens, screw presses, trafficable gravity settling basins, sedimentation and evaporation pond systems (SEPS) and rotating screens. By removing solids from the piggery waste stream, the treatment and sludge storage volumes of subsequent anaerobic ponds can be reduced, while still complying with the RDS loading rates. The resulting reduction in pond volume and surface area should reduce the overall pond odour emission. Subject to odour modelling, this may allow the development or expansion of a piggery at a site where current separation guidelines indicate that there is insufficient separation distance to nearby houses and/or towns. The other advantage is that the separated solids may be exported from the piggery property, thereby reducing the land area the applicant requires for sustainable application of piggery effluent. This may allow the establishment of piggeries on smaller properties.

In a group demonstration trial, Skerman and Collman (2006) found that highly loaded piggery solids settling ponds effectively removed 50-90% of the total solids and volatile solids discharged from a conventional 600 sow farrow-to-finish piggery in southern Queensland. The cost of establishing settling ponds is likely to be significantly less than the cost of installing more sophisticated mechanical solids separation equipment. However, the separated solids may be more difficult to manage because of their higher moisture content. Because group demonstration trials are preliminary in nature, the above results were based on limited data and there were some unresolved issues in relation to odour emissions and the rate of solids accumulation in the settling pond. The highly loaded settling pond approach is currently being examined in more detail and preliminary results suggest similar levels of solids removal to those found by Skerman and Collman (2006).

The settling pond being investigated in APL project 2108 is continuously crusted over due to the relatively high loading rate. The project will investigate whether the crust decreases odour emission in a similar manner to a permeable pond cover. Abattoir effluent ponds in Australia are commonly managed to maintain a crust as an odour management practice. This practice may also be applicable in the Australian pig industry.

### Highly loaded bio-digester incorporating an impermeable covered pond

This type of system may be used to recover bio-gas for use in piggery heating, power generation or to run an internal combustion engine for other purposes. Mechanical mixing and artificial heating of the pond effluent may be used to increase the efficiency of biogas recovery. Alternately, rather than reusing the gas emitted from the pond, gaseous pond emissions could be treated in a biofilter, before flaring or direct release to the atmosphere. This may be an effective method for addressing pond odour emission.

Dunlop *et al.* (2005) carried out preliminary trials using a biofilter to treat gaseous emissions from unsealed static pits located under commercial piggery sheds in southern Queensland. Using olfactometric assessment, they found that the biofilter reduced odour concentration by approximately 42%. Ammonia removal ranged from 80-95% while the concentrations of several odorants were significantly reduced. The researchers also reported that the nature of the odour exiting the biofilter was less offensive than normal piggery odour, smelling more like moist grass or earth. While this situation is not identical to a covered, highly loaded anaerobic pond, similar results could be expected.

In addition to limiting odour emission, a covered bio-digester could significantly reduce the emission of greenhouse gases, primarily methane. This objective will inevitably become more important as all industries face increased social, political and economic pressure to limit greenhouse gas emissions.

Covered pond bio-digesters would undoubtedly involve significantly higher capital and ongoing operational expenses than some of the other options mentioned previously. There are currently very few (if any) of these types of systems operating in Australian piggeries. However, APL is coordinating the installation of several pilot bio-gas recovery plants at commercial piggeries across Australia, to investigate the practicalities of establishing and operating this type of system. If greenhouse gas recovery incentives or regulatory requirements become more stringent in the future, this type of system may become more common-place.

### Regulatory agency acceptance of alternate technologies

As government entities, regulatory agencies have clear responsibilities to ensure that new developments do not impact adversely on the environment or community amenity. However, they are also obliged to embrace new technology that could potentially benefit industry, provided there is sufficient credible, scientific evidence suggesting that the new technology will not result in any significant detrimental impacts. The most immediate environmental risk associated with most of these alternate technologies is the possibility that nearby residents could experience increased odour nuisance.

Clearly, if regulatory agencies perceive an unacceptable level of uncertainty regarding the long-term performance of these technologies, they will not risk the possibility of any increased environmental harm or nuisance and the accompanying social and political fallout that may result. In some cases, regulatory agencies have preferred (and even insisted on) potential developers adopting 'tried and proven' methods, despite significant evidence supporting the effectiveness of alternate technologies. This reluctance to embrace new technologies is at least partly due to the failure of researchers, piggery developers and industry to package the information supporting new technologies in a manner that assists regulatory agencies in their decision making processes.

This highlights the need for researchers to liaise closely with both industry and regulatory agencies in identifying issues for investigation and preparing research project proposals. This will ensure that the experimental design is capable of providing results that will satisfactorily address any risks or uncertainties of concern to regulatory agencies. Ongoing liaison throughout the course of the research also provides opportunities to revise the experimental method to address any emerging concerns.

It is not adequate to present raw research findings to regulatory agencies without reasonably simple, practical guidelines and technical tools to assist in the understanding, adoption and implementation of the new technology. This highlights the need for applied development of raw research outcomes and findings, to produce practical solutions to real industry issues, and to address obstacles to adoption of new technologies by regulatory agencies.

## Conclusions

The RDS results in large anaerobic effluent treatment ponds. These ponds can be expensive to construct, particularly if they require clay or synthetic lining, and they occupy large areas of land. While they generally operate effectively with minimal management, up until their function becomes impeded by the accumulation of sludge (generally after 5 to 10 years), they are difficult and expensive to de-sludge, potentially disrupting the whole piggery operation.

Based on limited pond odour emission data presented by Hudson *et al.* (2004), there appears to be considerable scope for loading ponds at higher rates than those determined using the RDS. Despite relatively small increases in the odour emission rate per unit area, overall pond odour emissions are expected to decrease as the pond volume and surface area decrease.

Other options such as permeable pond covers, various solids separation methods (including highly loaded gravity settling ponds) and highly loaded bio-digester ponds with impermeable covers could potentially be used to reduce pond volumes. Some of these options may be used in conjunction with biofilters and/or systems designed for biogas recovery and use.

To ensure that new effluent management options receive endorsement from regulatory authorities, it is important for researchers to consider and address both industry and regulator concerns and uncertainties regarding new technology. Researchers should also demonstrate, beyond reasonable doubt, that new technologies do not increase the risk of adverse impacts on the environment or community amenity. The applied development of raw research outcomes to produce relatively simple, practical guidelines and implementation tools will increase the potential for acceptance and implementation of new technology by regulators and industry.

# Environmental regulation in the pork producing industry - bring on the research!

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## Abstract

Over the last 20 years, environmental management in Queensland has moved from the policy backwaters of government to the front line of operations by way of regulatory enforcement, industry programs and incentives. When the new Queensland Environmental Protection Act 1994 (EPA) came into effect, the business of environmental management has become a central feature of urban and rural development activity. The concept of environmentally sustainable development (ESD), has given life to the precautionary principle as a way for planners and regulators to place relevant controls on development. The planning, development and operation of pig farming systems has been effected by the new regulatory framework. Ever more definitive standards and approval permits have emerged which endeavour to achieve ESD. With these modern planning instruments in place, rural industry sectors have become, quite legitimately, concerned about future opportunities for research and innovation. This paper asserts that the capacity to engage in research and to achieve innovation in the pork producing industry is not hindered by Queensland environmental regulation frameworks. However, in order for research and innovation to prosper within these frameworks, some protocols need to be followed by the industry. What is at stake is community confidence.

## Introduction

The last twenty years have witnessed the strong emergence of environmental planning and development legislation in response to growing community expectations of land management and natural resource utilization, in the name of achieving ESD. According to WME Environment Business Magazine (Anon., 2007), ESD principles are embedded in some 130 pieces of legislation across Australia. ESD principles can be found in countless non-statute government documents such as policy statements, industry action plans, codes of practice and local and regional management schemes. The term 'environmentally sustainable' is the guiding aim of the National Environmental Guidelines for Piggeries (Tucker *et al.*, 2006).

A plethora of standards and codes, such as the National Guideline which set out to define ways to achieve sustainable operation and seek compliance with community and legislative requirements, followed the development of apparent universal adoption of ESD. This has narrowed the options for what a pig farmer, operating under the terms of an approval, can do in terms of experimentation and research.

## Science, development assessment and community perception

Government regulatory agencies are given the task of administering Acts and regulations strictly in the manner intended by the legislators through parliament. In Queensland, planning legislation – i.e. the Integrated Planning Act 1997 (IPA) - requires development to be carried out in a manner that achieves ecological sustainability. The agencies involved in assessing any particular proposal will depend on the nature of a particular development proposal and other circumstances such as local planning scheme requirements and jurisdictional matters outlined in the IPA.

The Intensive Livestock Environmental Regulation Unit (ILERU) of the Queensland Department of Primary Industries and Fisheries (DPI&F) is guided by the IPA when assessing an application to develop a new or expanding pig farming activity. The IPA cites the EPA. The EPA requires an assessor to consider ESD, among other values and standards in the EPA known as the standard criteria. This is the authorizing environment in which ILERU officers carry out the business of assessing and monitoring compliance of piggery development

Environmental Scientists within ILERU consider the most detailed and accurate information available about a development proposal before making a decision. Most importantly, a development proponent has the primary responsibility for presenting to ILERU the most relevant and comprehensive information available to describe the development completely and to accurately predict the total potential impact on the receiving environment. The ILERU staff will always take a proactive role in providing technical assistance or advice for proponents or their consultants to work through issues such as the use of new technology or alternative predictive tools, long before a development decision is due. 'Pre-lodgement meetings' are encouraged and could include DPI&F and local government planners even before the proponent hands over any application fees.

Assessment predictive tools such as odour dispersion or hydrological models are inherently complex. It is essential these tools are professionally accepted through widespread peer and industry review so their use can be defended in public and planning court arenas. Proponents need to ensure that before embarking on a development application, predictive tools used are accepted by the assessing agency because they are technically creditable and have sufficient validation. Assessing agencies support industry investment in any research that may produce validated assessing tools in the first instance or operational technologies that can achieve even higher levels of certainty in the decision making processes of agencies.

Behind all of the innovative technology and predictive tools there exists levels of scientific and experiential knowledge of varying degrees. In the community arena risk communicators will confirm that selling science to a sceptical public can be harder than 'selling coal to Newcastle'. The task is especially difficult where an issue involves the prospect of imposing change on a community and where the community senses a limited ability to influence the outcome. Arguing the science was not sufficient to convince enough ratepayers of Toowoomba that piping recycled water into the source of their water supply was an acceptable practice. The science asserted that the quality of the treated effluent was so good it would be sufficient for use in dialysis machines yet the community did not accept the proposal.

Residents of Narangba, north of Brisbane, have for some years been campaigning against a proposal for a food irradiation plant to be built in their area. Irradiation using Cobalt 60 is a fifty year old technology with the first commercial industrial plant in the world being built here in Australia. The process technology is remarkably simple. It involves the storage of Cobalt 60 in a heavily fortified concrete room containing a pool of water. A conveyor belt system directly above the water carries products that become irradiated with gamma rays when the rods are elevated out of the water. According to Queensland Health, Cobalt 60 presents no danger to the community or the environment when properly managed. However, in the public domain the voice of science and government has had to compete with a plethora of claims, sometimes emotive, still condemning the proposal, with allegations ranging from the production of radioactive food products through to nuclear terrorism.

Communities make their own judgements on the information presented about a development proposal and the science, no matter how simple or complex, will be only one of the considerations. In the case of piggery development, a range of predictive tools and standards have been adopted that theoretically predict the environmental impact that could be expected. The science that underpins these concepts is important, but few would assert the science is perfect. For ILERU, on-ground experience has played an important role over time. The guidelines ILERU uses to calculate separation distances were originally based on the best available local research. The final validation of those original decisions really came later when it was found that environmental nuisance was indeed being avoided through the use of separation distances based on those guidelines. For the community at large, this has been a successful outcome. For the pork producing industry the question remains - could the separation distances be reduced in certain circumstances or can innovation, open up new commercial opportunities without generating nuisance?

### **Research and innovation in a regulatory environment**

Commercially oriented development applications typically involve intentions to maximize investment returns through maximizing production potential from a given resource at a given site. This has the natural effect of pushing the boundaries of sustainability and this is an issue that confronts development assessors on a continual basis. Where the margins for error become tighter, the appropriate response is for the assessor to rely more heavily upon the most tried and tested measurement tools to predict the potential impact of the development. The more marginal the development proposal appears in terms of environmental compliance, the more likely it is that a conservative outcome will be required from the assessment process. The same would apply where lesser-known process technologies are presented in a development application. This is where the precautionary principle becomes particularly influential.

Improving environmental performance in piggeries today will result from specialized technically based marginal gains rather than quantum improvements. Where innovation is mixed with commercial intentions there is a relatively greater chance for the assessment agency to apply the rigour necessary for the agency to have confidence in its decision. Once a development is approved and established there are few options open to a community that is being effected unreasonably. This is of course part of the challenge facing contemporary researchers and regulators everywhere.

In the example of an existing piggery development, the matter of operating conditions and the potential for the conditions themselves to restrict research and innovation must be considered. In Queensland many lawfully existing piggeries are bound by a suite of operating conditions, while others are not bound by any conditions at all. Generally speaking, the difference is distinguished by the era in which the piggery was established. Piggeries with environmental approvals attached are typically more recently established or changed in some way.

That operating conditions could stifle commercial productivity, research or innovation is an argument supporting a performance-based approach. In such an approach the assessing agency minimizes its influence over how an activity actually operates at a basic level - by conditioning an approval only to clarify overall environmental performance targets. In this situation, an operator would be free to engage in all manner of trial and error research and innovation at an operational level because there would be no prescriptive conditions to conflict with. So long as control measures of whatever description were put in place to prevent unacceptable outcomes and breaches of those overall targets, the operator could engage in research trials and achieve desired innovations.

The performance-based approach could thus give producers the flexibility to conduct on-site research at their complete discretion. However, despite there being these advantages in performance-based approaches, such an approach can be difficult for a regulator to enforce, it has no widespread community support and is often not desired by developers themselves. Performance-based approaches as an alternative have been widely debated but agreement is elusive. For piggery development in Queensland, fundamental issues such as the precise location and nature of the piggery infrastructure and the scale of operation are matters that really require a degree of prescription in approval conditions. In order to maintain public confidence this sort of information needs to be stated at the application stage for advertising and scrutiny. The information is then defined when approvals are written. The EPA provides that an assessor may prescribe conditions, plant and equipment to be used and particular measures to be taken by an operator in order to achieve compliance with the EPA. This is the approach taken by ILERU for compliance with this Act. In reality the conditions typically imposed by ILERU are a mix of performance-based and prescriptive approaches.

In Queensland, pig farming on land is legitimized through the existence of planning approvals known as development approvals. These approvals attach to the land on which a pig farming activity is situated. The approvals typically authorize the activity at a certain scale, and where conditions exist, a piggery must operate within the bounds of those conditions. Quite a substantial number of Queensland pig farms pre-dated current planning and environmental laws. Generally speaking, activities that existed prior to the new laws were permitted to continue operating at the pre-existing scale. Where conditions do not exist piggery operators must still comply with the 'general environmental duty'.

The absence of specific operational conditions makes the prospect of carrying out research activities or introducing innovation potentially more feasible because there is no structured operating format to comply with. In these circumstances there is scope to adjust existing waste management systems or other operations. However, there is a limit to how much 'adjustment' can be made. Queensland planning legislation prescribes that any material change in the intensity or scale of an 'environmentally relevant activity' such as pig farming, may require a development approval to permit that change. Building work is another activity that requires approvals. Installing a new technology effluent treatment system could be considered a material change in use where the installation may, for example, create a new point source of odour that may, in turn, adversely affect neighbouring occupancies. Any proposed material change in an intensive livestock activity that could have a negative impact off-site is an issue that communities expect to be examined by the appropriate authority prior to the change being allowed. This is a fundamental tenet of planning laws.

Where a research project is proposed at a piggery, the legislation asks will the research activities cause a 'material change of use' of the piggery activity. A research project manager could approach the local government or, in the case of pig farming research, the ILERU for advice. It is possible that the research will not trigger this type of change, but it would be important for this matter to be resolved before any valuable research money is spent.

### **Cart before the horse?**

In 2004, in response to industry concerns about the perceived inflexibilities of embracing innovation within the development assessment process, the manager of ILERU engaged in discussion with Queensland pork producers about alternative ways of validating innovative practices for real life development proposals. The suggestion was made by industry to fast-track the acceptance of new technologies in pig farming development applications through collaborative analysis and prior agreement. The proposition was for experts from industry, academia and government (researchers and regulators) to form a group to peer-review innovative technologies for potential incorporation into new or expanded piggery development applications. The group would in theory seek agreement and this could allow regulatory assessors to more readily accept a new technology as being effective in achieving environmental standards (e.g. to reduce separation distances or to increase pig unit numbers).

Initially the proposition appeared to areas of merit. However, following further consideration and consultation, it soon became apparent that there were a number of significant problems in actual application. The most important, from a regulator's point of view, was that the peer review process could be perceived as being unreasonably influential to the development assessment process. To ensure the development assessment process maintains rigour, an assessor in a 'concurrency agency' assessment role must not be unduly influenced by a third party that has no statutory legitimacy. If an assessor was found to be influenced, this could be catastrophic in terms of community confidence. Such a group, if



formed, could also be viewed as not having the interests of the wider community completely in mind, given that there were to be no 'community members' involved in the group. Finally there would be logistical problems with setting up the group: what would the membership be, would members be paid and by whom, how often would the group meet and how could its decisions be robust enough to be accepted by the wider community, scientific or otherwise. There would also be the matter of reconciling the timing of the meetings and its outcomes with the actual submission of a development application incorporating the relevant innovation. In the end, the proposal was not progressed.

## Conclusion

The development planning and assessment regime in which ILERU operates is intentionally rigorous in process and precautionary in attitude. The planning legislation that applies to piggery development applies equally to shopping centre, oil refinery, and housing development. Embodied within the regime is the principle that development permits are issued to prescribe a certain type of development - a type that has undergone the process of assessment and scrutiny and that has allowed to be established at a place in a manner and form that can achieve compliance with environmental and community standards.

Assessing agencies refer to the best available information and use the best available measuring tools in order to predict and analyze in entirety the potential impact on surrounding environments. At times a new technology may be presented to an assessor that falls outside the usual convention. An assessor has an obligation to predict and analyze the impacts of the new technology. If common assessing tools are not up to the task, the assessor may require the proponent to present information that can assist the assessor in understanding the science behind the performance. The ILERU will always be in a position to engage with proponents and their consultants with a view to collectively seeking this understanding in order for considered decisions to be reached. Simple is good because if technology is not easily understood it will not only be a challenge for the regulator - the developer will also have the wider community to contend with.

Research and innovation can prosper within the current regulatory environment but communication with regulatory agencies is essential. Older piggeries where no operating conditions exist are likely to be places where more operational flexibility is possible in the name of research. Research projects are more likely to fall under the regulator's radar where they are carried out at piggeries that do not have tight margins in terms of separation distances to housing or sensitive environments, or where a developer has maximized the use of on-site resources for commercial or other reasons. The key is to consult with ILERU during the site selection process so that advice can be given about risk factors and compliance with operating approvals. The local government planning authorities should be involved in the early consultation process, as most development decisions concerning intensive livestock development ultimately rest with councils. In starting this consultation process early, and before the research dollars are spent, industry and regulators can benefit from research and development outcomes and work towards maintaining community confidence.

## Symposium – effluent management

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### Conclusions

The intention of this symposium was to address one of the more challenging environmental issues facing the pig industry - effluent management - using contributors to cover a very diverse range of expertise and backgrounds.

In the first area - food-borne pathogens and effluent - Chinivasagam and her colleagues used the analogy of a 'spoke in the wheel' to look at the issue. A 'spoke in the wheel' can be viewed in a negative sense (something that blocks the operation of the wheel) or in a positive sense (something that is actually part of the operation of the wheel). There is no doubt that there is a deep concern about food-borne pathogens and their impact on both the public and the general environment. The work of Chinivasagam and her colleagues - funded by the pig industry - has been a major provider of critically important baseline information. The pig industry and the regulators now have a good knowledge of both the pathogens present in piggery effluent and, most importantly, their level. As noted in the paper, food-borne pathogens will be present in piggery effluent. However, the reality is that these organisms are part of the normal bacterial population and can be managed by appropriate guidelines. Clearly, the 'spoke in the wheel' is a positive and supportive one!

In the second challenging area - termed the Big Pond Debate - Alan Skerman has questioned the validity of a rigid application of the Rational Design Standard (RDS) to design ponds for anaerobic treatment. Work performed by the Queensland Department of Primary Industries and Fisheries - again with pig industry funding - has provided some data, albeit limited, which suggests that ponds can be loaded at higher rates than those determined by RDS without greatly increasing odour emission rates. With pig industry fund, the DPI&F has also at various additional options (pond covers, solid separation methods, highly loaded bio-digestors) which could reduce pond volumes. Skerman makes the clear point that both researchers and regulators have responsibilities in the adoption of new effluent management options. The researchers need to work closely with both industry and regulatory authorities to ensure that new technologies are validated at a level that ensures regulatory confidence in the technology. The researchers need to ensure that research moves beyond the raw state to a level of practical outcomes that will encourage the regulatory authorities to embrace new technologies.

In the final controversial area, Cowan has provided an insight from the regulatory viewpoint of how to balance two viewpoints - the need for on-farm environmental research and the need for farms to stay within environmental regulation frameworks. A common theme of all the papers, and a key point identified by Cowan was the need for communication. There is clearly a great interest by regulators in the progress of innovative environmental research - ('Bring on the Research!' is a frank and supportive message). The industry, the funding bodies and the researchers need to work closely with each other and the regulators.

Overall, this symposium has added some light to an area where the debate is often more marked by 'heat' than 'light'. All three speakers have been positive, innovative and have provided clear pathways to a better future. Our researchers are clearly committed to sound science that - in partnership with industry and regulatory authorities - delivers effective outcomes in terms of innovative effluent management options. While maintaining their rigorous and precautionary principles, the regulatory authorities are clearly keen to see the progressive, innovative, on-farm research.

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# 5

## Dietary supplements and disease control

## Digestible energy, starch and cyanide content of sun-dried cassava residue in Vietnam

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Soyaiya and Omole (1983) showed that cassava meal and cassava peel meal can be used in pig diets. In Vietnam, cassava is used as an animal feed and also to produce flour for human nutrition. Processing plants are used to extract high value cassava starch for flour production while the wet cassava residue is sun-dried to remove any cyanide (HCN) present and used to produce animal feed. However despite its availability and relative cheapness, feed mills are reluctant to increase the use of dry cassava residue due to concerns over possible cyanide toxicity in pigs and limited information on its nutritive value. In the current study we analyzed the cyanide content of cassava residue samples from 16 Vietnamese mills and also assessed the energy value of the sun-dried cassava residue.

We assessed the digestible energy (DE) value of a basal diet (based on 90% maize and a 10% basal component consisting of casein, vitamins, minerals, lysine and oil) and an experimental diet that replaced 30% of the basal diet with cassava residue. Four entire male Large White pigs (about 22 kg) were housed individually in metabolism crates. Total faecal collections were carried out over five days following an initial eight day pre-collection diet adaptation period. Ferric oxide was added to the diet as a faecal dye to indicate the start and end of collection. There were two replicates in time for each experimental diet using a crossover design.

The cyanide content in the cassava residue following traditional sun-drying was reduced to about 4 mg HCN/kg, well below the generally accepted target of 50 mg HCN/kg of diet (Table 1). The resultant dry matter of the sun-dried cassava residue was 89.1%. The pig digestibility study indicated that the faecal digestible energy content of cassava residue was still relatively high. Further chemical examination revealed that a large proportion of the starch present in cassava root still remained even after processing, indicating that the extraction process for cassava starch production was inefficient.

**Table 1. Nutritive and cyanide content of dried cassava residue**

	Dry Matter (%)	Cyanide mg/kg DM	Starch (%)	GE MJ/kg DM basis	DE MJ/kg as fed
n=	80	80	5	5	4
Cassava residue	89.1	3.66	61.6	17.2	11.5
SD	0.99	2.24	4.83	0.35	0.78

This study indicates that cassava residue could be a potentially useful energy source in pig diets. Under current processing systems the residual cyanide has been substantially reduced and should pose little risk even when dried cassava residue is used at higher than current inclusion levels in pig diets in Vietnam (currently 8-10% inclusion).

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## Increasing ractopamine levels in finisher pig diets improves growth performances in light, medium and heavy gilts

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The  $\alpha$ -agonist ractopamine (RAC) has been approved in Australia since January 2004 as a dietary ingredient to improve the production efficiency in pigs. Dose response studies have suggested that daily gain is maximized at 5 ppm RAC although carcass gain may be maximized at higher doses (Watkins *et al.*, 1990; Armstrong *et al.*, 2004). Also, many of the studies have been carried out with heavy slaughter weights more suitable to US than Australian and New Zealand markets. Therefore, the objective of this study was to determine the dose response to RAC in light, medium and heavy gilts.

Ninety-six individually penned pigs were assigned to a 3x4 design with the respective factors being starting weight - Light (65kg), Medium (80 kg) and Heavy (95 kg) - and dietary RAC - 0, 5, 10 or 20 ppm RAC (Paylean<sup>®</sup>, Elanco Animal Health Pty Ltd) for 28 days. All diets were formulated to contain 0.62 g available lysine/MJ DE and 13.9 MJ/kg. Pigs were weighed at -7, 0, 7, 14, 21 and 28 days and voluntary feed intake (VFI) determined at day 7, 14, 21 and 28 days. Backfat at the P2 site and leg fat were determined with an ultrasound on days 0, 14 and 28. At the end of the study pigs were slaughtered and the carcass and P2 backfat determined. Data were analyzed by analysis of variance.

ADG increased linearly ( $P=0.006$ ) but not quadratically ( $P=0.25$ ) with increasing dose of RAC and increased linearly ( $P=0.004$ ) and quadratically ( $P=0.034$ ) with starting weight (Table 1). Likewise, there were linear but not quadratic responses to dietary RAC ( $P=0.004$  and 0.61, respectively) and starting weight ( $P=0.001$  and 0.59). VFI was not affected by dietary RAC ( $P=0.84$ ) but increased linearly ( $P<0.001$ ) and quadratically ( $P=0.044$ ) with starting weight. Carcass weight increased linearly ( $P=0.006$ ) but not quadratically ( $P=0.37$ ) with dose of dietary RAC. Dressing percentage (data not shown) increased linearly ( $P=0.043$ ) while dietary RAC did not affect carcass P2 ( $P=0.31$ ). These data indicate ADG, FCR and carcass growth increase in a linear manner to dietary RAC up to at least 20 ppm and that the responses are similar regardless of starting weight between 65 and 95 kg.

**Table 1. Effect of starting weight and dietary ractopazine dose on voluntary feed intake (VFI), average daily gain (ADG), feed conversion ratio (FCR) and carcass weight (HSCW) and P2 backfat in finisher gilts**

Weight (W)	Light				Medium				Heavy				Significance <sup>2</sup>		
	Dose (D, ppm)	0	5	10	20	0	5	10	20	0	5	10	20	SED <sup>1</sup>	W
ADG, kg	0.97 <sup>a</sup>	1.09 <sup>b</sup>	1.04 <sup>b</sup>	1.08 <sup>b</sup>	1.10 <sup>a</sup>	1.14 <sup>a</sup>	1.23 <sup>b</sup>	1.19 <sup>b</sup>	1.07 <sup>a</sup>	1.17 <sup>b</sup>	1.13 <sup>ab</sup>	1.24 <sup>b</sup>	0.071	0.002	0.023
VFI, kg/d	2.71	2.86	2.69	2.69	3.18	3.22	3.25	3.20	3.32	3.38	3.45	3.41	0.165	<0.001	0.84
FCR	2.78	2.64	2.59	2.51	2.93	2.83	2.63	2.70	3.11 <sup>a</sup>	2.93 <sup>a</sup>	3.13 <sup>a</sup>	2.77 <sup>b</sup>	0.157	<0.001	0.029
HSCW, kg	70.0	72.7	71.1	74.1	83.5	86.6	87.9	86.5	94.4 <sup>a</sup>	99.5 <sup>b</sup>	96.2 <sup>a</sup>	100.9 <sup>b</sup>	2.39	<0.001	0.010
P2, mm	9.4	9.5	9.9	9.4	10.9	10.1	11.9	11.0	13.1	11.8	12.6	11.6	0.99	<0.001	0.31

<sup>1</sup> Standard error of the difference for Weight x Diet, <sup>2</sup> No significant Weight x Dose interactions. Within a row and starting weight category, means lacking a common superscript letter differ ( $P<0.05$ ).

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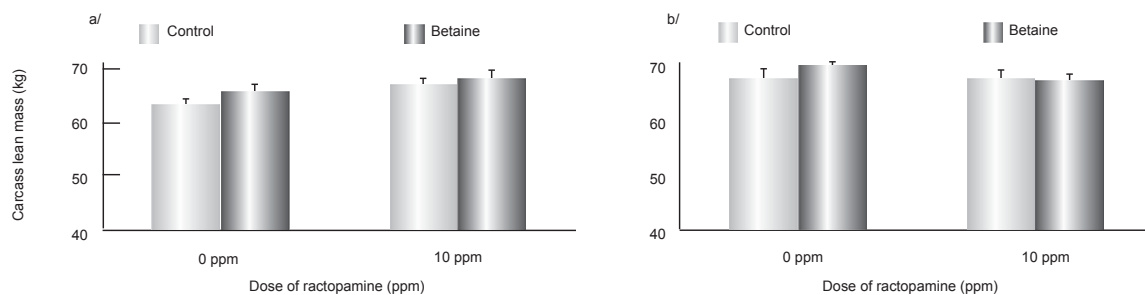
## Dietary betaine and ractopamine have additive effects on lean tissue deposition, particularly in restrictively-fed gilts

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Dietary ractopamine supplementation dramatically increases the rate of carcass lean deposition, with the lean tissue responses most evident at *ad libitum* feed intakes (Dunshea *et al.* 1998). Dietary betaine can improve growth by reducing maintenance requirements, with the greatest responses generally occurring when DE is limiting. Previously we have shown that dietary betaine can stimulate lean tissue deposition in boars consuming 80% of *ad libitum* and that these effects were additive to effects of porcine somatotropin (Suster *et al.* 2004). Therefore, the aim of the present study was to investigate the interactions between dietary ractopamine and betaine on growth performance and carcass characteristics in restrictively-fed finisher pigs.

Forty individually penned pigs (initial weight 58.4 kg) were allocated to a 2 x 2 x 2 factorial experiment with the respective factors being sex (gilt or boar), dietary betaine (0 and 1.25 k/kg Betafin, B) and dietary ractopamine (0 and 10 ppm Paylean, RAC). Pigs were weighed weekly and feed adjusted to ensure that feed intake was restricted to that observed on commercial farms (35 MJ DE /day) (Dunshea, 2005). Diets contained 72% wheat and 16% canola meal and were formulated to 0.56 g available lysine/MJ DE (1.0% total lysine). Body composition was determined by dual energy X-ray absorptiometry at 0 and 35 days.



**Figure 1:** Effect of dietary betaine and ractopamine on lean tissue mass as slaughter in a/ gilts and b/ boars

Over the first 14 days of the study daily gain was increased in pigs fed betaine (+8%,  $P=0.04$ ), was greater in boars (+12%,  $P=0.005$ ) but was not effected by RAC ( $P=0.18$ ). Over the same period, there were similar improvements in feed efficiency. However, growth and efficiency responses were not maintained over the entire study. Lean deposition tended to be greater in pigs fed betaine (+5%,  $P=0.08$ ), was greater in boars (+6%,  $P=0.006$ ) but was not affected by RAC ( $P=0.57$ ). However, there was an interaction ( $P=0.03$ ) between RAC and sex such that RAC increased lean deposition in gilts but not boars. As a result betaine and RAC had additive effects on lean mass in gilts (+5.1 kg) but not boars (Figure 1). Fat deposition was less in pigs fed RAC (-8%,  $P=0.05$ ), was lower in boars (-17%,  $P<0.001$ ) but was not affected by betaine ( $P=0.81$ ). However, there was an interaction ( $P=0.04$ ) between dietary RAC and sex such that RAC decreased fat deposition in gilts (-14%) but not in boars. In conclusion, dietary betaine and RAC can have additive effects on lean deposition and improve body composition, particularly in restrictively-fed gilts.

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## Factors affecting feed intake during lactation for primiparous sows

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It is generally accepted that high feed intake of lactating sows is desirable as it reduces excessive sow body weight loss during lactation and improves piglet weaning weights (Eissen *et al.*, 1999). However, there is relatively little detailed information about factors that affect feed intake during lactation, including whether individual sow appetite is a limiting factor. This paper investigates whether knowledge of traits recorded on gilts at selection or farrowing can be used as predictors of their feed intake during lactation when feed delivery is 'to appetite'.

Lactational feed intake (LFI) records were available for 546 litters from two lines (L1 and L2) of primiparous sows recorded in seven monthly contemporary groups (CGP: levels 7) under two gestational feeding treatments (TMT: levels 2). Traits recorded for selection at 20 weeks of age included average daily gain (ADG) and backfat depths at the P2 and P4 sites (P2 and P4). After selection, gilts were recorded for individual feed intake in group-pens under *ad libitum* feeding (ADI) over a five week period. Pre-mating body weight at 29 weeks (BW29) was recorded along with P2 and P4 (MP2 and MP4). Weight (WT110) and sow body composition (P2100 and P4110) at 110 days of gestation were known. Litter size at birth (TB: total born and NBA: number born alive) and after cross-fostering on day 1 (ND1) were recorded, along with total litter weight at birth (LBW) and after fostering on day 1 (LWD1). The number of stillbirths (SB) was used to categorize probable ease of farrowing into normal (CAT=1:  $\leq 1SB$  for  $TB \leq 14$ ;  $\leq 2SB$  for  $TB > 14$ ) or difficult (CAT=2:  $> 1SB$  for  $TB \leq 14$ ;  $> 2SB$  for  $TB > 14$ ). The significance of all above systematic effects was examined using GLM procedures (SAS Inc v.9.2) using a step-wise approach. Only significant effects were retained in the final model.

Growth rates or weights (ADG, BW29) and fat depth at the P2 site (P2, MP2) were not associated with LFI. Significant ( $P < 0.0001$ ) line differences between gilts for ADI (L1: 2.62 kg/day vs. L2: 2.52 kg/day) were not mirrored by similar or significant differences in LFI. Sow weight and body condition at day 110 of gestation was not significantly associated with LFI, with the exception of P4110. The final model for LFI contained TMT, FCGP, and CAT as class effects, along with LWD1 and P4110 as linear covariates. This multi-variate model explained 12.5% of variation in LFI. Seasonal effects ( $P < 0.01$ ) accounted for ~3% of variation, whereas TMT ( $P = 0.02$ ) and CAT ( $P = 0.02$ ) accounted for ~1% each. Gilts with difficult farrowings ate 275 g/day less than those defined as having normal farrowings. Regression coefficients for P4110 and LWD1 (both  $P < 0.0001$ ) were  $-0.049 \pm 0.01$  kg/mm and  $0.065 \pm 0.02$  kg/kg, respectively. Results indicate that sows adapt lactational feed intakes primarily according to season and their body composition, with respect to both protein and fat accretion. However, additional factors affecting individual sows such as ease of farrowing and post-fostering litter quality (number and weight) were also important.

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## Antimicrobial use in the Australian pig industry – which drugs, how often and why?

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There are widespread concerns that antimicrobial use in food animals selects for resistant bacteria and/or their resistance genes that could then be passed on to humans to cause adverse health outcomes. Collecting information on the use of antimicrobials in food animals is a key component of efforts to reduce the emergence of antimicrobial resistant bacteria. In the pig industry, antimicrobial use is widespread and obtaining information on usage is a priority. We surveyed Australian specialist pig veterinarians on how frequently they treat common bacterial infections in their herds and the type of antimicrobials used. Eight questions related to frequency of treatment of specific bacterial diseases of pigs using antimicrobials while 14 questions asked which antimicrobial drugs had been used in the previous 12 months and their route of administration. The data were captured using an anonymous web-based questionnaire and analyzed in Stata (StataCorp LP v.9) software. Simple descriptive analysis of the reasons for and frequency of antimicrobial use are given in Table 1.

**Table 1. Frequency of antimicrobial use against bacterial diseases (% herds, n=197)**

Disease	Not answered	Not at all	Not much	Somewhat	Very much	A great deal
<i>Mycoplasma</i>	1.0	47.2	9.1	23.4	10.2	9.1
<i>Actinobacillus</i>	1.0	75.1	10.7	6.1	4.0	3.1
Glasser's	1.6	60.9	17.3	12.7	5.6	2.0
<i>Lawsonia</i>	1.0	14.2	2.0	24.9	32.5	25.4
Swine dysentery	1.0	78.7	3.0	6.6	5.1	5.6
<i>Salmonella</i>	1.6	96.0	1.0	1.5	0.0	0.0
<i>E. coli</i>	1.0	29.4	23.9	30.5	12.7	2.5
Other	4.1	64.0	16.8	11.7	3.6	0.0

Antimicrobials used in the 12 months before the survey were: penicillins (95% of herds), tetracyclines (85%), macrolides (75%), sulphonamides (71%), olaquinox (53%), lincomycin/spectinomycin (35%), ceftiofur (26%), dimetridiazole (13%), "other" (11%), tiamulin (11%) and florfenicol (5%).

The majority of antimicrobial use in Australian pig herds is directed against *Lawsonia* (the cause of proliferative enteropathy) and *E. coli* (typically enterotoxigenic strains that cause pre- and post-weaning colibacillosis) - confirming the need for effective control measures against these pathogens. *Salmonella* in pigs is important from an antimicrobial resistance perspective because it can be transferred via food to humans to cause disease. However, these findings suggest that salmonellosis in the pig industry does little to drive consumption of antimicrobials. Respiratory disease caused by *Mycoplasma* is problematic in about half the herds (52.8%) and explains in part why use of macrolide drugs (including tylosin) is widespread. Antimicrobials are being used to treat and/or control swine dysentery in about 20% of herds, and the inevitable withdrawal of dimetridiazole suggests that eradication of this disease should be a priority for the industry. Ceftiofur causes a dilemma because it belongs to a class of drugs that are critically important in humans while in pigs it reduces mortality due to enteritis caused by *E. coli*, which in some herds is showing resistance to other antimicrobial agents including aminoglycosides.

The results of this research will help promote improved use of antimicrobials in the pig industry.

This work is supported by Australian Pork Limited.

## Resistant phenotypes of enteric microbial communities from Australian pig farms

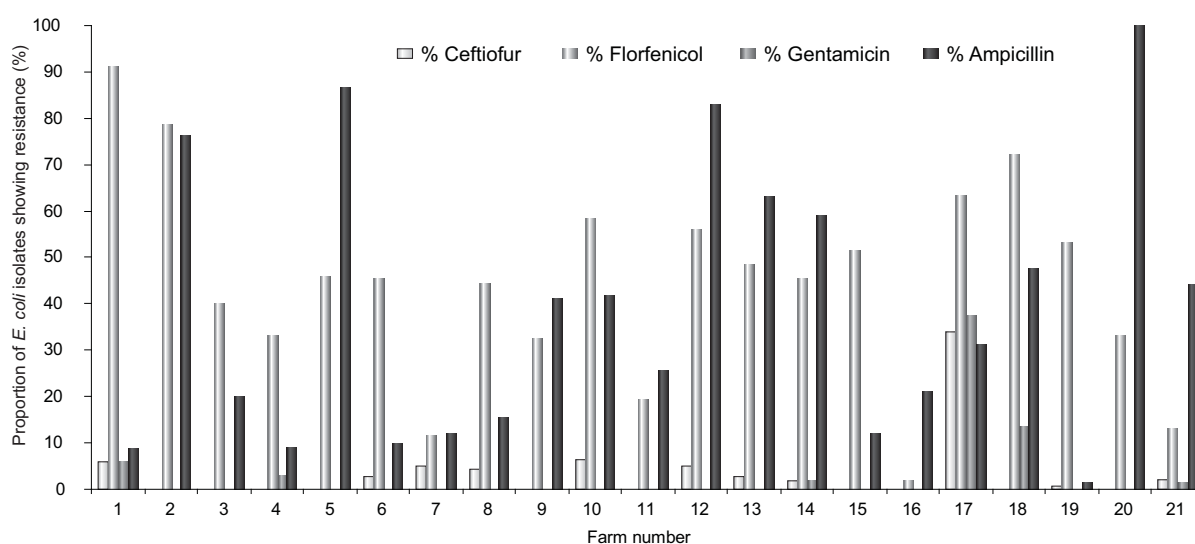
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Enterotoxigenic *E. coli* (ETEC) associated diarrhoea (colibacillosis) is a common disease affecting neonatal and post-weaning pigs that prompts prophylactic and therapeutic use of antimicrobials. Use of such antimicrobials can lead to antimicrobial resistance in gut organisms and potential exposure of humans to these bacteria following consumption of pork products. Judicious use of antibiotics is therefore being encouraged to slow the spread of resistance development, particularly where relevant to human health (Nataro and Kaper, 1998). This study used a mass-screening technique to assess which antimicrobial resistant phenotypes are present in commensal *E. coli* obtained from Australian slaughter pigs. Resistance to four antimicrobials was assessed. Of these, ceftiofur and gentamicin are drugs of high importance in human medicine and it is hypothesized that the prevalence of resistance to ceftiofur and gentamicin will be low in Australian pigs, while resistance to florfenicol and ampicillin will be higher.

Faecal samples (n=30/farm) were sourced from 21 farms with various histories of antimicrobial use. Samples were pooled, serially diluted and plated onto MacConkey agar (MCA). Samples were further diluted to obtain 200-300 representative colonies plated onto a hydrophobic grid membrane filter (HGMF) MCA master plate. This was then replicated onto selective MH plates containing ceftiofur (8 µg/mL), florfenicol (4 µg/mL), gentamicin (4 µg/mL), ampicillin (32 µg/mL) and a non-selective Chromogenic agar plate. Growth of colonies on HGMF grids was assessed by computer assisted image analysis software (HGMF-RES).

Eleven farms recorded some ceftiofur resistance, although only one farm had resistance levels >5% (farm 17: 33.9% resistance) (Figure 1). Resistance to gentamicin was <10%, apart from two farms (17: 37.6% and 20: 13.4%). A high prevalence of resistance was observed to florfenicol, with eight farms having more than 50% of all colonies resistant. Moderate resistance to ampicillin was identified, with six farms demonstrating >50% resistance. Resistance profiles for an additional 60 farms will now be determined.



**Figure 1.** Percentage of *E. coli* isolates within each herd showing resistance

This work was supported by Australian Pork Limited .

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## Easy profiling of *Haemophilus parasuis* serovars present on a farm – is it possible?

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*Haemophilus parasuis* (Hps), the causative agent of Glässer's disease, can be isolated from the nasal cavity, tonsils and trachea of pigs via swabs from Hps-challenged pigs (Kirkwood *et al.*, 2001). As Hps is part of the natural flora of pigs, non-pathogenic strains are also found in the nasal cavity. These non-pathogenic isolates of Hps have a high prevalence in the nasal cavity, while pathogenic isolates have a low prevalence (Oliveira *et al.* 2003). Colonization levels of Hps in a litter increase with increasing time spent with the sow (Kirkwood *et al.*, 2001) and litters from older sows have a higher colonization level than younger sows (Cegielski *et al.*, 1999). Therefore, nasal swabbing piglets from old sows at weaning should be the optimum time to recover Hps from the nasal cavity. In this work, the potential of nasal swabbing as a tool for serovar profiling was evaluated.

Eleven Australian pig farms were sampled over a year. Nasal swabs were either taken from pigs of old sows at weaning time or from sick pigs displaying symptoms of Glässer's disease (coughing, anorexia). Swabbing was done with Amies transport swabs, kept on ice until inoculated on to BA/SN agar (Turni and Blackall, 2007) and blood agar, the latter being cross-streaked with a nurse colony of *Staphylococcus hyicus*. All suspect colonies of Hps were selected and subcultured for DNA processing, storage and serotyping. Isolates were confirmed as Hps by Polymerase Chain Reaction (PCR) (Oliveira *et al.*, 2001). Enterobacterial Repetitive Intergenic Consensus (ERIC) PCR as described by Oliveira *et al.* (2003) was used to group the isolates according to genotype profile. A representative isolate of each genotype was then serotyped (Turni and Blackall, 2005).

Key findings of the work were: 1) Hps was found on farms without clinical signs of Glässer's disease, which is not surprising as Hps is part of the natural flora. 2) On a farm where serovar-4 had in the past caused an outbreak of Glässer's disease the disease causing organism was not recovered during sampling of weaned pigs. However, it was recovered in a sick pig. Sampling pigs that were negative at weaning over time revealed that a nontypeable (NT) serovar on this farm spread very rapidly. Serovar-4 was recovered at 72 days of age when up to six colonies of Hps were isolated for each pig and a total of 103 samples compared using ERIC PCR. While all 23 pigs yielded NT isolates, only six pigs yielded serovar-4. This could indicate that multiple serovars are not easily detected if one serovar is more prevalent than the other. 3) Sampling healthy weaned pigs does not necessarily reveal all pathogenic strains, as samples from necropsied pigs yielded additional pathogenic strains. 4) Sampling sick pigs alone might not reveal known non-pathogenic strains (serovars 3, 6, 7, 9, 11). 5) Combined sampling of weaned and sick pigs seems to enhance the chances of finding pathogenic strains.

Overall, sampling appropriate pigs and using a combined genotyping/serotyping approach offers a practical tool for the profiling of Hps serovars present on a farm.

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## Reducing survival and transmission of *Lawsonia intracellularis* in conventional piggeries

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Australian and overseas attempts to eradicate proliferative enteropathy (PE) have focused on depopulation and repopulation of herds and high levels of medication for all new and existing pigs. Almost all of these eradications have failed within one year, with PE reoccurring in the herd (Johansen *et al.*, 2002; Collins *et al.*, 2006). Failure to eradicate PE from the herd may be due to unsuccessful elimination of the causative agent from the pig or from the environment. *Lawsonia intracellularis* can survive in porcine faeces in the environment for at least two weeks and infect naïve pigs (Collins *et al.*, 2000). We hypothesised that the survival and transmission of *L. intracellularis* could be significantly reduced with cleaning and disinfection of pens.

Twelve Large White x Landrace finisher pigs shedding *L. intracellularis* in their faeces were used to contaminate conventional pens over a three week period. The pigs were removed and pens were left vacant for two weeks. During this period, half of the contaminated pens were power hosed and disinfected with Virkon S<sup>®</sup>, and left to dry for three days. The other pens were left empty and uncleaned. Two groups of naïve weaner pigs were introduced into these pens: one group (n=10) to the uncleaned environment and the other (n=12) to the cleaned environment. Faeces and blood were collected weekly to detect individual faecal shedding of *L. intracellularis* by polymerase chain reaction (PCR) and serum IgG antibodies to *L. intracellularis*. Pigs were necropsied 56 days after introduction, and ileum and colon tissue was collected for polymerase chain reaction (PCR) detection of *L. intracellularis*. Minimum and maximum temperatures, recorded daily throughout this period, ranged from an average overnight minimum of 9°C to an average daily maximum of 18°C.

Transmission of *L. intracellularis* infection was demonstrated in naïve pigs introduced into the uncleaned pens, with faecal shedding of *L. intracellularis* detected in two of 10 pigs 21-35 days post exposure. Antibodies to *L. intracellularis* were detected in eight of 10 of the same pigs 28-42 days post exposure. *L. intracellularis* DNA was not detected in the ileum or colon of these pigs necropsied 56 days after entering the uncleaned pens. No evidence of *L. intracellularis* transmission was demonstrated by PCR or serology in the 12 pigs introduced into the cleaned environment for up to 56 days after exposure to the cleaned environment.

*L. intracellularis* is able to survive in faeces in uncleaned conventional pens for at least two weeks at temperatures ranging between 9°C and 18°C and subsequently colonize naïve pigs. Power hosing and disinfection of conventional pens contaminated with faeces containing *L. intracellularis* was able to significantly reduce the survival and transmission of *L. intracellularis* to naïve pigs introduced into that environment (P<0.05).

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## Effect of pneumonia on body temperature in grower gilts

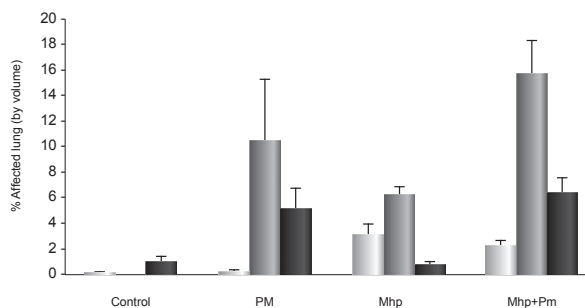
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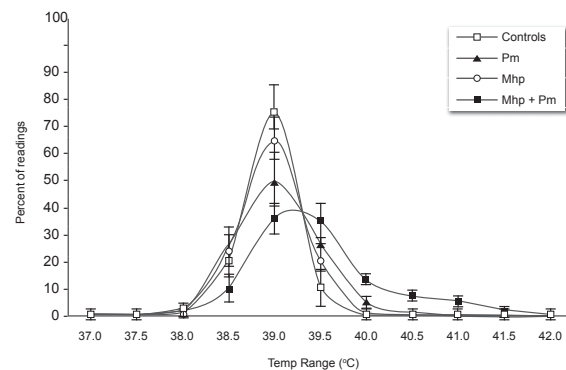
A non-invasive procedure to monitor body temperature (BT) responses associated with pneumonia continuously may provide useful data for the AUSPIG model. Two important respiratory disease agents that occur widely in the Australian pig industry, *Mycoplasma hyopneumoniae* (*Mhp*) and *Pasteurella multocida* (*Pm*), were used to infect grower gilts experimentally derived from a PIC genotype high health-status herd and the effects of single and combined sequential challenge were evaluated. Four groups of 16-17 gilts were housed in individual pens within separate rooms of a controlled environment facility. Two groups were challenged intratracheally at 11 weeks of age with a NSW field strain of *Mhp*. Three weeks later, a third group and one of the two *Mhp* challenge groups were challenged intratracheally with *Pm* strain PM508. The fourth group remained as unchallenged controls. Pneumonia severity was assessed by computer tomography (CT) scanning on three occasions: before *Pm* challenge; at 2.5-3 weeks and at 5-6 weeks after *Pm* challenge.

To avoid the problem of elevated BT readings caused by continuous monitoring of BT using restraint, we used a novel approach of intravaginal data loggers on silicone inserts. By staggering applications among groups, data capture over a 51 day period was readily achieved on two separate, two-week periods for each gilt, commencing six days before *Pm* challenge.

Among the three CT scan times (Figure 1), pigs had more severe pneumonia 2.5 to three weeks after *Pm* infection (sub-acute phase) and gilts with combined *Mhp* and *Pm* infection were most severely affected in both the sub-acute and chronic (5-6 weeks post challenge) phases. One to six weeks after *Pm* challenge, there was considerable divergence between the two groups challenged with *Pm* and either control and *Mhp*-only pigs in terms of the proportion of temperature readings across the range 37-42°C. Pigs infected with *Pm* had a marked shift in mean temperature and a marked spread of temperature responses (Figure 2) particularly in weeks one and two, and combined infection had a greater and more sustained effect up to week five. Strategic application of data loggers on silicone inserts provided a practical method to collect continuous BT data in populations of females over 55 kg.



**Figure 1.** Mean pneumonia severity (as % affected lung + SE) among treatment groups at three sequential CT scan times



**Figure 2.** Proportion of body temperature readings ( $\pm$ SE) ranges during the 2nd week after *P. multocida* challenge

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# 6

## Sow reproduction

# The role of the conceptus in determining placental efficiency and the capacity of the uterus

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## Abstract

Uterine capacity has been used to describe simply the number of conceptuses that can be gestated until the term of gestation. Furthermore, there has been widespread acceptance of the concept that ovulation rate and uterine capacity are the two major determinants of litter size. Placental efficiency has emerged in the last decade as a novel component of litter size and may impact both postnatal survival and growth. Grossly, placental efficiency has been described as the weight of the neonate (or foetus) divided by the weight of the placenta. Although numerous reports have indicated the strong positive relationship between foetal weight and placental weight, variation in placental weight does not completely describe foetal weight. It has been proposed that the residuals from the line describing the relationship between foetal weight and placental weight reflect variation in placental efficiency. It has been argued that this measure provides some indication of the capacity of the placenta to maintain growth of the foetus throughout gestation. It also predominantly reflects variation in placental weight and not variation in foetal weight. In numerous reports the negative correlation between placental weight and placental efficiency has been described including as early as day 25 of gestation. The Meishan breed exhibits a very high placental efficiency (nearly three to four times that of controls) and this appears to play a role in this breed's prolificacy. Tremendous variation in placental efficiency exists both within and among litters of commercially relevant breeds and lines. A significant amount of the variation in placental efficiency can be described by the density or number of blood vessels in the placenta and to a lesser extent the adjacent endometrium. Little to nothing is known about how specific transport mechanisms may influence placental efficiency and this is an area of research ripe for investigation.

## Introduction

Placental efficiency is a recently recognized, difficult to evaluate, but likely a critical component of the reproductive efficiency of the female and may impact the productive efficiency of her offspring (Ford, 1997; Wilson *et al.*, 1999). Placental efficiency has been grossly defined as the weight of the foetus or neonate divided by the weight of the placenta. In reality, the efficiency of the placenta is a combination of placental blood flow and vascular density, the density and activity of, sometimes complex, placental transport mechanisms and the degree of three dimensional complexity of the interface for nutrient exchange, including the primary and secondary folds of the placental membrane as well as the complexity of the microvillar surface. An often underappreciated component of placental efficiency is the rate of nutrient delivery which itself is a combination of uterine blood flow, the density and activity of endometrial nutrient transporters and the matched three dimensional complexity of the endometrial surface.

In a pig production enterprise, the reproductive efficiency of the sow is critical to the overall performance of the operation (Foxcroft *et al.*, 2006). The capacity of the uterus determines the upper limit of litter size in the sow (Fenton *et al.*, 1972). This capacity has several contributing components: on the maternal side, the size and biochemical capacity for nutrient exchange, including blood flow to the gravid uterus, and on the foetal side, the size and efficiency of nutrient absorption by the placenta. In addition to the impact of placental efficiency and/or uterine capacity on determining litter size, recent evidence has accumulated leading to the suggestion that the efficiency of nutrient delivery to individual conceptuses will influence not only their birth weight, but can have long term impacts on the efficiency and rate of growth postnatally (Foxcroft *et al.*, 2006; Wu *et al.*, 2006).

## Defining uterine capacity

Recent reviews of prenatal conceptus growth and development (Foxcroft *et al.*, 2006; Wu *et al.*, 2006) recount the early demonstrations by Dziuk (1968) and Bazer *et al.* (1969a,b) that in cases in which the number of embryos present was well in excess, the limitations of the uterus would lead to considerable conceptus loss. In fact Dziuk (1968) used several different methods to insure that the number of embryos was in excess (i.e. strategic ligation of the uterus to create a roomy and crowded compartment within each gilt, unilateral hysterectomy and ovariectomy, superovulation and superinduction) and in all cases in which the conceptuses were actually crowded in excess of seven per horn or 14 total there was a dramatic reduction in survival. Fenton *et al.* (1972) described uterine capacity as 'the ability of the



uterus to support only a limited number of embryos'. Utilizing superinduction, they found that in contrast to previous reports (Bazer 1969a), increasing the number of embryos increased the number of conceptuses present. However, when they increased the number of embryos further by transferring embryos from two donors into already pregnant females, there was a limitation on litter size imposed by the uterus (Fenton *et al.*, 1972). There are two important concepts relating to these studies that are worth noting. First, these authors all assumed that all conceptuses were equal, which is not true and the variation that exists at least contributes to uterine capacity. Second, these authors were all utilizing genetics that had ovulation rates of 10.0-13.5, while a number of recent reports have indicated that ovulation rates in modern commercial sow lines average greater than 22.1 (Vonnahme *et al.*, 2002; Town *et al.*, 2004; Foxcroft *et al.*, 2006).

This limitation of the uterus appears to set a ceiling on the number of conceptuses that can survive to term. This concept formed the basis for an experimental approach to increase litter size by selecting for uterine capacity (Christenson *et al.*, 1987). These authors employed unilateral hysterectomy in conjunction with ovariectomy to challenge uterine capacity. The removal of one ovary results in a near doubling of ovulation rate on the remaining ovary, referred to as ovarian compensatory hypertrophy. By also removing the ipsilateral uterine horn, the number of embryos per uterine horn is dramatically increased so that litter size is determined by uterine capacity. Following 11 generations of selection, these authors evaluated uterine capacity (utilizing unilateral hysterectomy and ovariectomy) and litter size in control and selected individuals. Selection for uterine capacity resulted in an increase in uterine capacity of ~one pig per uterine horn; however, when females from the selected line were left intact, there was no correlated increase in litter size (Leymaster and Christenson, 2000). It appears that the observed response to selection of an increase in uterine capacity was only realized in situations in which the number of conceptuses was nearly twice what was observed in control animals (i.e. the increase in uterine capacity was only observed if the animals were unilaterally hysterectomized and ovariectomized, but not if they were left intact).

### Defining placental efficiency

Although the term 'placental efficiency' has been used to better describe the components of uterine capacity in the pig recently (Ford, 1997; Biensen *et al.*, 1998), the term has been utilized to describe the same physiological phenomenon for some time. Thirty years ago Battaglia's group described the relationship of human foetal and placental weight and described the foetal:placental weight ratio as placental efficiency (Molteni *et al.*, 1978). The introduction of the term to describe variation in placental efficiency amongst litter mates in the pig grew out of a series of studies in the 1990s designed to determine the mechanism(s) by which the prolific Meishan pig was able to give birth to 3-5 more pigs per litter than the commercial, Western breeds to which it was compared. Previously it had been demonstrated that the ovulation rate in Meishan and control gilts of similar physiologic age (i.e. the same number of post-pubertal oestrus cycles) were the same (Bazer *et al.*, 1988). In addition, the gross size of the uterus was similar between Meishan and control breed females (Bazer *et al.*, 1988). Therefore, it was reasoned that the difference in litter size had to be controlled by the conceptus. Although Meishan neonates were smaller than controls at birth, in a clever experimental design, Meishan and control embryos were co-transferred to control recipient females. Surprisingly, the litter mate Meishan and control neonates were born at a similar body weight; however, the control placentae were nearly 50% heavier than their Meishan litter mates. In reporting these data, the authors first utilized the term placental efficiency to describe this notion that individuals from a litter can vary dramatically in the relationship between foetal weight and placental weight and they suggested that the variation in this relationship might be a result of variation in the density of blood vessels in the placental membranes (Ford, 1997). The measured placental efficiency of Meishan conceptuses was 8.7 whereas the controls had an average placental efficiency of 3.4. These authors speculated that this marked difference in placental efficiency, which could be attributed almost entirely to differences in placental weight, likely contributed significantly to the ability of the Meishan to gestate more foetuses to term of gestation than controls, despite the similarity in uterine size and ovulation rate (Ford, 1997).

Numerous reports have demonstrated a positive correlation between foetal weight or birth weight and the weight of the placenta (Knight *et al.* 1977, Wootton *et al.*, 1977; Mesa *et al.*, 2003; Town *et al.*, 2004; Vianna *et al.*, 2004). However, regardless of the stage of gestation in which the relationship is measured, the weight of the placenta only describes at best 50% of the variation in foetal or neonatal weight. Therefore, variation in the efficiency of placental function clearly exists. In effect, placental efficiency describes the residuals of the regression of foetal or neonatal weight on placental weight. Those above the line having higher than average placental efficiency and those below the line having lower than average placental efficiency. This is true regardless of the shape of the line. Mesa *et al.* (2003) indicate that the relationship is quadratic, in that those conceptuses with the heaviest placentae did not exhibit a linear increase in birth weight, but the quadratic component of the line describing their data set is three orders of magnitude less than linear effect of placental weight on foetal weight (birth weight=341+(6 x placental weight)+(0.006 x placental weight<sup>2</sup>)). Furthermore, in both their control and selected lines, the variation in placental weight did not

describe more than 52% of the variation in birth weight. Similarly, Vallet *et al.* (2002) described the relationship as quadratic, but they also reported that the quadratic component is three orders of magnitude less than the linear effect (foetal weight =  $83.6 + (5.6 \times \text{placental weight}) + (-0.008 \times \text{placental weight}^2)$ ) and the quadratic component appears from their figure to simply describe those individuals in which the placenta is very large (likely due to a very roomy uterine environment) and the foetus is growing at or near its potential.

### Evidence for variation in placental size and function

Following the dramatic demonstration that the Meishan placental efficiency was twice that of controls, Wilson *et al.* (1999) reported that within litters of purebred Yorkshire females there was a significant variation in placental efficiency within litters. Across eight litters, in which they had tagged individual umbilical cords at farrowing so that they could determine the efficiency of each conceptus, the average placental efficiency was  $4.2 \pm 0.2$ . However, the range of individual placental efficiencies varied from 2.7-7.4. The authors emphasized that placental efficiency observed within a single litter ranged from 3.8-7.4, indicating that there is tremendous variation within a single litter in which the individual conceptuses should be exposed to a relatively uniform environment. Additionally, in selecting offspring in a situation in which uterine capacity has been challenged based simply on the number of conceptuses present could conceivably result in the selection of individuals with a low placental efficiency in a litter that is large as a result of the littermates having very efficient placentae.

The variation in the efficiency of the placenta did not appear to be related to the variation in foetal weight (Wilson *et al.*, 2001). However, there was a pronounced negative correlation between placenta efficiency and placental weight ( $r = -0.72$ ,  $P < 0.0001$ ). Others have reported similar negative correlations between placental efficiency and placental weight ( $r = -0.53$  to  $-0.75$ ) with little to no association between placental efficiency and foetal weight at or near term of gestation (Vallet *et al.*, 2002, Mesa *et al.*, 2003; Town *et al.*, 2004, 2005). This has been interpreted as indicating that both small and large foetuses can grow on relatively efficient and relatively inefficient placentae, but that small placentae tend to be relatively efficient and large placentae tend to be relatively inefficient. Therefore, placental weight can serve as a proxy for placental efficiency, as long as birth weight is acceptable. Unfortunately, in order to know which placenta goes with which foetus both measures must be taken and therefore placental efficiency can be calculated.

In a data set collected from a herd depopulation that included 244 sows from parity 1-14, placental efficiency varied from 0.2-0.9 on day 25 of gestation, from 0.06-0.25 on day 36 of gestation and from 0.10-0.30 on day 44 of gestation (Vonnahme *et al.*, 2002). Somewhat surprisingly, the placental efficiencies in early gestation (days 25, 36 and 44) were also negatively associated with placental weight in a cubic manner, with variation in placental weight accounting for 89-96% of the variation in placental efficiency (Vonnahme *et al.*, 2002). As with the late gestational measures of placental efficiency, there was no relationship between placental efficiency and foetal weight. Others have described a similar negative correlation between placental efficiency and placental weight early in gestation (i.e. day 20-30) in which approximately 40% of the variation in placental efficiency was described by placental weight (Town *et al.*, 2004, 2005). Interestingly, utilizing an experimental paradigm of unilateral oviduct ligation (which should increase the amount of uterine space for embryos derived from ovulations from the contralateral ovary), there was no difference in the average placental efficiencies for conceptuses gestated in a relatively crowded versus relatively roomy uterine environment (Town *et al.*, 2004). The degree of variation observed within a litter of full siblings assumedly exposed to the same environment and the apparent lack of plasticity in this trait would support the suggestion that placental efficiency is controlled by the conceptus.

### Determinants of placental function

The initial descriptions of placental efficiency in the pig suggested that variation in efficiency reflected variation in the density of blood vessels in the placenta (Ford, 1997; Biensen *et al.*, 1998). This early speculation was in large part a result of the observation of two very distinct types of placentae observed at farrowing in the litters of control gilts that had gestated both Meishan and control conceptuses described above. In addition to the observation that the control placentae were 50% heavier than the litter mate Meishan, there was a dramatic difference in the apparent vascularity between these placental types that was evident even in the grayscale photograph of representatives of these two distinct placental types (Ford, 1997). The density of blood vessels in the placenta of purebred Yorkshire conceptuses appeared to be relatively constant throughout the last 40 days of gestation (Biensen *et al.*, 1998). Conversely, the vascular density of the Meishan placenta nearly doubled during the last 40 days of gestation (Biensen *et al.*, 1998). Furthermore, across litters of Yorkshire conceptuses the measured placental efficiency was positively correlated to vascular density of both the placenta ( $r = 0.70$ ,  $P < 0.02$ ) and to a lesser extent the endometrium ( $r = 0.26$ ,  $P = 0.07$ ) (Wilson *et al.*, 2001). This relationship is also observed in cases where the number of placental vessels are counted instead of the percentage of the tissue occupied by blood vessels ( $r = 0.48$ ,  $P < 0.002$  between number of placental

vessels and placental efficiency and  $r=0.40$ ,  $P<0.05$  between number of endometrial vessels and placental efficiency) (Vonnahme *et al.*, 2001). Conversely, Mesa *et al.* (2003) found no association between placental vascularity and placental efficiency. This may have been because of their estimation of vascularity was a subjective colour scoring on a scale of 1 to 5 instead of a quantitative histological approach.

The arrangement of the endometrial and placental vasculature has been described as cross-countercurrent and the absorption of nutrients by the placenta is in large part determined by the rates of maternal and foetal blood flow (Leiser and Dantzer, 1988). From day 44 until 112 days of gestation, there was a dramatic linear increase in the proportion of placental tissue occupied by blood vessels and this was matched to a linear increase in the potent angiogenic factor vascular endothelial growth factor (Vonnahme *et al.*, 2001). These authors reported a strong positive correlation between the placental expression of vascular endothelial growth factor and the efficiency of the placenta throughout gestation ( $r=0.66$ ,  $P<0.0001$ ). In addition these authors report an increase in vascular endothelial growth factor in foetal blood during the latter portion of gestation and indicated that the foetal plasma concentration of vascular endothelial growth factor is positively correlated with both foetal weight ( $r=0.45$ ,  $P<0.01$ ) and placental efficiency ( $r=0.40$ ,  $P<0.01$ ).

In addition to the relative vascularity of the placenta, the degree of three-dimensional interdigitation between the chorioallantoic membrane of the placenta and the folds in the endometrium could contribute to the relative efficiency of the placenta. The degree of folding in primary and secondary rugae increased from day 70-110 of gestation, but did not differ between control and Meishan placentae despite the dramatic difference in placental efficiency between these breeds (Biensen *et al.*, 1998). Recently, Vallet and co-workers (unpublished) have also investigated the degree of folding at the maternal foetal interface. They also found that the degree of interdigitation increased as gestation progressed from day 45-105, but was not different amongst lines selected for ovulation rate or uterine capacity and was only weakly ( $r=0.19$ ,  $P<0.05$ ) related to placental weight across all days. It is possible that the degree of interdigitation is limited by the inherent folding of the endometrium, and as individuals within a litter vary widely in their placental efficiency the degree of folding may not contribute significantly to the variation in placental efficiency.

A number of transport mechanisms exist in the tissues of both the endometrium and the placenta that are responsible for the regulated transfer of nutrients and wastes between the maternal and foetal blood (Sibley *et al.*, 1997). Transported nutrients would include glucose, amino acids, ions (e.g. sodium, potassium, calcium and protons) (Wilson and Ford, 2001). In addition to these nutrient building blocks there are some large macromolecules that appear to be taken up by placental areolae (specialized structures on the surface of the placenta that interact with the very active uterine glands). Unfortunately, there is a dearth of information on the relative activities of these transporters in either the endometrium or the placenta. Self *et al.* (2004) and Wu *et al.* (2005) have described concentrations of amino acids and a number of amino acid catabolizing enzymes in placental tissues throughout gestation, but as yet have not described variation among placentae of different relative efficiencies within a given day of gestation. Furthermore, a comparison of the activity of sodium dependent- and independent-leucine transport between the smallest and an average foetus in litters at 45, 65 and 100 days of gestation only indicated that the placentae from normal sized foetuses switched from sodium-independent to a mix of sodium-independent and sodium dependent transport mechanisms at day 100 of gestation (Finch *et al.*, 2004). This same transition was not observed in placentae from the smallest foetus in the litter which maintained just sodium-independent transport. Clearly, there is a tremendous need for additional information about the types of transporters present in both the endometrium and the placenta and whether they vary with placental efficiency.

### Determinants of uterine function

Probably the most significant determinant of uterine function during gestation is blood flow to the gravid uterus (Ford, 1995). Unfortunately, the measure of blood flow is to the entire gravid uterus and can not be attributed to individual conceptuses. The rate of uterine blood flow during the last three weeks of gestation remained relatively constant until approximately one hour after parturition, at which time it precipitously dropped (Ford *et al.*, 1984). When expressed on a per foetus basis, uterine blood flow was approximately 250 ml per minute per foetus, but the authors were not actually able to determine what percentage of total flow (1.5 L per minute per uterine horn) actually went to which foetus (Ford *et al.*, 1984).

As indicated above, the density or number of microscopic blood vessels in the endometrium at the maternal-foetal interface was at least weakly associated with placental efficiency (Vonnahme *et al.*, 2001; Wilson *et al.*, 2001). There was also an observed positive association between the density or number of microscopic blood vessels in the endometrium ( $r=0.68$ ,  $P<0.05$ ) and placenta ( $r=0.64$ ,  $P<0.0001$ ) (Vonnahme *et al.*, 2001; Wilson *et al.*, 2001). The coordinated development of both the placental and adjacent endometrial microvasculature probably reflects the local angiogenic stimulus produced at least in part by the placental expression of vascular endothelial growth factor noted above.

### Heritability of placental size and function

In a paper that really put the spotlight on placental efficiency and its potential contribution to uterine capacity, Wilson *et al.* (1999) selected animals for high and low placental efficiency out of the litters described above. In these litters, there was almost 300% variation in placental efficiency, including a range from 3.8 to 7.4 in a single litter. Care was taken to select litter mates for each group that were born at a similar and acceptable birth weight (i.e. >1.2 kg). Therefore, the selection of animals to form each group really reflected high and low placental weight. Gilts were then bred to boars within each group and gilts produced both a gilt and parity one litter. At each farrowing individual placentae were tagged and those individuals that had been selected for a high placental efficiency gave birth to offspring that had an elevated placental efficiency and those that were selected for a low placental efficiency gave birth to offspring with a low placental efficiency. Notably, although there was a slight reduction in birth weight in the offspring of the high placental efficiency group, they were born in litters that averaged 12.5 pigs per litter as compared to the 9.6 pigs per litter born to the low placental efficiency group. The offspring from females selected for a relatively high placental efficiency had placentae that were nearly 40% lighter than offspring born to litter mate females that had been selected for relatively low placental efficiency. Although this was a very small experiment, the realized heritability of placental efficiency was 0.37. It is likely worthwhile noting that the average placental efficiency in those selected for relatively high placental efficiency was approximately 5.5, still well below the observed placental efficiency of average Meishan conceptuses (8.7) (Ford, 1997). Upon further investigation, it was demonstrated that at day 90 of gestation individuals from the relatively high placental efficiency group exhibited approximately 30% greater placental expression of vascular endothelial growth factor than the individuals from the relatively low placental efficiency group (Vonnahme and Ford, 2003).

Mesa *et al.* (2005) reported the results of a similar selection experiment, with the exception that they developed an index for selection that included total born, birth weight and placental weight. Following four generations of selection they reported a decrease in placental weight with a concomitant increase in placental efficiency. They estimated the additive heritability for these two traits to be 0.25 and 0.18, respectively. They report that the improvement in placental efficiency did not result in an increase in the total born or the number born alive, although they only report the overall mean for these which were both better than average (11.0 and 10.6, respectively). Moreover, as there is no information about these animals, or ones with a similar genetic base, on their ovulation rate one can not discern whether uterine capacity was challenged enough to observe an impact of improved placental efficiency. However, in commercial dam lines in which indirect selection for ovulation rate has dramatically increased the potential number of embryos to the point that nearly 50 % of the potential conceptuses are lost (reviewed in Foxcroft, 2006), there is still potential for a benefit from selection for placental efficiency.

### Environmental influence on placental growth

The best evidence for factors that appear to influence placental efficiency comes from the comparisons between the Meishan and control breeds (Ford, 1997, Wilson and Ford, 2001). In addition to having smaller, more vascular and more efficient placentae than control breeds late in gestation, Meishan embryos grow more slowly from the early blastocyst stage through until immediately before elongation (Rivera *et al.*, 1996; Wilson and Ford, 1997). They also produce significantly less oestrogen, which appears to elicit a reduced secretory response from the endometrium (Anderson *et al.*, 1993; Youngs *et al.*, 1994). This may lead to the reduced number of cells present as the embryo initiates elongation and/or completes the elongation process (Youngs *et al.*, 1994; Ford, 1997). The argument was put forward that in order to test the hypothesis that oestrogen production by the blastocyst influenced uterine secretion, which in turn influenced placental size and efficiency one would either need to decrease oestrogen exposure in a control animal or provide exogenous oestrogen to a Meishan. As the latter was more technically feasible, Wilson and Ford (2000) gave exogenous oestrogen to pregnant Meishan gilts only at the time of embryo elongation and then observed placental size at day 112 of gestation. Treatment with exogenous oestrogen only at the time of elongation resulted in a 40% increase in placental size by day 112 of gestation. This marked increase in placental size did not increase foetal weight and so the treatment also reduced placental efficiency. Recently, Wilmoth *et al.* (2006) have reported that a similar oestrogen treatment in control animals resulted in a 50% increase in the rate of proliferation of the trophoblast of embryos collected following the oestrogen administration. The apparent lack of plasticity in placental efficiency alluded to above may in fact not represent a programmed potential for placental development, but may instead reflect the result of the development of the embryo during the first two weeks of gestation.

### Potential for placental efficiency to impact postnatal survival and growth

In the last several years a tremendous amount of research in prenatal growth and development has been focused on 'developmental programming'. Although the first data were collected more than 60 years ago when Hans Selye reported long term behavioral alterations in offspring following application of a stressor to the pregnant dam,

the intensity of research rapidly increased following several reports by David Barker which outlined what has become known as the Barker Hypothesis. Following some refinement, the hypothesis basically postulates that during the development of organ systems in utero, particularly those that often go underdeveloped to ensure appropriate brain and cardiovascular development, an insult can have long term deleterious consequences on the functioning of that organ system, ultimately leading to an increased risk of adult onset disease.

Placental efficiency (i.e. the relative ability of a gram of placenta to provide support to a number of grams of foetus) would likely modulate the ability of an individual foetus to assimilate nutrients. It is not unreasonable then, that placental efficiency may have consequences well beyond serving as a component of litter size. In fact, in a large population with different genetic merit for postnatal survival, in which the difference in estimated breeding values for postnatal survival varied by >16%, an investigation of potential prenatal indicators of postnatal survival was undertaken (Leenhouwers *et al.*, 2002). They observed that those litters with the highest estimated breeding values for postnatal survival also had the smallest and most efficient placentae. Furthermore, although foetal length was reduced in these litters, the weight of the liver, adrenal and small intestine were increased. They also found that liver and muscle glycogen content were increased with increasing genetic merit for postnatal survival.

In the experimental paradigm described above in which one oviduct is ligated to reduce the number of conceptuses and generate a 'roomy' uterine environment, Town *et al.* (2004) collected semitendinosus muscles to compare the effect of the availability of uterine space on muscle growth and development. The underlying hypothesis was that the numbers of conceptuses surviving during early gestation exceed uterine capacity for normal development, especially in commercial dam lines in which ovulation rate is very high. Subsequently, conceptuses that do survive to term have had to compete for space and nutrients during early stages of muscle development. They found that at day 90 of gestation, the weight and the cross sectional area of the semitendinosus muscle were both reduced by nearly 20% in those foetuses in the relatively crowded uterine environment compared to those in the roomy environment (Town *et al.*, 2004). They pointed out that the ovulation rate in the intact animals was only 19 which is only about two-thirds of the ovulation rates that have been reported for some dam lines. The implication is that the higher ovulation rate animals there may be even more detrimental impact on muscle development. This potential reduction in muscle protein accretion during development in utero may dramatically hamper later lean tissue growth and development, slowing growth and limiting muscle protein accretion in the production system.

## Conclusions

Overall, placental efficiency is an important trait. Ultimately, placental efficiency is a component of uterine capacity and it is important to realize that individuals within a litter may all respond differently to what should otherwise be a fairly similar uterine environment. It exhibits wide variation amongst individuals, but appears to exhibit a relatively high heritability, especially for a reproductive trait. It is somewhat technically challenging to measure, particularly in a commercial production system. However, if we can uncover the mechanistic regulation of the observed variation in placental vascularity, as well as begin to establish other biochemical mechanisms that contribute to variation in placental efficiency, perhaps we can define markers of placental efficiency that may be more practical selection tools. This may be especially important as we learn more about how the relative development of the foetus during gestation and its ability to compete for nutrients may have dramatic impacts on postnatal survival and growth potential.

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## Effects of maternal somatotropin injections or feeding ractopamine in early-mid pregnancy in gilts and sows

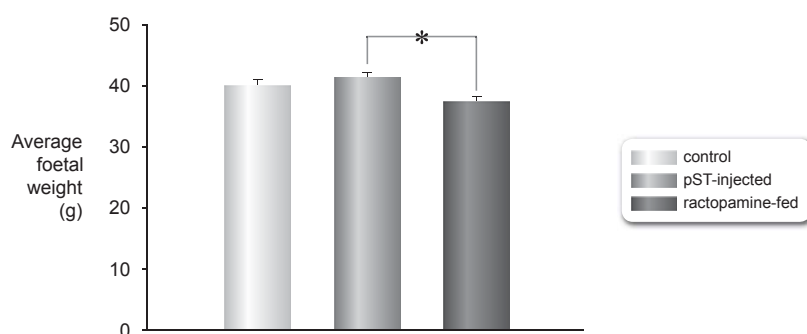
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Foetal size and birth weight positively predict neonatal survival and postnatal growth rates and efficiency. Daily porcine somatotropin (pST) injection of gilts in early-mid pregnancy increases fetal growth and may improve progeny growth performance and muscularity (Rehfeldt *et al.*, 2004). Limited data suggest that feeding sows with  $\alpha$ -adrenergic agonists (e.g.  $\alpha$ -ractopamine) may have similar effects (Hoshi *et al.*, 2005). We hypothesized that pST or  $\alpha$ -ractopamine treatment from day 25-50 of pregnancy would increase foetal growth in gilts and sows.

Large White x Landrace F1 gilts (day-25 weight: 136 $\pm$ 2 kg, mean $\pm$ SEM) and sows (parities 3-6, day-25 weight: 221 $\pm$ 6 kg) were injected daily with water (control) or with recombinant pST as Reporcin<sup>®</sup> (pST-injected, gilts: 2 mg/day, sows: 3.5 mg/day,  $\sim$ 15 $\mu$ g/kg/day), or fed  $\alpha$ -ractopamine as Paylean<sup>®</sup> (ractopamine-fed, 20 ppm), from day 25-50 of pregnancy. Gilts were fed 2.2 kg/day and sows were fed 2.5 kg/day of a dry sow ration (13.0 MJ DE/kg, 15.2% protein). Maternal weight and P2 backfat depth were measured on days 25 and 50. Dams were humanely killed at day 50 and foetuses and placentae dissected, weighed and measured. Data from dams with a litter size <9 were excluded (remaining n=3-6 dams/group). Effects of treatment and parity were assessed using two-way analysis of variance with litter size as a covariate.

Treatment affected maternal weight gain in sows (P=0.009) but not in gilts (P=0.6). Sows treated with pST gained more weight (21.4 $\pm$ 1.4 kg) than control (14.0 $\pm$ 0.7 kg) or ractopamine-fed sows (10.8 $\pm$ 2.5 kg). Treatment did not alter P2 backfat depth (1.1 $\pm$ 0.8 mm). Average fetal weight correlated negatively with litter size (P=0.032) and varied with treatment (P=0.019) but not with maternal parity. Fetuses from pST-injected dams were heavier than those from ractopamine-fed dams (Figure 1). Placental weight (P=0.017) correlated negatively and the ratio of fetal: placental weight (P=0.042) correlated positively with litter size, but neither was altered by maternal parity or treatment (P>0.1 for each).



**Figure 1.** Effects of treatments on foetal weight (adjusted to a mean litter size of 11.3); \*differences between treatments were significant, P<0.05

Injecting pregnant sows with pST increases their food conversion efficiency and weight gain, which may improve sow longevity. Fetuses from pST-injected dams were heavier than those from ractopamine-fed dams, but not from controls.

We thank OzBioPharm Pty Ltd for donating Reporcin<sup>®</sup> and Elanco Animal Health Pty Ltd for donating Paylean<sup>®</sup> for this study.

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## Induction of oestrus during lactation using an injection of gonadotrophin and piglet separation

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The idea of inducing oestrus in the sow during lactation is not new. Studies over the last 50 years (reviewed by Armstrong *et al.*, 1999) have produced conflicting results when gonadotrophins were used during lactation to promote follicular growth and induce ovulation. We speculate that these strategies were unsuccessful because 24-hour suckling activity limits the release of luteinizing hormone, which restricts gonadotrophin support for the final stages of follicle maturation and oestradiol production. The objective of this study was to prove the concept that oestrus can be induced during lactation using a combined injection of pregnant mare serum gonadotrophin (PMSG) and human chorionic gonadotrophin (hCG) at three weeks after parturition followed by piglet separation from the sow for part of the day.

The study was replicated in time using two groups of four multiparous hybrid (mainly Large White x Landrace) sows. Both groups of sows were housed in farrowing crates in the same room. Parturition occurred within four days in each group of sows and litter size varied from 6-10 (mean 7.8) piglets. Each sow was offered voluntary access to a pelleted, commercial lactating sow diet and water was provided from a nipple drinker. Piglets in each litter were provided with supplementary heating and voluntary access to a pelleted, commercial piglet diet and a nipple drinker in each pen. Sows in each group were injected intramuscularly with 400 IU of PMSG + 200 IU of hCG (PG600; Intervet Inc) on the same day at 19-24 (mean 20.3) days after parturition. Following injection with PG600, oestrus was checked in each sow using the back-pressure test in the presence of a boar in front of the farrowing crate at 1600 hours and 0800 hours of each day until mating. After injecting each sow with PG600, piglets were separated at 1630 hours on each day until mating using a metal-mesh partition attached to the side of the farrowing crate. The metal-mesh partition was removed at 0830 hours on the following day to return the piglets to the sow. Oestrus was detected at  $5.1 \pm 0.2$  days (mean  $\pm$  SD) after injection with PG600 in seven of the eight sows. Each of the seven sows was removed from their farrowing crate and mated naturally to ensure a standing response in the presence of a boar. These seven sows were subsequently artificially inseminated within 8-16 hours in the farrowing crate using fresh semen. Piglet separation ceased after artificial insemination and the piglets remained with the sow until weaning at 35 days after parturition. The sow not detected in oestrus was re-mated six days after weaning. Mean piglet live weight was  $5.1 \pm 0.3$  kg and  $8.2 \pm 0.5$  kg at 20 and 35 days of age, respectively. Pregnancy was confirmed in all sows using ultrasound at 28 days after mating.

This study provided 'proof of concept' that oestrus can be induced during lactation using an injection of gonadotrophin at 19-24 days after parturition, combined with boar exposure and piglet separation until mating. The results suggest that separation of the piglets overnight for 16 hours per day allowed follicle development in the sow and the release of luteinizing hormone to induce ovulation. Importantly, pregnancy was maintained when piglets remained on the sow for a further 10 days after mating until weaning at 35 days of age. This study uncouples weaning from re-breeding in the sow and offers a new management strategy to re-mate the sow during lactation. Potential benefits of this strategy include an increase in the number of pigs born per sow per year and an increase in piglet weight at weaning.

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## Oestrus and ovulation responses of gilts vary with different gonadotrophin treatments

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When gilt availability is limited, a common method used to stimulate the onset of puberty is the injection of gonadotrophins such as 400 IU eCG plus 200 IU hCG (PG600). However, up to 30% of treated gilts do not show an oestrus response within seven days (Kirkwood, 1999). We hypothesize that this non-oestrus response is due to the hCG component of PG600 causing some gilts to either ovulate or luteinize follicles. In either event, the production of progesterone (P4) will block behavioural oestrus. If true, then injection of eCG alone (i.e. no hCG component) should result in an improved oestrus response.

To examine gilt responses to different gonadotrophin preparations, 109 prepubertal Yorkshire x Landrace gilts (90 kg, 153 d) were assigned by weight and age to receive an injection of 600 IU eCG (Pregnecol®, n=45), 400 IU eCG plus 200 IU hCG (PG600®, n=45), or serve as un-injected controls (n=19). From day two to day seven after time of hormone injection, gilts were exposed to a mature boar for 15 minutes daily to facilitate detection of oestrus. Fewer control gilts were employed because we anticipated few, if any, would exhibit oestrus during the seven-day study period. Blood samples were obtained from all gilts at hormone injection (day zero) and at day three, seven and 10 and assayed for P4 concentrations. All gilts had non-detectable P4 concentrations on day zero. An elevation in P4 to >1 ng per mL on day three was indicative of premature ovulation (or luteinization), while low levels on day three followed by elevations on day seven or day 10 was indicative of a normal ovulatory response. Before hormone injection, the first 25 gilts assigned to each hormone treatment were subject to an ultrasound examination of their ovaries and the diameters of the largest three follicles recorded. Treatment effects on follicle size and oestrus responses were examined by analysis of variance and Chi square, respectively (NCSS, 2005).

Oestrus response by day seven was 33/45 (73.3%), 7/45 (15.5%), and 0%, for PG600, eCG, and controls, respectively (Table 1). All oestrus gilts ovulated except for four of the PG600 gilts, and non-oestrus in PG600 gilts was not associated with a premature rise in P4. These results refute the hypothesis that non-oestrus in PG600-treated gilts is due an hCG-associated follicular luteinization or ovulation. The poor response to eCG suggests that at the gilt age used, follicular immaturity requires both FSH (eCG) and a LH-like (hCG) stimulation in order to drive follicular growth and eventual ovulation.

**Table 1. Effect of eCG (Pregnecol) or eCG plus hCG (PG600) on oestrus and ovulation in gilts**

	PG600	Pregnecol	Control
No. of gilts	45	45	19
Follicle diameter, mm <sup>1</sup>	2.4±0.1	2.3±0.1	--
Oestrus by day 7 (%)	73.3	15.6	0
Oestrus with elevated P4	64.4	15.6	0
Oestrus with no increase in P4	8.9	0	0
Anoestrus with elevated P4	2.2	0	0

<sup>1</sup> Means ± SE

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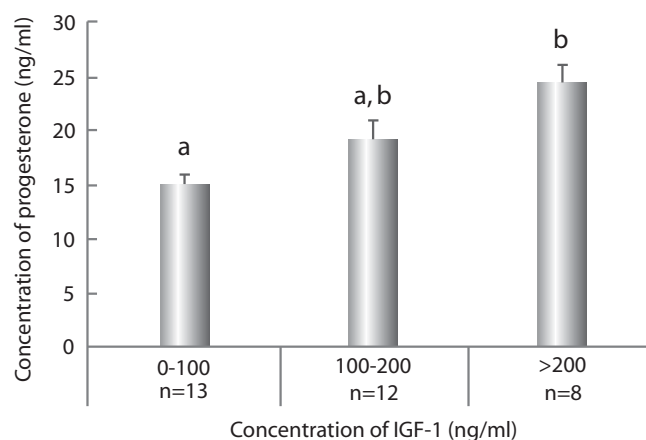
## Early luteal progesterone secretion is related to insulin-like growth factor 1 (IGF-1)

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Formation of corpora lutea (CL) and secretion of progesterone during the early luteal phase (first week after ovulation) occurs independently of lutenizing hormone (LH). Corpora lutea are not sensitive to LH in this period and even hypophysectomy does not prevent formation of normally functioning CLs after ovulation. There are other factors that influence the development of CLs and secretion of progesterone in this period (e.g. angiogenic and growth factors). Insulin-like growth factor 1 (IGF-1), for example, has been shown to stimulate progesterone secretion by luteal cells *in vitro*. The objective of this paper was to determine whether there is also a relationship *in vivo* between IGF-1 and progesterone secretion. Data were derived from multiparous (n=12) and primiparous (n=21) sows. Blood samples were taken at 6-12 hour intervals to quantify the postovulatory rise in progesterone. Seventy-two hours after ovulation, the progesterone concentration was estimated by linear fit of progesterone values for individual sows. One blood sample for IGF-1 was taken about one day after ovulation and at least four hours after the last feeding.

IGF-1 concentration varied considerably between sows, ranging from 36-351 ng/ml. Peripheral concentration of progesterone varied between 7.7-31.4 ng/ml. The progesterone concentration at 72 hours after ovulation was higher for sows with a higher IGF-1 concentration (Figure 1). Overall, the correlation between peripheral concentration of progesterone at 72 hours after ovulation and concentration of IGF-1 on the first day after ovulation was 0.6 (P<0.05). In conclusion, these data show a positive relationship between peripheral IGF-1 concentrations and progesterone secretion *in vivo* during the early luteal phase.



**Figure 1.** Relationship between peripheral IGF-1 concentration on the first day after ovulation and peripheral concentration of progesterone at 72 hours after ovulation; <sup>a,b</sup>P<0.05

## Supplementing lactating sow diets with ractopamine: effects on sow weight loss, milk composition and piglet growth

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Excessive mobilisation of skeletal protein during lactation is thought to be one of the primary causes of delayed return to reproductive function in weaned primiparous sows. Supplementing the diets of growing pigs with the beta-agonist ractopamine (Paylean®) increases net protein accretion, and causes redistribution, but not a reduction, in fat deposition (Dunshea, 1993; Bell *et al.*, 1998). The current study tested the hypothesis that ractopamine supplementation of lactating sows reduces maternal weight loss and alters milk composition and piglet growth.

Thirty six Large White x Landrace first parity sows were allocated to one of two treatment groups (n=18 sows). One group (Control) received a standard lactation diet (0.71 g avail. lys/MJ DE) throughout lactation, while the other group (Ractopamine) received the same standard lactation diet but supplemented with ractopamine at 10 parts per million (ppm) from days 1-13 of lactation and 20 ppm from day 14 of lactation until mating. The amount of feed offered each day was stepped up gradually, reaching 6 kg/day by day seven of lactation, fed over three meals per day. Sows were weighed and P2 backfat and maximum eye muscle depth measured by ultrasound on days 1, 14 and 20 of lactation, with litter size standardized to nine piglets within 24 hours of farrowing (day 0). Piglets received only maternal milk as their feed source and were weighed on days 1, 7, 13 and 20 of their mother's lactation. Maternal milk samples were collected on days 3, 13 and 20 of lactation and analyzed for fat and protein content.

Although total lactation weight loss was similar for Control and Ractopamine sows, weight loss between days 14 and 20 tended to be lower for Ractopamine compared to Control sows ( $0.36 \pm 0.73$  vs.  $2.5 \pm 0.98$  kg;  $P < 0.1$ ). Compared to Control sows, milk taken from Ractopamine sows contained a significantly higher fat concentration on day three of lactation, and significantly lower protein concentrations on days 13 and 20 of lactation (Table 1). Piglet weight gain between days 1-14 of lactation was unaffected by maternal diet; however, between days 14-19 of lactation weight gain was significantly lower for piglets suckling Ractopamine as opposed to control sows ( $260 \pm 0.01$  vs.  $310 \pm 0.01$  g/day;  $P < 0.01$ ).

**Table 1. Effect of dietary ractopamine in lactation on sow milk composition**

Lactation diet	Milk fat (g/l)			Milk protein (g/l)		
	Day 3	Day 13	Day 20	Day 3	Day 13	Day 20
Control	$66.4 \pm 2.04^a$	$59.8 \pm 1.43$	$57.7 \pm 1.53$	$45.6 \pm 0.59$	$42.3 \pm 0.37^b$	$41.6 \pm 0.51^b$
Ractopamine	$79.3 \pm 3.20^b$	$60.4 \pm 1.70$	$55.5 \pm 2.43$	$45.6 \pm 0.83$	$38.7 \pm 1.24^a$	$38.1 \pm 0.60^a$

<sup>ab</sup> within column indicates significant difference;  $P < 0.05$ .

The current data indicate that dietary ractopamine alters milk composition. Specifically, the decreased protein concentration in the milk of ractopamine supplemented sows supports the suggestion that Paylean® has an inhibitory effect on protein degradation which may be beneficial during periods of increased metabolic demand, such as lactation.

Paylean® provided by Elanco Animal Health Pty Ltd.

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## Increasing dietary energy levels minimizes weight loss but does not improve lactation performance in first-litter sows

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Tritton *et al.* (1996) demonstrated that high protein diets offered to lactating first litter sows could improve lactation performance, whereas increasing the energy level of diets failed to improve litter gain. However, body reserves of weaned sows were substantially depleted under low dietary energy intakes. Since these studies, first-litter sows have become leaner and voluntary feed intake may have reduced as a consequence of selection for progeny efficiency. Poor lactation performance and slow pre-weaning growth rates remain a major constraint to post-weaning growth, especially in gilt progeny. We tested the hypothesis that litter gain would be increased with dietary energy levels of lactation diets offered to genetically lean first-litter sows during summer.

Commencing in December, 284 Large White x Landrace F1 cross gilts at 110 days of pregnancy were selected for the study over 14 weeks. The gilts were allocated to one of five digestible energy (DE) levels offered in a mash form. The lowest and highest energy diets were formulated using similar ingredients varying in the dietary level of tallow. The low DE diet contained 13.0 MJ DE/kg, 218 g crude protein/kg, 11.7 g lysine/kg and 33 g crude fat/kg. The high DE diet contained 15.3 MJ DE/kg, 259 g crude protein, 13.8 g lysine/kg and 96 g crude fat/kg. Intermediate treatments were manufactured by blending of the low and high energy diets. Sows were fed three kilograms of their treatment diet daily to the day of farrowing and thereafter offered *ad libitum* until weaning at 26.9±0.1 days (mean±SE). Data were analyzed by General Linear Model analysis of variance.

Litter size born (10.5±0.1) and piglet birth weight (1.44±0.01) were unaffected by dietary energy level after dietary treatment feeding for six days (Table 1). Post-farrowing weight and backfat were similar between treatments (201.4±1.0 kg; 19.9±0.2 mm). Sow live weight loss between farrowing and weaning at 27 days was negatively related to dietary energy level in response to the extra energy intake. Increasing dietary energy content did not increase lactation performance in a linear response. First-litter sows restricted in energy intake will mobilize body tissue reserves, notably lean tissue, for milk production which may impact on subsequent reproductive performance. The results signify there are factors other than energy constraining lactation in commercial genotypes.

**Table 1. The effect of dietary digestible energy level on the performance of first-litter sows**

Dietary energy level (MJDE/kg)	13.0 (n=57)	13.6 (n=58)	14.2 (n=54)	14.7 (n=59)	15.3 (n=56)	SED	Linear response to DE level
Litter growth rate (kg/d)	1.79	1.64	1.84	1.78	1.79	0.03	NS
Piglet growth rate (kg/d)	0.201	0.192	0.209	0.205	0.209	0.002	NS
Piglet wean weight (kg)	6.9	6.7	7.1	7.0	7.1	0.07	NS
Sow intake (kg/d)	4.7	4.7	4.6	4.7	4.7	0.04	NS
Sow weight loss (kg)	18.6	12.5	15.4	12.4	10.2	0.7	P<0.01
Sow P2 loss (mm)	3.5	3.3	3.4	2.7	2.8	0.2	NS

NS Non significant response (P<0.05).

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## Impact of time of grouping after insemination on the fertility of sows

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While gestation stalls enable ease of artificial insemination (AI) and individual feeding, they are criticized on animal welfare grounds because of the degree of physical restriction they impose. This has resulted in an increased movement towards group housing of sows during gestation. The usual field advice is that sows should not be mixed until 30 days after breeding to allow for the completion of placentation. However, to our knowledge, this recommendation is not evidence based so we performed an experiment to test the hypothesis that mixing sows before 30 days of gestation would reduce sow fertility. A total of 617 mixed-parity weaned sows were assigned at the time of AI either to individual gestation crates or to groups of 15. Each group of 15 unfamiliar sows comprised about three sows at each of 2, 7, 14, 21 and 28 days from breeding. All sows in a group were mixed at one time. Sows were floor-fed once daily (~2.5 kg per sow) using a standard gestation diet. After five weeks in groups, sows were re-housed in individual stalls until farrowing. Litter size data were examined by GLM analysis of variance and the effect of day of gestation at grouping on farrowing rate was compared to the non-grouped control sows using Log likelihood Chi square test.

There was no effect of grouping *per se*, or day of gestation when grouped, on subsequent litter sizes (Table 1). However, the farrowing rate tended ( $P=0.085$ ) to be reduced for sows mixed at 14 days of gestation. These data indicate that sows can be successfully mixed and group-housed during gestation. However, there was a tendency for a lower farrowing rate of sows mixed during the period around 14 days after breeding (i.e. the period of initial maternal recognition of pregnancy). A reduction in farrowing rate for sows mixed at 10 days post insemination has been noted earlier (Te Brake and Bressers, 1990). Therefore, it would be prudent to avoid mixing sows 10-14 days after insemination.

**Table 1. Effect of time of grouping after insemination on farrowing rate and mean ( $\pm$ SE) litter size of group-housed sows**

Pregnancy day	No. sows	Farrowing %	Litter size (total)	Litter size (live)
Control <sup>a</sup>	122	82.0	11.6 $\pm$ 0.3	10.6 $\pm$ 0.3
Day 2	98	77.5	11.0 $\pm$ 0.4	10.2 $\pm$ 0.4
Day 7	97	75.3	11.2 $\pm$ 0.4	10.3 $\pm$ 0.4
Day 14	101	72.3 <sup>b</sup>	11.6 $\pm$ 0.4	10.7 $\pm$ 0.3
Day 21	101	83.2	11.4 $\pm$ 0.4	10.4 $\pm$ 0.3
Day 28	98	82.6	11.5 $\pm$ 0.3	10.6 $\pm$ 0.3

<sup>a</sup> Sows maintained in individual stalls; <sup>b</sup> different from Control,  $P=0.085$  by Chi Square.

Supported by the USA National Pork Board and the University of Guelph/Ontario Ministry of Agriculture, Food and Rural Development Animal Research Program.

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## Mixing in early pregnancy: effects on embryo survival

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Group housed sows typically have lower pregnancy rates and litter sizes (Kongsted, 2005). While the stress resulting from sow aggression following mixing during the period of embryo implantation can be detrimental to embryo development, the effects of remixing and stress during the pre-implantation period (days 1–10 of gestation) on embryo development and survival are poorly documented (Razdan *et al.*, 2003). In light of this, we determined the effects of re-grouping and the timing of re-grouping during the pre-implantation period on embryo mortality. Forty-eight Large White x Landrace crossbred sows were selected at 18 weeks of age and housed in unchanging groups until their second oestrus. At 25 weeks of age, the combination of PG600 and 20 minutes of daily, physical boar contact was used to stimulate puberty, with boar contact resuming 12 days after first detection of oestrus and gilts receiving two artificial inseminations (AIs), 24 hours apart, at their second oestrus. After their first AI, gilts were allocated to one of four treatment groups (n=12 gilts/treatment). Gilts in one treatment group were housed individually in stalls (STALL). The remaining gilts continued to be housed in their pre-AI groups and were either not re-mixed (NOMIX), or remixed to form new groups on day 2/3 (REMIXD2/3) or day 8/9 (REMIXD8/9) of gestation (day 0 = day of first detection of second oestrus and first insemination). Group housed gilts were housed in groups of six, with a space allowance of 2.4 m<sup>2</sup>/gilt, and were fed once each day (2.2 kg/gilt). Reproductive tracts were collected on day 26±0.2 of gestation, and the number of corpora lutea (CL) and viable embryos counted.

Embryo survival on day 26 of gestation was similar for all four treatment groups (P>0.05) (Table 1). Compared to gilts in the STALL and NOMIX groups, a numerical, but not significant, increase in the number of embryos present on day 26 of gestation was observed for gilts remixed on days 2/3 or 8/9 (P>0.05); however, this reflected the higher ovulation rate of these animals, rather than any increase in embryo survival rate. In conclusion, the current data indicates that mixing gilts during the first 10 days of gestation does not have a negative effect on embryo development and survival.

**Table 1. Effects of re-mixing sows in early pregnancy on embryo survival**

Gestation housing treatment (n=12 gilts/treatment)	Ovulation rate (number of corpora lutea)	Number of embryos	Embryo survival (%)
Stall	14.9±0.14	12.8±1.07	86.0±0.06
No-mix	14.8±0.57	12.2±0.85	83.0±0.05
Re-mix 2/3	15.6±0.75	14.6±0.94	93.0±0.03
Re-mix 8/9	16.0±0.82	14.0±1.07	89.0±0.05

Project funded by the South Australian Pig Industry Advisory Group.

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# 7

## Gut health and development

## Symposium – gut health in the pig

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### Introduction

‘Gut health’ is a term that has only relatively recently entered the lexicon used in pig production. Its genesis essentially coincided with the events in Europe associated with restrictions and then a ban on the use of antibiotic growth promotants in diets. Since the late 1990s, the expression has been omnipresent in the pig literature as a plethora of opinion, products and research have appeared on the subject. The factors and conditions involved in ‘gut health’ are multi-factorial, complex, and in general incorrectly interpreted. In addition to enteric disease, other influences will also impact upon ‘gut health’ such as responses occurring in the gastrointestinal tract after weaning, any changes that might occur after a change in diet, and the situation in the gut before, during and following a disease challenge. Unfortunately there is some confusion as to what an appropriate definition of ‘gut health’ actually is, and this has probably caused some confusion and misinformation in relation to the issue.

The first paper in this symposium by John Pluske and colleagues attempts to address this question. The paper provides a broad overview of ‘gut health’ and introduces a working definition, which essentially relates to a generalised condition of homeostasis in the gastrointestinal tract in view of the various challenges and perturbations that occur to, and in, this important organ system. This paper addresses some key and salient aspects associated with gastrointestinal health with a particular focus on enteric disease, which is where most of the attention in relation to ‘gut health’ has been focused. A prime example of this is with post-weaning colibacillosis and the use of replacement feed additives to antibiotic feed additives.

The second paper in the symposium by Colm Moran provides an industry-oriented and pragmatic approach to how replacement products to antibiotic feed additives could be developed. An understanding of the various mechanisms of action of antibiotic feed additives is a requisite first step in such a process, and although understanding in this area is not complete, sufficient evidence exists to allow the development of such products.

The overall aim of this symposium is to inform and educate the attendees in an area of pig science that we have all heard and read about a lot in recent times. We hope that this symposium will set a platform for discussion of ‘gut health’ in the context of the Australasian pig industry.

# Gut health in the pig

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## Abstract

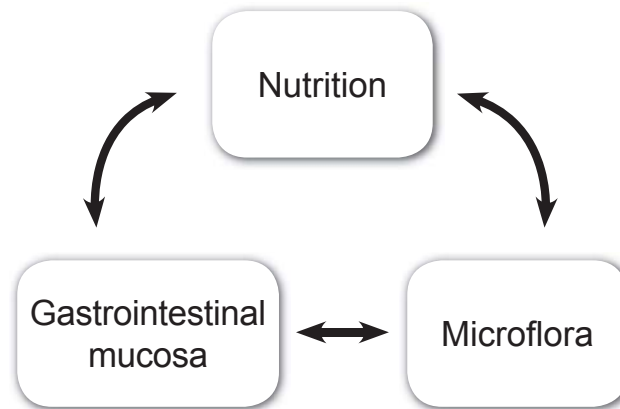
Gastrointestinal disturbances can cause large economic losses in the pig industry. Diseases and conditions of the gastrointestinal tract (GIT) that can cause economic loss have generally been controlled by the use of dietary (and or in the water) antimicrobial compounds, such as antibiotic feed additives and (or) minerals such as zinc and copper. However the implementation of legislation in some parts of the world, for example the European Union, and a growing sentiment worldwide to reduce the use of dietary antimicrobial compounds, has caused a reassessment of measures to influence GIT 'health' and caused enormous interest in alternative means to control diseases and conditions of the GIT. There are now available a wide array of products and strategies available to the pig industry that influence 'gut health'. The products in the market place are characterised predominately not only by their (claimed) different modes of action, but also by the variation in responses seen when offered to pigs, and not only in the post-weaning period. This variation is presumably a consequence of the many different conditions of management that pigs are under, that in turn influences factors such as composition of the microbiota and mucosal immunity. Other strategies, such as the manipulation of particle size and changing the protein content of a diet, might also be adopted to influence the expression of enteric pathogens and the expression of disease. Ultimately, the cost-benefit of adopting such practices to influence gastrointestinal 'health' requires consideration.

## Introduction

'Gut health' is a term that we define as describing a generalised condition of homeostasis in the gastrointestinal tract (GIT) of the pig. The factors and conditions involved in 'gut health' are multifactorial, complex, and currently poorly described and sometimes incorrectly interpreted, although it is evident that perturbations of the GIT that cause disease, even sub-clinical disease, are a disturbance of this homeostasis. In addition to enteric disease, other influences will also impact upon gut health, such as the responses occurring in the GIT in the period immediately after weaning, any changes that might occur after a change in diet, and (or) disruptions to meal patterns and hence the flow of nutrients through the GIT.

Simplistically, 'gut health' can be viewed as an outcome (positive, negative, the status quo) of the complex interactions occurring in the GIT between nutrition (e.g. feed type, feed composition, presence or absence of antibiotic feed additives), the mucosa of the GIT (e.g. type of receptors, mucin, inflammatory activity, cytokine production, barrier function), and the microbiota (e.g. density, location, pathogenicity, extent of colonisation) (Figure 1). The vast array of possible interactions means that analysis that encapsulates the dynamic nature of this organ cannot be achieved with many of the analytical tools currently available. Consequently scientific analysis, and hence interpretation, is usually resistant to a reductionist approach to these issues (Niewold, 2006). Examining the system holistically has not been possible previously, however gene expression profiling using microarray technology (e.g. Niewold, 2006) and (or) the emerging field of metabolomics (e.g. Bertram *et al.*, 2007) hold promise as means of unravelling the regulatory mechanisms and transcriptional networks that underlie these most complex of biological processes. Further description and interpretation of this methodology is beyond the scope of this paper, however.

Nevertheless, when attempting to assess gut function and gut health, it is useful to view any particular assessment in a number of ways that extend from the whole body level to the molecular level. To understand the basic mechanisms during normal and pathological gut development, a useful approach is to assess gut function at as many levels as possible as this will provide maximum knowledge about any treatments applied and each experimental animal used (Figure 2). Analysis of many different parameters can cause conflicting observations that are difficult to interpret. However, relying too much on single parameters is more dangerous as this may lead to erroneous conclusions.



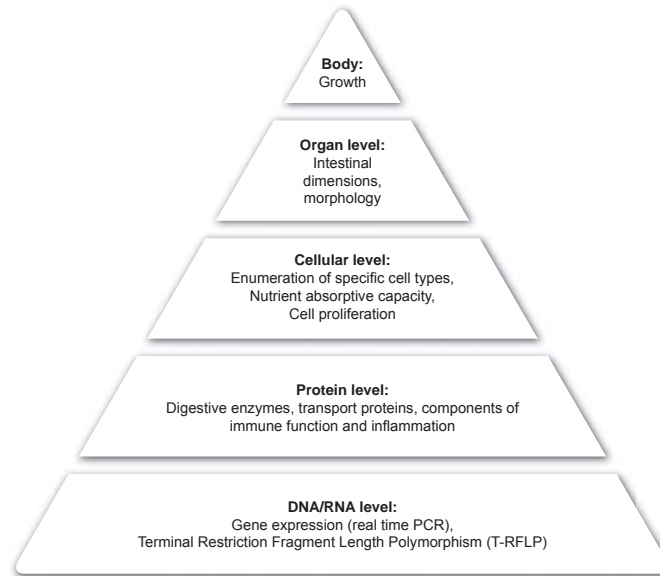
**Figure 1.** A schematic representation of the complex mutual interactions that occur between ‘nutrition’, the ‘gastrointestinal mucosa’, and the ‘microflora’ (adapted from Nisnevold, 2006)

A discussion of ‘gut health’ cannot proceed without acknowledgement of the various mechanisms mooted for the positive actions of antibiotic feed additives on the host. These have been described previously (see reviews by Visek, 1978; Anderson *et al.*, 2000; Gaskins, 2001; Pluske *et al.*, 2002; Page, 2006; Wegener, 2006), have been covered in the companion paper by Moran (2007), and will not be reiterated in this paper. Nevertheless, the benefits of antibiotic feed additives to pig production can broadly be categorized into production benefits, disease control benefits, prevention of metabolic and (or) fermentative disorders, and numerous other related benefits such as protein and energy sparing (Page, 2006). In turn, these can have environmental benefits, for example through reduced nutrient excretion. It is rare therefore, if not impossible, to find a feed additive/nutritional strategy that can elicit the same effects that an antibiotic feed additive has when benefits are realised. In this respect, it is unlikely that any single feed additive/nutritional strategy will fully enhance the ‘health’ of the GIT to the same degree as an antibiotic feed additive when the GIT is compromised.

The intent in this paper is to provide some context for the use of alternatives to antibiotic feed additives, by briefly discussing modes of action and attempting to define and highlight the basic principles of ‘gut health’. The companion paper in this symposium by Dr Colm Moran will cover this in more detail. This paper will then discuss some of the biological issues surrounding the notion of ‘gut health’, using examples where appropriate. It is hoped that this paper will provide readers with a greater understanding of some of the major issues surrounding ‘gut health’ and instigate debate and discussion of the topic, particularly in an Australasian context.

### Antibiotic feed additives

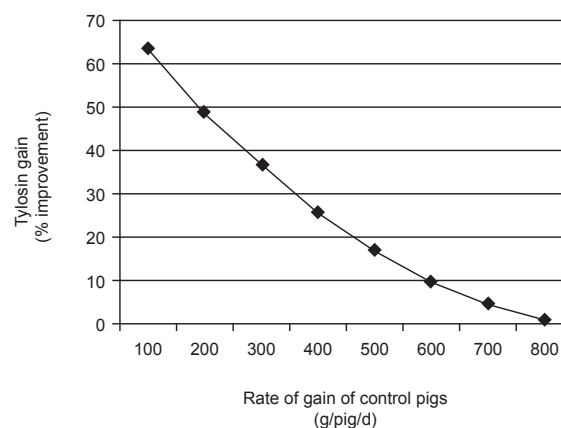
Antibiotic feed additives have been used extensively and successfully in pig feeds over many years to improve health and performance, but only appear to be effective under certain circumstances. Risks associated predominately with antibiotic resistance of microbes in man have caused the banning or restriction of antibiotics as feed additives for pig production in some parts of the world (the precautionary principle). This has caused an explosion in both the use and development of alternative strategies to antibiotic feed additives, and in particular those feed additives/nutritional strategies that alter (or are claimed to alter) the microbiota of the gastrointestinal tract (GIT) to somehow modulate the ‘health’ of this organ system. It has also caused a number of large collaborative projects [e.g., “Sustainable Systems for Weaner Pig Production” (a UK-based project), “Defining and validating gut health criteria in young pigs, based on digestive physiology, microbiology and mucosal immunology investigations for testing alternative strategies to in-feed antibiotics” (an EU project), “Feed for Pig Health (Development of Natural Alternatives to Antimicrobials for The Control of Pig Health and Promotion of Performance) and REPLACE (Plants and Their Extracts and Other Natural Alternatives to Antimicrobials in Feeds)” (EU projects)] aimed at understanding further the mechanisms underlying this phenomenon. Although these projects mainly concern the newly-weaned pig, it is important to consider that other pig phases, for example growing-finishing pigs and sows, also suffer from GIT disruptions causing decreased production, morbidity, and sometimes death.



**Figure 2.** An example of the organisational hierarchy to define the parameters of gut function

The change in production systems in the 1940s and 1950s from one predominately based on pasture, where plant and animal foods were consumed, to controlled indoor environments, where pigs were fed prepared vegetable-based diets, underscored the inadequacy of diets based on plants for optimum production. Including animal protein products in diets for indoor pigs showed that normal growth could be restored (Page, 2006). Subsequently it was found that the factor responsible for this restoration in growth was vitamin B12. In a case of serendipity, researchers trying to find a reliable and high-producing source of vitamin B12 in *Streptomyces aureofaciens* isolated chlortetracycline, which was the growth-promoting fraction over and above the independent effect of vitamin B12 (Page, 2006). Since this time a plethora of studies have been conducted attempting to explain the mode(s) of action of antibiotics and, unsurprisingly given the diversity of antibiotics present, no common mechanism(s) has/have ever been found. Coates *et al.* (1952) commented that “it is unlikely that a single mode of action can explain all the results reported in the literature”, with respect to the use of antibiotics in chickens. Little has changed in 55 years.

A feature of using antibiotic feed additives in pig production is the variation in responses that occur. Braude *et al.* (1953; cited by Page, 2006) summarised a large number of experiments from the time and concluded that the relative improvement in growth rate resulting from adding antibiotics to diets for pigs was inversely related to the growth rate of control animals. This is corroborated by the research of Melliere *et al.* (1973) (Figure 3). Hays (1991) commented that the “response is greater during critical stages of production such as weaning...”, and that “environmental stresses such as inadequate nutrition, crowding, ... poor sanitation ... also contribute to increased responses”. A study by Dritz *et al.* (2002) with more than 24,000 pigs in three multi-site production systems in Kansas showed that only nursery pigs responded to in-feed antimicrobials, noting that the performance of non-supplemented control pigs was very high consistent with decreased microbial challenge and high levels of hygiene.



**Figure 3.** The impact of the performance of control animals on the magnitude of the response to tylosin in pigs (redrawn from Melliere *et al.*, 1973)

What does all this mean for 'gut health' in pigs? The implication is that responses to any in-feed alternatives to antibiotic feed additives are also likely to be variable, and therefore it is no surprise that such findings are found commercially. The literature is littered with studies that show beneficial responses and enhanced 'gut health' in pigs and poultry to alternative dietary products to antibiotic feed additives, but less evident are studies showing a lack of, or even negative, responses. Critically, the circumstances and (or) conditions whereby these (lack of) effects occur are often not cited, making it even more difficult to discern the reasons for the variability that occurs. Examples of exceptions are the meta-analysis described by Miguel *et al.* (2004) assessing the efficacy of the product BioMos®, and the work described by Rosen (2006) focusing on the construction and application of holo-analytical predictive empirical models to objectively assess alternatives to antibiotic feed additives in the intensive animal industries. Studies such as these at least document the variation that surrounds a response to a feed additive, but they offer little or no comfort to those seeking a unifying product or strategy (elixir) to replace antibiotics in diets, if indeed that is the intention.

Many hypotheses have been proposed to explain the mode(s) of action of the antibiotic feed additives without any consistent consensus. Germ-free animals benefit very little from dietary antibiotics clearly showing that the microbiota is a critical intermediary in any positive effects observed. Page (2006) summarised the hypotheses already proposed and tested to explain these effects, and they include:

- Reductions in total numbers of bacteria in the GIT with decreased competition between the microflora and the host for nutrients;
- Inhibition of 'harmful' bacteria that may be pathogenic and (or) capable of producing toxic metabolites, particularly from protein fermentation;
- Inhibition of bacterial urease, to prevent formation of NH<sub>3</sub>;
- Improved energetic efficiency of the GIT through reduced weight, reduced enterocyte turnover and increased rate of glucose uptake;
- Inhibition of bacterial cholytaurine hydrolase activity, which reduces the level of lithocholic acid;
- Nutrient sparing;
- Improved nutrient absorption effected by morphological changes to the small intestinal epithelium;
- Modification of intestinal enzyme activity, since the characteristics of intestinal enzyme activity are influenced significantly by the microflora;
- Reduced immune stimulation, via a reduction in sub-clinical microbial challenges; and
- Stimulation of intestinal synthesis of vitamins by bacteria.

Saliently, these hypotheses confirm that any alternative/replacement feed additives are most unlikely to fully capture the multitude of effects imparted by the use of antibiotics. Furthermore, it is probable that no one single antibiotic feed additive can achieve all these effects because they have different modes of action.

### **Barrier function of the gastrointestinal tract**

Any discussion pertaining to 'gut health' cannot occur without reference to the microbiota that inhabit the GIT of the pig and their influence on barrier function. It is not our intention to describe the generalized features of the GIT microbiota because these have been adequately portrayed in many other review articles. However, there is a diverse assemblage of bacteria that varies in population density and diversity in different compartments of the GIT and at different stages in the life of a pig (Hampson *et al.*, 2001; Zoetendal *et al.*, 2004). Additionally, the microbiota (commensal, pathogenic) are intimately involved in 'cross talk' between the enteric bacteria and the host, with the chemistry and distribution of bacterial binding sites on gut mucosal surfaces playing key roles in determining host and tissue susceptibility and in triggering host responses, especially in young animals (Kelly and King, 2001). Individual mucin carbohydrates have the capacity either to repel or bind to microbial surface adhesins. In the case of pathogenic bacteria, protection against microbes lies in the capacity of mucin carbohydrates, particularly in the small intestine, to either repel or bind microbial adhesins (Belley *et al.*, 1999). The nature of the diet, the microbiota, and interactions between them influence the composition and functional characteristics of intestinal mucins (Montagne *et al.*, 2004). The taxonomy and distribution of bacterial groups that preferentially reside within the mucous layer must be better defined to ascertain the role of 'normal' gut bacteria in mucogenesis and mucolysis (Deplancke and Gaskins, 2001).

A key issue when discussing 'gut health' is that of barrier function. A good example where barrier function is compromised occurs in the immediate post-weaning period, where weaning causes a 'leakier' small intestine

(Spreeuwenberg *et al.*, 2001; Verdonk *et al.*, 2001; Boudry *et al.*, 2004). Inflammation of the intestine is associated with increased permeability that may lead to translocation of toxins, allergens, viruses or even bacteria. If and when bacteria cross this first line of defence and reach the lamina propria, their metabolites or mediators liberated from epithelial cells may cause an inflammatory response (Gaskins, 1997), and in this case the measurement of pro-inflammatory cytokines provides some information as to the degree of local inflammation (Johnson, 1997). Therefore weaning *per se*, and essentially the period of anorexia that occurs immediately after weaning, causes an inflammatory response (McCracken *et al.*, 1995; Pie *et al.*, 2004) that initiates perturbations to 'gut health'. In this case, simply encouraging pigs to eat more feed after weaning should ameliorate these responses and stabilize 'gut health'.

### Factors influencing the activity and composition of the GI microbiota

Myriad of factors influence the diversity and activity of the GIT microbiota, including the age of the pig and the environment it inhabits, antimicrobial agents (antibiotic feed additives and minerals such as Zn and Cu), dietary composition (e.g., carbohydrate type and content, protein type and content), feed additives (e.g., organic acids), feed processing (grinding, meal vs pellets), feeding methods (e.g., fermented liquid feeding), disease load, weaning, season, stress and genetics. These factors, which can interact, can make the study of the gut microbiota difficult.

Colonization of the GIT by the microbiota plays a critical role not only for the overall well-being of the pig, but also for its nutrition, performance, and quality of the products produced (e.g., production of skatole in the hindgut and effects on meat quality). Apajalahti and Kettunen (2006) reviewed the microbiological activities that could be considered important in assessing the health and (or) performance of the host. These included activities affecting nutrient and energy partitioning between the microbiota and the host, those connected to pathogenesis and disease development, activities supporting or suppressing immunological mechanisms, and activities of various metabolic pathways of bacteria that may manufacture harmful and (or) beneficial end products during their metabolism. Table 1 summarizes these principles and shows that all phenomena are connected to the microbiota, their outer cell wall structure, their growth and their general metabolism. Apajalahti and Kettunen (2006) remarked that because microbiota in the GIT obtain the majority of substrate for growth and metabolism from the host's diet, then it is evident that the listed characteristics of the intestinal microbial community can be influenced by feed composition and (or) feed type. This can have consequences for all phases of pig growth and pig production, from the lactating sow to suckling and newly weaned pigs, through to the finishing pig.

**Table 1. Examples of microbial activities relevant for the health and performance of animal hosts (from Apajalahti and Kettunen, 2006)**

Example of microbial activity	Target Level	Risky Level
General bacterial growth in small intestine	Restricted	Overgrowth
Effect on intestinal morphology	Stabilising	Disrupting/deforming
Pathogen growth	Suppressed	Favoured
Pathogen adherence	Blocked	Facilitated
Immune activation	Stimulatory	Suppressive
Lactic acid production	Normal	Accumulation
Putrefactive activity (e.g., indole, cresol, skatole, NH <sub>3</sub> )	Restricted	Strong

### Some influences on the GI microbiota and 'gut health'

It is beyond the scope of this paper to provide a comprehensive discussion of all nutritional influences on the GIT microbiota. A plethora of papers and reviews coinciding with the lead-up to the European Union's ban on antibiotic feed additives have been written (e.g., Jensen, 1998; Gaskins, 1997, 2001; Hillman, 2001; Pluske *et al.*, 2002; Hopwood *et al.*, 2005; Lallès *et al.*, 2007). However, one question that generally arises in discussions relating nutrition to the GIT microbiota, and particularly the area of 'gut health', is: What is 'normal' when referring to the health of the pig GIT?

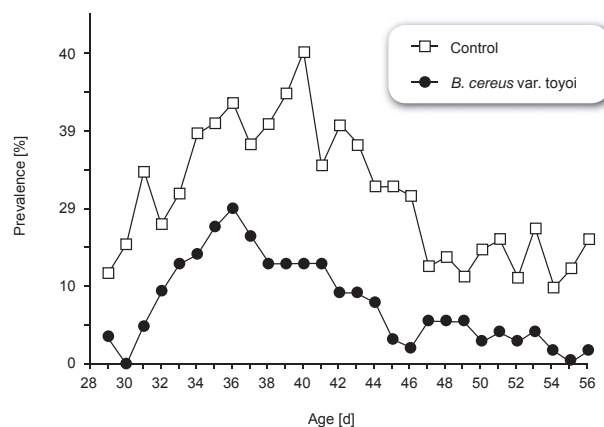
Hillman (2004) suggested that emphasis should be placed on an 'optimal' microbiota being present in the GIT rather than a 'normal' microbiota, mainly because defining what is 'normal', given the wide array of conditions pigs are grown under and methodological shortfalls, is difficult and problematic. Furthermore, there is widespread use, particularly in the popular press, of the description of bacteria as being either 'beneficial' (good) or 'detrimental' (bad).

For example, it is generally assumed that *Bifidobacteria* and *Lactobacillus* are 'good' bacteria whereas *Enterobacteriaceae* (i.e. the coliforms) are 'bad' bacteria, despite a large number of species and strains in the latter being of a non-pathogenic (avirulent) nature (Chapman *et al.*, 2006). Potential enterotoxigenic strains of *E. coli* exist in all herds, but post-weaning colibacillosis (PWC) does not necessarily occur in these herds. The presence or absence of a pathogenic organism, therefore, may not necessarily predict that disease will occur unless numbers proliferate to such an extent to overwhelm the general microbial population in the GI tract, or more specifically a specific region of the GI tract.

Hillman (2001) showed that intestinal *lactobacilli* having anti-pathogenic ('probiotic') activity against pathogenic *E. coli* that possessed K88 fimbriae were not evenly distributed across 19 Scottish pig farms. The variation in anti-pathogenic efficacy could be responsible for the variation seen across farms in, for instance, the efficacy of probiotics and other additives that purportedly affect specific bacterial populations. This is because adding an antimicrobial to the GIT where there is a population of bacteria already possessing a high indigenous antimicrobial activity is likely to be less effective than adding it where there is a population that shows little or no pre-existing antimicrobial activity (Hillman, 2001). In this regard, Hillman (2001) commented that it is the consistency of activity (e.g., across farms, across diets, across seasons) rather than the degree of activity that is currently poorly understood with respect to the large number of antibiotic replacements/alternatives on the market at present, and this possibly precludes their wider use. A current project within the Pork CRC under the supervision of Dr James Chin (NSW Department of Primary Industries) is investigating this aspect of probiotics further.

With regard to efficacy of probiotics after weaning, is the fact that we are relying on the intake (feed, water) of the bacteria/bacterium at a time when feed intake is both low and variable (Pluske *et al.*, 1997) simply may not be giving the product(s) any chance at all of succeeding? The published data defining the benefits of probiotics for nursery pigs are equivocal, which is no real surprise given the different species and strains that are used and the wide array of weaning and feeding conditions that products work under (Pluske, 2006). Differences in herd-health status undoubtedly contribute to the ambiguity in efficacy seen, as alluded to previously. In addition, recent research (e.g., Liong *et al.*, 2007) shows that oral synbiotics (probiotics + prebiotics) can have powerful metabolic effects in the pig, so should these type of combinations be investigated further? Regardless of this, is there a way of delivering probiotics that can circumvent the usual problems of low feed/water intake after weaning? Indeed, should probiotics be administered at birth when the GIT is sterile?

Work with fermented liquid feed (Demeckova *et al.*, 2002) and *Bacillus cereus* var. *toyoi* (e.g., Taras *et al.*, 2005) suggests that an altered microbiota in the faeces of the dam caused by changing the microbiota of the sow exerts a beneficial influence on both pre- and post-weaning development of the young pig. There is also some suggestion that feeding spores of *B. licheniformis* and *B. subtilis* (Alexopoulos *et al.*, 2004) alters milk quality in sows, which is pertinent given the current interest in Australia and the Pork CRC regarding the performance of progeny derived from gilt or sow litters. In the study by Taras *et al.* (2005), one group of sows were fed for 17 weeks, from day 24 after mating to day 28 after farrowing, and the piglets from these sows were fed for 6 weeks, from day 15 of lactation to 8 weeks of age. The control group of sows/piglets did not receive the probiotic strain. The *Bacillus cereus* var. *toyoi* strain was recovered from the faeces of sows and piglets throughout the trial, including the period 0-14 days of age before introduction of the starter diet occurred, and there was an improvement in FCR of pigs in the post-weaning period derived from sows fed the probiotic during pregnancy and lactation. Of particular interest in the weaned pigs offered the probiotic was a significant reduction in the incidence of liquid faeces (Figure 4) and post-weaning diarrhoea. Diets did not contain any antimicrobial agents, suggesting that this particular probiotic strain reduced the proliferation of enterotoxigenic *E. coli* in the GIT of weaned piglets.



**Figure 4.** Prevalence of liquid faeces (consistency score 4-5) during the total post-weaning period (d 29-56) of piglets in the Control (open boxes) and probiotic (*B. cereus* var. *toyoi*) group, respectively (after Taras *et al.*, 2005)



### Good bacteria versus bad bacteria: is there such a thing?

In an attempt to develop a predictor of 'gut health', Hillman (2004) advocated the use of a faecal lactobacilli:coliform bacteria ratio as an indicator of a pig's ability to resist infection and therefore enhance/maintain 'gut health'. A higher ratio (at least 100:1) indicates a 'healthy' gut while a low ratio (less than 100:1) places the pig at a greater risk of infection. Hillman (2004) commented that a simple test such as this using faeces would help ensure that pigs can be maintained as close as possible to optimum performance because gut 'health' is optimised.

Such a predictor obviously requires further validation, however it is seemingly ironic that Anderson *et al.* (2000) remarked that the class of microbes that appear to depress pig growth the most through toxin production, namely the Gram-positive facultative anaerobes that includes strains of *Lactobacillus* and *Enterococcus*, are also often used as probiotics; in the case of *Lactobacillus*, this would increase the ratio when perhaps it should be decreased. Furthermore, Mikkelsen *et al.* (2003) reported that the population of *Bifidobacteria* in GIT samples from piglets was numerically very low, which further complicates any clear explanation of links between 'good' bacteria and overall GIT function given the massive microbial diversity and overall population size present.

Further complications arise when the type of antibiotic feed additive is investigated. In an excellent study using molecular PCR-DGGE and quantitative polymerase chain reaction (qPCR) techniques to identify bacteria in the ileum of weaned pigs fed either tylosin or an antibiotic rotation sequence (week 1: chlorotetracycline sulfathiazole penicillin; week 2: bacitracin and roxarsone; week 3: lincomycin; week 4: carbadox; week 5: virginiamycin), Collier *et al.* (2003) found that sequence analysis of treatment-specific DNA bands identified three *Lactobacillus*, one *Streptococcus* and one *Bacillus* species that were diminished with the antibiotic rotation treatment, whereas tylosin selected for the presence of *L. gasseri*. *Lactobacillus*-specific qPCR was performed and analyzed as a percentage of total bacteria and demonstrated that total bacteria were decreased by tylosin and the antibiotic rotation treatments, whereas the percentage of lactobacilli increased by d 14 and through d 28 in tylosin-treated pigs. Collier *et al.* (2003) concluded that the decrease in total bacteria by antibiotics might reduce host-related intestinal or immune responses, which would divert energy that could otherwise be used for growth. Conversely, the ability of tylosin to improve animal growth may relate to its apparent selection for lactobacilli, commensals known to competitively exclude potentially pathogenic species from colonizing the intestine.

An issue that is increasingly discussed with reference to 'gut health' and the GIT microbiota is that of diversity, with the general view being that a greater bacterial diversity is beneficial for 'gut health'. But is this view consistent, and does it provide a meaningful basis for comparing antibiotic feed additives to alternatives? Konstantinov *et al.* (2003) reported that feeding fermentable carbohydrates in the form of sugar-beet pulp (100 g/kg diet) or fructooligosaccharides (FOS; 2.5 g/kg diet) to piglets after weaning (at 28 days) increased bacterial diversity and promoted a more rapid stabilisation of the bacterial community (by day 5 post-weaning) compared to pigs fed neither of these products. This was related to the predominance of several bacterial genera and species that, the authors postulated, were involved in the utilisation of dietary fibre sources such as sugar-beet pulp and FOS. However, no production benefits or effects on diarrhoea were reported. Conversely, Collier *et al.* (2003) found that the use of antibiotics caused a homogenization (decrease in diversity) of the microbiota they the authors' believed might explain, in part, the homogenized growth performance commonly observed in groups of animals fed antimicrobial growth promoters (Schwarz *et al.*, 2001). Collier *et al.* (2003) commented that microbial homogenization might ultimately prove to be a target mechanism for alternatives to antimicrobial growth promoters in diets for pigs.

### Zinc oxide – why does it work?

Zinc oxide (ZnO) is a non-antibiotic product that appears to influence and benefit 'gut health'. Numerous studies have shown the production and (or) anti-diarrhoeal benefits of including ZnO at supraphysiological levels (i.e. 2000-3000 mg/kg or ppm) in the diet after weaning, but what is less clear is/are the mechanism(s) whereby ZnO exerts its beneficial effects. Reported effects include the increased gene expression of antimicrobial peptides in the small intestine (Wang *et al.*, 2004), positive effects on the stability and diversity of the microflora, particularly with respect to coliforms (Katouli *et al.*, 1999), increased IGF-I and IGF-IR expression in the small intestinal mucosa (Li *et al.*, 2006), bactericidal functions (Jensen-Waern *et al.*, 1998) and reductions in electrolyte secretion *in vitro* from enterocytes (Carlson *et al.*, 2006). Hedemann *et al.* (2006) found changes in some pancreatic enzymes and mucin staining but concluded that there were no definite answers as to how the growth promoting and diarrhoea-reducing effects of excess dietary Zn were exerted. Nevertheless, some studies have reported no benefits of feeding ZnO (e.g., Jensen-Waern *et al.*, 1998; Broom *et al.*, 2006).

A study by Hojberg *et al.* (2005) using 2500 ppm ZnO showed reduced bacterial activity (ATP accumulation) in digesta from the gastrointestinal tracts of newly-weaned piglets compared to that in animals receiving 100 ppm ZnO. The numbers of lactic acid bacteria and lactobacilli were reduced, whereas coliforms and enterococci were

more numerous in animals receiving the high ZnO dose. These authors surmised that the influence of ZnO on the GIT microbiota resembled the working mechanism suggested for some growth-promoting antibiotics, namely the suppression of Gram-positive commensals rather than potentially pathogenic Gram-negative organisms. Hojberg *et al.* (2005) suggested that reduced fermentation of digestible nutrients in the proximal part of the GIT might render more energy available for the host animal and contribute to the growth-promoting effect of high dietary ZnO doses. In contrast, dietary CuSO<sub>4</sub> inhibited the coliforms and thus potential pathogens as well, but overall the observed effect of CuSO<sub>4</sub> was limited compared to that of ZnO.

Despite the ambiguity related to the exact mechanism(s) of action of ZnO to elicit its beneficial effects, it is likely that it will continue to be studied because it is a cost-effective nutritional tool that appears to function well in situations where 'gut health' might be compromised, such as after weaning. Unraveling the mode(s) of action of this chemical compound could provide a means for the rational development of alternatives to antibiotic feed additives.

### Dietary Associations with Enteric Diseases

The study of a specific bacterial infection offers a means of assessing the usefulness of nutritional strategies on the survival of that particular pathogen in the GIT and its ability to affect production, morbidity and mortality. There are a number of well-known enteric bacterial diseases that occur throughout Australasia and indeed the world, and each is relatively unique in that it generally occurs at different phases of pig growth and (or) in different regions of the GIT. There is a relative paucity of studies examining the effects of a specific pathogen on the overall balance and diversity of the GIT microbiota, and how alterations to a community, for example by feeding a particular substrate, can influence pathogenesis. Even where the addition of a dietary component/nutrient/additive is known to stimulate proliferation of specific groups of resident bacteria, little is really known about the way in which these bacteria interact with pathogenic species of bacteria. This lack of information makes it difficult to predict how a given dietary component could be used to indirectly influence a given enteric pathogen (see review by Hampson *et al.*, 2001).

The following section deals with the influences of nutrition/nutritional management on two important diseases, namely swine dysentery (SD; caused by *Brachyspira hyodysenteriae*) and salmonellosis (*Salmonella* spp). We have intentionally highlighted these two diseases because they are pathogenic in different parts of the GIT (SD in the large intestine, salmonellosis mainly in the terminal small intestine/large intestine), highlighting possibly different mode(s) of action, and the approach to their control is quite marked.

### Swine dysentery

Swine dysentery (SD) is a mucohemorrhagic colitis occurring mainly in grower pigs that involves the caecum, colon and rectum, and is caused by the anaerobic spirochaete *Brachyspira hyodysenteriae* (Hampson *et al.*, 2006). Clinical manifestations vary greatly, and include both mild and sub-clinical disease. In typical cases, infected pigs initially show a slight depression and reduced feed intake, they develop diarrhoea, and this can progress to consist of mucus plugs, fibrin, epithelial cell casts, and flecks of fresh blood. Affected animals have faecal staining of the hindquarters, become dehydrated and appear gaunt, with a tucked-in abdomen and an arched back. If left untreated, around 10% of affected pigs can die within five days of first showing clinical signs (Hampson and Trott, 1995).

The exact pathogenesis of SD is not clear, however it is apparent that the disease does not always express itself clinically in pig herds despite the presence of the bacterium (Hampson *et al.*, 2006). Numerous factors are implicated in the aetiology of SD (see reviews by Hampson and Trott, 1995; Harris *et al.*, 1999; Pluske *et al.*, 2002; Hampson *et al.*, 2006), including nutrition, and this fact resulted in a considerable body of work being conducted at Murdoch University in the 1990s examining the relationships between cereal type, processing, enzyme addition, the microbiota and the clinical expression of SD following experimental challenge. These data have been reported previously (see Pluske *et al.*, 2002; Hampson *et al.*, 2006).

Briefly, our data showed that feeding a diet low in both soluble non-starch polysaccharides (NSP) and resistant starch (RS) generally afforded protection against *B. hyodysenteriae*. However, the manner in which the grains have been processed also appears to be important, especially with cereals inherently low in NSP (< 1 g/100 g soluble NSP). Our data suggested that a reduction in RS levels (e.g. via extrusion, steam flaking) would only prove effective against SD if the grain in question has an inherently low NSP level to begin with (Durmic *et al.*, 2002). In this regard, Durmic *et al.* (2002) reported with multiple regression analyses across numerous studies that colonisation by spirochaetes was highly related to the dietary concentrations of soluble NSP, while development of SD was similarly influenced by the RS content of the diet.

Other researchers, however, have failed to confirm our findings, and this has caused some doubt as to the ability to ameliorate against SD using diet. Leser *et al.* (2000) did not detect the same synergistic bacteria in pigs infected with

*B. hyodysenteriae*, although they did report changes in bacterial populations when pigs were fed either a cooked rice diet or a fermented liquid feed following infection with the causative agent. Kirkwood *et al.* (2000), Leser *et al.* (2000) and Lindecrona *et al.* (2003) failed to duplicate our previous results showing that feeding pigs a diet lower in soluble NSP and RS ameliorated the clinical expression of SD. Subtle diet differences existed between the studies, and these could have accounted for such discrepancies. For example, Kirkwood *et al.* (2000) and Lindecrona *et al.* (2003) fed parboiled rice, which has a higher RS content, whereas we fed cooked (autoclaved) medium-grain rice of a lower RS content. In turn, the amount of RS reaching the lower bowel could have been responsible for conditions that favoured/prevented colonisation of the spirochaete (Kim *et al.*, 2006). German researchers (Baumann and Bilkei, 2002) reported that high levels of highly fermentable fiber (9.6% highly fermentable neutral detergent fiber) may increase health and performance of pigs experimentally infected with *B. hyodysenteriae* compared to pigs fed a diet containing 6.1% low fermentable neutral detergent fiber. Moreover, a recent study by Thomsen *et al.* (2007) examined the effects of two diets based on triticale and barley and supplemented with either rape seed cake or dried chicory root and sweet lupins. The study showed that diets supplemented with highly fermentable carbohydrates from dried chicory roots and sweet lupins can protect pigs against developing SD. Finally, Piao *et al.* (2007) concluded that lower concentrations of iso-valerate and iso-butyrate in digesta, associated with feeding non animal-protein-based diets and therefore in contrast to our general proposition that feeding animal proteins is protective against SD, was “likely associated with development of pathogenic spirochete infection”. However, Piao *et al.* (2007) made this conclusion based on “3 samples collected from one farm”, and their description of the diets used was vague.

Species differences within a particular pathogen might also influence the success, or otherwise, of a nutritional treatment. For example, Lindecrona *et al.* (2003) found no protective effect of feeding a (parboiled) cooked rice-based diet on the development of SD in 18-30 kg pigs, but Lindecrona *et al.* (2004) reported a protective effect of feeding cooked rice on the development of *Brachyspira pilosicoli* (the agent of porcine intestinal spirochaetosis - PIS - or porcine colonic spirochaetosis) in pigs. In the same studies, Lindecrona *et al.* (2003) found a protective effect of fermented liquid feed on the development of SD but with *Brachyspira pilosicoli*, there was no significant effect of fermented liquid feed (Lindecrona *et al.*, 2004).

Less attention has been given to general microbiological changes that occur in association with a specific disease. In the case of SD, other bacterial species such as *Fusobacterium*, *Clostridium* and *Bacteroides* need to be present in the lower GI tract for the disease to occur (see review by Pluske *et al.*, 2002). Durmic *et al.* (1998) showed diet-associated differences in the genera and species present in the large intestine of pigs experimentally infected with SD, with changes in bacterial populations consistent with those that occur in the natural disease. Leser *et al.* (2000), using 16S ribosomal DNA sequence analysis, did not detect the same synergistic bacteria in pigs infected with *B. hyodysenteriae*, although they did report changes in bacterial populations when pigs were fed either a cooked rice diet or a fermented liquid feed, following infection with the spirochaete and subsequent destabilisation of the colonic bacterial community.

Results such as these make generalisations about the effect of nutrition on SD difficult to make. Unfortunately, such conclusions are not made any clearer when researchers speculate beyond the confines of a particular experiment. For instance, Hogberg *et al.* (2004) conducted a study using pigs fitted with post-valve T-caecum (PVTC) cannulae on the effects of diets differing in NSP content on the diversity of GIT coliforms, using terminal restriction fragment length polymorphism (T-RFLP). These authors reported that the ratio of soluble and insoluble NSP influenced the coliform diversity in the large intestine, which is plausible, but then drew inferences between their data and the link to SD. They commented that: “Previous findings suggest that it is the soluble fraction of NSP that predisposes pigs to swine dysentery. Diets based on cooked rice and animal protein reduced the clinical expression of *Brachyspira hyodysenteriae*, presumably by limiting the amount of fermentable substrates entering the large intestine (Pluske *et al.* 2002). However, we found no difference in coliform diversity between the Low NSP diets on day 17, possibly indicating a stabilisation over time. Further, we found no difference in the mean coliform diversity in connection to varying total NSP level measured in the rectal samples. Indeed, other researchers have failed to prevent development of swine dysentery with low fibre diets based on cooked rice and animal protein (Kirkwood *et al.* 2000, Lindecrona *et al.* 2003).”

Such data need to be questioned. First, the use of pigs fitted with a PVTC cannula, in which a significant proportion of the caecum is removed, casts doubt on hindgut function, particularly with a disease such as SD that can develop in the caecum. Second, the genus *Brachyspira* does not belong to the family *Enterobacteriaceae*, which are commonly called the ‘coliforms’. To then draw conclusions linking coliform ‘diversity’ to SD is clearly wrong, and these authors should be challenged. Nevertheless, Hogberg *et al.* (2004) did say that the balance of the intestinal flora, as well as pig genotype and microbial environment, are factors of significance for the development of SD, although this has been said previously (e.g., Hampson and Trott, 1995).

What is the way forward with the nutritional effects on SD? It is evident that dietary fibre has an impact on proliferation of the disease, and more precise delineation of this in any diet-related investigations into SD is required. In addition, an area that has received very little attention is the influence of protein entering the large intestine on SD

(although see Jacobsen *et al.*, 2004), and possible interactions with dietary fibre. A comparison of different pathogenic strains species found in field cases coupled to techniques such as T-RFLP and RT-PCR that can identify and enumerate other bacteria might also signal new directions.

### Salmonellosis

The major route for transmission of *Salmonella* in pigs is the oral route (Fedorka-Cray *et al.*, 2000) after which the basic virulence strategy common to *Salmonella* is to invade the intestinal mucosa and multiply in the gut-associated lymphoid tissues. From the infected tissue the bacteria are drained to the regional lymph nodes where the host defence mechanism can prevent further spread, in which case the infection remains localised to the gut and often manifests itself as acute enterocolitis. If the macrophages in the draining lymph nodes are unable to limit spread, *Salmonella* can cause systemic disease (Baumler *et al.*, 2000).

Therefore clinical porcine salmonellosis can be separated into two groups with distinctly different symptoms. The first, most severe, form is associated with septicaemia and involves the host-adapted *S. choleraesuis* that is common in the U.S.A but absent in many European countries. The second disease is associated with enterocolitis often connected with *S. typhimurium*, which may be a more important pathogen in a number of countries (Fedorka-Cray *et al.*, 2000; Griffith *et al.*, 2006). A wide variety of other serovars have been isolated from pigs and although they occasionally may cause disease, in general, infected pigs may remain healthy carriers (Fedorka-Cray *et al.*, 2000). In many countries, the problem of *Salmonella* infection in pigs is consequently mainly a question of food safety as sub-clinical *Salmonella enterica* infections in pig herds are recognised as important sources of human salmonellosis and hence a potential threat to human health through food-borne disease outbreaks.

Numerous epidemiological reports have shown that the physical characteristics of feed influence the susceptibility of pigs to *Salmonella*, as the *Salmonella* prevalence was higher in pigs fed heat-treated pelleted feed compared with feeding meal or liquid feed (Dahl, 1997; Wingstrand *et al.*, 1997; Stege *et al.*, 2000). For example, Wong *et al.* (2004) conducted a survey using 359 finishing-pig herds in Germany, Denmark, Greece, The Netherlands and Sweden, between 1996 and 1998, to find herd factors associated with pigs testing seropositive for *Salmonella*. These data showed that pigs fed non-pelleted feed (dry or wet) had a 2- and 2.5-times lower odds of seropositivity compared to pigs fed pelleted feed, and that pigs given whey (to drink or as the liquid part of the diet) had 2.6-times lower odds to test seropositive than pigs not receiving whey.

To understand this phenomenon further, Mikkelsen *et al.* (2004) and Hedemann *et al.* (2005) conducted a 2x2 factorial experiment studying the effects of feed grinding (fine and coarse) and feed processing (pelleted and non-pelleted) on physicochemical properties, microbial populations, morphological characteristics in the small intestine, caecum and colon and *in vitro* survival and adhesion of *Salmonella enterica* serovar *Typhimurium* DT12 in the GIT of pigs. These authors demonstrated that feeding a coarsely ground meal feed to pigs changes the physicochemical and microbial properties of the stomach contents, predominately an increase in undissociated lactic acid concentrations, which caused a higher death rate of *S. enterica* serovar *Typhimurium* DT12 and consequently decreased this bacterium's survival during its passage through the stomach. Knarreborg *et al.* (2002) previously reported that high levels of undissociated lactic acid inhibited the growth of enterobacteria. In this way the stomach acts as a barrier preventing harmful bacteria from entering and proliferating in the lower part of the gastrointestinal tract. In addition the adhesion was 60% less to the ileal tissue of pigs fed the non-pelleted diets than to those fed pelleted diets. Further, pigs fed pelleted diets secreted mucins capable of binding *Salmonella enterica* serovar *Typhimurium* DT12 and thereby allowing for colonization.

Regarding dry feed, several Danish studies have confirmed that offering meal feed to growing-finishing pigs reduces the *Salmonella* prevalence compared with feeding heat-treated pelleted diets (Hansen, 2004; Jørgensen *et al.*, 1999). However in the same studies it was repeatedly shown that offering a dry meal diet to growing-finishing pigs reduces the performance of the pigs. This poorer performance can be explained by differences in particle size distribution and hence utilisation of the available energy and nutrients, but again it underlines that improving GIT health is often associated with increased cost of production.

Fermented liquid feeding is another means of manipulating the GI microbiota, in both the fermentation tank and the GI tract (e.g., Mikkelsen and Jensen, 2000). Fermented liquid feed (FLF) is characterised by high numbers of lactic acid bacteria, high numbers of yeast, a low pH (< 4.0), and a high concentration of lactic acid (132-244 mM) (only the undissociated form of lactic acid is bactericidal/bacteriostatic), and typically results in reduced numbers of coliform bacteria in the feed, provided that fermentation conditions are correct. Van Winsen *et al.* (2001) investigated the effects of fermented (liquid) feed on bacterial populations along the GI tract, and reported significant negative correlation in pigs fed fermented feed between the concentration of disassociated lactic acid and *Enterobacteriaceae* numbers in the stomach. Van Winsen *et al.* (2001) concluded however that the direct influence of lactobacilli on *Enterobacteriaceae* numbers could not be demonstrated.

### Can nutrition of the suckling piglet influence gut health and lifetime performance?

To conclude this part of the symposium, we wish to broach a subject that questions the timing and nature of a dietary intervention to effect changes in 'gut health' that, in turn, might influence whole-of-life performance. Pluske *et al.* (2005) reviewed the influence of nutrition of the young pig in relation to lifetime performance. In this review, an experiment conducted by Hugh Payne was described where the effects of three pre-weaning nutritional treatments on lactation and lifetime performance were assessed. The three treatments during lactation were: (i) no creep feed, (ii) litters offered a commercial, pelleted creep feed, and (iii) litters offered a mixture of sow feed, fresh straw and soil and organic matter (e.g., faeces, stubble) in a ratio of approximately 5:1:25 (hereafter referred to as the 'outdoor mix'). The third treatment was used to simulate materials that outdoor born and reared piglets might encounter and consume under commercial outdoor conditions. After weaning, piglets were all fed and housed indoors under identical conditions until slaughter at approximately 105 kg. The diets used in treatments (ii) and (iii), and the diet fed after weaning, did not contain any growth promoting antibiotics or pharmacological levels of Zn or Cu.

A summary of the data are presented in Pluske *et al.* (2005) and Table 2, and shows that feeding the 'outdoor mix' during lactation appeared to have a stimulatory effect on both hot carcass weight and dressing (killing out) percentage, after statistical correction for birth weight, sex, P2 backfat depth and final live weight, at slaughter. These data infer that some element(s) associated with young pigs being exposed to an 'outdoor' environment has/have a beneficial effect on carcass conformation 19-20 weeks later.

What might precipitate such a marked effect? A subsequent study using objective PCR-DGGE methodology to assess the diversity of the microbiota in the large intestine of piglets from indoor and outdoor housing systems revealed differences in the gastrointestinal ecology of pigs born outdoors and raised on deep litter (Pluske *et al.*, 2007). These data suggest that changes caused to the GIT microbiota, and hence possibly immune function, by nutrition early in life have whole-of-life effects on growth and production (Payne *et al.*, 2003).

**Table 2. Effects of offering no creep food, a commercial creep feed, or an outdoor mix on the performance and carcass characteristics of pigs from birth to slaughter (from Pluske *et al.*, 2005)**

	Treatment in lactation			LSD <sup>B</sup> (5% level)
	No creep feed	Commercial creep feed	Outdoor mix <sup>A</sup>	
<b>Body weight (kg)</b>				
Weaning (28 d)	9.1	8.7	8.9	0.9
7 d post weaning	9.7	9.8	10.0	0.8
28 d post weaning	18.7	19.3	19.9	1.6
Final weight	107.1	106.2	107.4	1.4
<b>Daily gain (g)</b>				
Birth-weaning	266	256	260	31
Weaning -7 d after weaning	60	123	131	63
Weaning - 28 d after weaning	344	366	378	63
Birth - 28 d after weaning	307	316	325	23
Birth - finisher	677	677	693	23
Weight/age <sup>C</sup> , g/d	686	687	701	24
HCW <sup>D</sup> (Trim 13) (kg)	70.4	70.3	71.7	1.27
P2, mm	12.4	12.7	13.2	1.78
Dressing, %	65.7	65.9	66.8	1.08

<sup>A</sup>Outdoor mix consisted of creep feed, straw, and soil + organic matter.

<sup>B</sup>LSD: least significant difference.

<sup>C</sup>Weight/age = (Final weight/final age)\*1000.

<sup>D</sup>HCW: hot carcass weight.

### Conclusions

'Gut health' is an often contentious and puzzling subject area and one that continues to attract attention worldwide. Important progress has been made in a relatively short period of time in relation to our understanding in this field at the GIT level, with reference to the mechanisms underpinning the physiology, microbiota and localised immune system. Some concepts have emerged, for example, the notion of stimulating/nullifying specific groups of bacteria in the GIT to modify the GIT environment, however it will not be until the system is viewed and analyzed

holistically that major advances will be made. In the post-weaning period, where most attention has been focused, the compromised state of the young pig makes it an ideal candidate for the range of dietary products that might influence 'gut health'. However, pigs in later stages of growth also suffer from diseases and conditions that can dramatically influence production and survival, so 'gut health' needs to be viewed from the whole-of-life perspective. In this sense, and as discussed in this symposium previously, interventions through the sow (gestation, lactation, gestation plus lactation) could be more appropriate in some cases to modify GIT health in the offspring, via colostrum and milk, than via the newly-weaned pig, for example. There is already a large body of research, and this continues to grow, in regard to the various interventions that might influence GIT health. Some of these data are published while other data remains unpublished. Regardless, the issue of variability in responses seen to interventions needs to be considered in any recommendations that might be made.

## Designing a new growth promoter in the 21st century

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Antibiotic growth promoters (AGP) have been supplemented to pig diets at sub-therapeutic levels since about the early 1950s after veterinarians realized the value of antibiotics when penicillin left over from World War II was used for intramammary treatment of bovine mastitis (Gustafson and Bowen, 1997). In recent years, societal concerns about drug-resistant microbes, fast-food chain and grocery store company purchasing requirements, and government regulations to promote food safety are increasingly restricting the use of AGPs. However, the search for alternatives to or replacements for AGPs is hampered by a lack of knowledge about the modes of action by which antibiotics alter intestinal microbial populations and/or cause immunomodulation to enhance growth performance of pigs (Niewold, 2007). This paper addresses some of the key steps behind selection of a new growth promoter, and identification of the key personnel involved in the development of a new product. A case study for a novel non-antibiotic anti-inflammatory growth promoter is presented to highlight the research tools establishing product efficacy. The identification of key biomarkers for inflammatory responses (TLR-4, cytokines, iNOS etc) has allowed the development of cell culture assays for the rapid screening of compounds of interest. The application of a soluble mannan product extracted from *S. cerevisiae* to an immortal chicken cell line (MQ-NCSU) demonstrated that an anti-inflammatory response was achieved through the down-regulation of NF  $\kappa$ B, IL-6-like activity and a concomitant reduction in nitric oxide being produced. In addition, the mannan preparation appeared to bind to TLR-4 thereby preventing the interaction with LPS when challenged as demonstrated by Western Blot analysis. The validation of the cell culture assay by running a broiler production study proved that the dietary supplementation of MRF resulted in a significant body weight improvement.

### Antimicrobial resistance problem in human medicine

The emergence of antimicrobial resistance has led to increasing concerns about the use of antimicrobial products in human medicine, veterinary medicine, animal agriculture, and horticulture. There is a general agreement that the use of antibiotics in animal production leads to increases in the incidence of resistant bacteria in the intestines of animals exposed to antibiotics (World Health Organization, 1997; Witte, 1998), and there is evidence that antibiotic resistance genes can be and are transmitted from animal to human microbiota (Greko, 2001).

The use of noursesthrin, an aminoglycoside, in East Germany gave strong evidence for a 'cause and effect' relationship between antibiotic use in agriculture and increasing resistance in the commensal microflora of farm animals (Witte 1998). Within two years of the introduction of this antimicrobial, resistance to noursesthrin was discovered and the incidence of resistance increased until its use was discontinued. Noursesthrin had not been used in human medicine and resistance appeared to be causally related to its use in agriculture.

Other studies in Europe demonstrated a strong association between the use of avoparcin as a growth promoter for animals and the occurrence of vancomycin-resistant enterococci in animals and food (Bates *et al.*, 1994; Klare *et al.*, 1995; Bager *et al.*, 1997; Wegener *et al.*, 1998). Vancomycin was considered to be a last resort antibiotic for cases of multiple resistant strains of *Enterococcus faecalis*, a major cause of hospital-acquired infections (Levy, 1998). There is increasing evidence that bacteria can develop resistance to antibiotics related in chemical structure to those to which they have been exposed. Cross-resistance to therapeutic drugs exists in most cases where the growth promoter belongs to the same class of antibiotics as the therapeutic drug (Wegner *et al.*, 1998). For examples of closely related AGPs and human medicines, see Table 3.

**Table 3. Associated growth promoters and human medicines (Wegener *et al.*, 1998)**

Antibiotic growth promoter	Human medicine
Avilamycin	Ziracin
Avoparcin	Vancomycin
Tylosin phosphate	Erythromycin
Virginiamycin	Synercid

The development of antibiotic resistant strains has been associated with continuous use of sub-therapeutic quantities of antibiotics but development of viable antibiotic resistant strains is a complex process (Salyers, 1999). When antibiotic use is transient, bacteria resistant to the antibiotics, by survival, are selected initially, but these may then be unable to compete with susceptible bacteria when the antibiotic selective pressure is removed. However, if antibiotic selection is continuous over long periods of time, bacteria have the chance to accumulate compensatory mutations that allow them to retain their resistance phenotype without losing fitness to thrive in their ecosystem (Morrell, 1997; Salyers, 1999). Resistance to an antibiotic may sometimes improve a pathogenic bacterium's competitiveness in the gut and therefore permit it also to dominate a foreign ecosystem (Salyers, 1999). Once a drug-resistant mutant pathogen has emerged in a bacterial population, the resistance can be transferred to other bacteria by the mechanisms of transformation, transduction, or conjugation (Salyers, 1999). The indiscriminate use of antibiotics in animal production appears to have exacerbated some of the disease conditions that their development was intended to prevent and to have allowed the resistant strains to reach humans. Traces of antibiotics and other medicines are also showing up in rivers and municipal water supplies, and experts believe these chemicals may harm the environment and human health (Salisbury *et al.*, 2002).

### **Bans on AGPs in animal production**

In 1986, Sweden became the first country to ban antibiotics for use in animal growth promotion. Avoparcin was banned in Denmark in 1995 and in the European Union in 1997. In 1998, Denmark banned virginiamycin, and Danish cattle, poultry, and finisher pig producers voluntarily stopped use of all AGPs. In 1999, the European Union Commission banned tylosin, spiramycin, bacitracin and virginiamycin because they belonged to classes of antimicrobials also used in human medicine and olaquinox and carbadox which were considered to have unacceptable occupational toxicity risks. In December 1999, the Danish swine producers voluntarily stopped use of all remaining AGPs in pigs under 35 kg body weight, and Denmark has restricted the use of antimicrobials to therapeutic use, by prescription only, since January 1, 2000 (Dibner and Richards, 2005). In 2006, the European Union Commission removed from the market the last four antimicrobial products used in animal production (avilamycin, flavophospholipol (Flavomycin), monensin sodium, and salinomycin sodium) (Anonymous, 2006).

The European Union ban on antibiotics greatly affected Brazilian animal feed additive use because of exports of poultry and livestock products. Brazilian producers must follow strict regulations governing animal welfare and consumer safety (antibiotic ban) in order to ship products to the European Union. As far as the situation in Brazil is concerned, the ban on antibiotics is unlikely to be applied to meat produced for internal consumption any time in the near future (Frost and Sullivan, 2007).

In the United States, bans against the use of nitrofurans, chloramphenicol, and ampicillin in animal feeds have been in place for many years (Johnston *et al.*, 2007). The U.S. Food and Drug Administration proposed banning fluoroquinolones (useful against pathogenic *E. coli*) in poultry in 2000, but the drug manufacturer fought the ban until it was finally enacted in September 2005 (Elmin, 2006). Japan has banned some antibiotics, others are still used in animal production, and some production is voluntarily antibiotic free. Ionophore coccidiostats remain permitted.

Understanding how these antibiotics work as growth promoters provides a guideline for investigators to search for future alternatives and/or replacements to AGPs. An "alternative" is described in the dictionary as "a necessary or remaining course or choice" whereas a "replacement" is "a substitute or equivalent in the place of" (Webster's New Universal Unabridged Dictionary, 1992).

### **Microflora-management related mechanisms of AGPs**

In order to find substitutes for AGPs in animal production, it is important to consider our knowledge of the ways antibiotics may enhance growth and reduce pathogen counts (especially *Salmonella* species, *Campylobacter jejuni*, *Escherichia coli*, *Clostridium perfringens*, and enterococci). At least four major mechanisms have been proposed to explain antibiotic growth promotion in animals (Gaskins *et al.*, 2002; Dibner and Richards, 2005; Page, 2006) and in which intestinal microbiota is the target: 1) AGPs inhibit endemic subclinical infection and thereby reduce the metabolic costs of the innate immune system, 2) AGPs reduce concentrations of growth-depressing metabolites such as ammonia and bile degradation production generated by microbes, 3) AGPs reduce microbial consumption of nutrients in digesta, and 4) AGPs enhance the uptake and use of nutrients because the intestinal wall in AGP-fed animals is thinner.

### **Host animal anti-inflammatory effect of AGPs**

A proposed fifth mechanism deals directly with the host animal as the target. The mechanism is based on the observation that AGPs have a nonantibiotic anti-inflammatory effect which improves growth performance (Niewold,



2007). The rationale behind this hypothesized mechanism is that: 1) AGPs have a similar effect on various production animals such as poultry and pigs which differ considerably in the composition of intestinal microbiota (and further change during phases of feeding), 2) AGPs differ widely in chemical classes and microbial spectra of activity (Gram-positive or Gram-negative), and 3) not all antibiotics have growth-promoting activity whereas they should be expected to according to the microflora-management theory. As cited by Page (2006), earlier authors concluded “there appears to be no obvious explanation for the great variation in growth-promoting activity between the different classes of antimicrobial substances studied”.

According to Niewold (2007), what a variety of antibiotics have in common is that they accumulate in inflammatory cells (van den Broek, 1989; Labro, 1998, 2000). Most accumulated antibiotics enhance the killing of bacteria and inhibit parts of the innate immune system. Inhibitory effects of antimicrobial compounds on phagocytic cells (macrophages and polymorphonucleocytes) have been well documented (van den Broek, 1989; Schoevers *et al.*, 1999; Labro, 1998, 2000). Antibiotic compounds can be divided into three groups: 1) non-accumulating, 2) accumulating without inhibition of function, and 3) accumulating with inhibition of function. Although some antibiotics have been claimed to be nonabsorbable by the intestinal tract, this may be true in the healthy intestine, but episodes of enhanced intestinal permeability are not uncommon in production animals (Niewold *et al.*, 2000). One of the consequences of intestinal inflammation is increased macromolecular intestinal permeability (MacDonald and Monteleone, 2005) which certainly would facilitate local penetration and accumulation of low molecular weight antibiotics.

Phagocytic cells can accumulate antibiotics, in some cases two- to 100-fold the ambient (extracellular or intestinal lumen) concentration (Labro 1998, 2000 reported cyclines, 2x; streptogramin peptide antibiotic, 40x; macrolides, 10x-100x accumulations). The relevant effect of this accumulation of an antibiotic(s) in phagocytic inflammatory cells would be attenuation of the inflammatory response. Consequently, the levels of proinflammatory cytokines would be lower and therefore the catabolic stimulus would be lower than in untreated animals. This is consistent with a growth-permitting rather than a growth-promoting effect. Concerning bacitracin, van den Broek (1989) described an inhibitory effect of bacitracin on phagocytosis. Alloza and Vandebroek (2005) suggested an anti-inflammatory effect whereas Higuchi *et al.* (2004) showed a possible proinflammatory role for bacitracin.

The intestines have been described as an organ in a state of more or less constant controlled inflammation (Biancone *et al.*, 2002). Antibiotics have been reported to inhibit one or more of several different functions of inflammatory cells, chemotaxis (movement of an organism in response to a chemical concentration gradient), the production of reactive oxygen species, and proinflammatory cytokine production, which is the most important effect on growth. Upon release of these cytokines, an acute phase response occurs, causing an overall catabolic effect (Niewold, 2007). The response includes a shift in hepatic protein production toward acute phase protein synthesis, catabolism of muscle tissue, and a loss of appetite (Gruys *et al.*, 2006). The acute phase response incurs the greatest physiological expenses (Humphrey and Klasing, 2003), and because of the magnitude of this effect, one would expect measurable effects from inhibitors of cytokine production such as antibiotics.

Based on the foregoing discussion, one would expect the largest effect of AGPs in less optimal conditions with the most inflammatory responses (Niewold, 2007). Use of macrolide antibiotics in human medicine for pulmonary inflammatory conditions (Hoyt and Robbins, 2001), to take advantage of “the non-antibiotic effect of antimicrobial compounds” (Labro, 2000), suggests that AGPs may act similarly in the intestinal mucosal system to help manage inflammation. Niewold (2007) suggested that the different intestinal microbial populations when using AGPs may be a consequence of altered immune status rather than of a direct effect on the *microbiota*.

Intestinal inflammation usually causes an accumulation of inflammatory cells in the mucosa leading to a thicker intestinal wall. The thinner intestinal wall observed when supplementing an AGP is consistent with reduced tissue inflammation, which is associated with a reduced influx and accumulation within inflammatory cells (Larsson *et al.*, 2006).

### Alternatives to AGPs for growth promotion

A wide variety of products have been used to replace AGPs in animal feeds, and these can be grouped into categories according to their physical composition and activity. Among these products are bacteriophages, betaine, bioactive peptides, botanicals or herbs, copper sulfate and zinc oxide, dietary ingredients, direct-fed microbials (probiotics), enzymes, essential oils and spice extracts, humic substances, 25-hydroxy-vitamin D3 (25-OH-cholecalciferol), ionophore coccidiostats, mannan oligosaccharide (yeast cell wall; certain sugars), organic acids (acidifiers), plasma protein and specific antibody egg yolk, prebiotics (such as inulin), sodium bicarbonate, and vaccines. As evidenced by the list of potential alternatives in Table 4, very few of the candidates can be considered to be replacements, especially if one considers the anti-inflammatory mechanism of action.

**Table 4. Some alternatives to sub-therapeutic antibiotics in animal production with brief comments regarding their use for pigs and poultry**

Category or product	Comments based on scientific research
Bacteriophages	Bacteriophages (phages) are viruses that infect bacteria. These agents are widespread in nature and have evolved into a wide variety of types to target specific microbes (Huff <i>et al.</i> , 2003)
β-D-glucans	Derived from a variety of fungal and algae sources. These compounds have powerful immunostimulatory effects. They have been used to promote performance in pigs but questions still remain as to whether they can be classified as growth promoters after Klasing showed the cost of stimulating the immune system. It appears that they have a specific application in herds which have immunosuppressive problems (Zekovic <i>et al.</i> , 2005).
Betaine	Dry or liquid application; osmotic function supports intestinal integrity to help resist invasion of pathogens; benefits in heat stress; methyl donor spares some methionine and replaces some choline; enhanced energy utilization in pigs allows moderate rather than high energy diet formulations (Eklund <i>et al.</i> , 2006).
Bioactive peptides	Certain naturally occurring peptides can provide early protection for animals by stimulating innate immunity, directly killing pathogens, and increasing the magnitude of the immune response following vaccination (Anonymous, 2007).
Botanicals and herbs	Plant parts such as roots, bark, aerial portion, leaves, whole plants, or extracts from “nature’s green pharmacy”; sometimes difficult to get regulatory approval, especially for blends, to use in animal feeds (even though used in human dietary supplements); may be bacteriocidal or improve immune response; botanical is from any plant whereas herb is from a flowering, non-woody stemmed plant (Ilsley <i>et al.</i> , 2005).
Copper sulfate; zinc oxide	Copper sulfate providing 200 to 250 ppm copper for pigs or about 150 ppm for meat birds improves growth; zinc oxide at 2,000 to 3,000 ppm for 2 weeks after weaning pigs decreases scours, improves performance; tribasic copper chloride (TBCC) has higher copper bioavailability (estimated +20%) therefore requires lower levels of inclusion; organic copper or zinc supplements may have good bioavailability and less excretion (Broom <i>et al.</i> , 2006; Holberg, 2005; Mahan, 1990).
Direct-fed microbials (probiotics)	Competitive exclusion strains colonize and protect the intestinal mucosa with lactic and acetic acid, or antimicrobial substances such as reuterin; pass through lactic acid producing bacteria; <i>Bacillus</i> spores are pelletable, economical, and consume oxygen, creating more anaerobic condition favoring <i>Lactobacilli</i> ; improve gut health and performance (Schneitz <i>et al.</i> , 1998; Chow, 2002).
Enzymes	Phytases enhance digestibility of phosphorus, calcium, other minerals, and amino acids by unbinding them via action on phytate; xylanase and beta-glucanase release energy from cereal grains; protease and beta-mannanase act on protein and insoluble carbohydrate portion of soybean meal, respectively; and amylase hydrolyzes starch; enzymes facilitate digestibility and nutrient availability, reducing pathogens (Bedford, 1999).
Essential oils and spice extracts	Essential volatile oils (e.g. eugenol, thymol) from plants used either singly or in proprietary blends against specific pathogens or for specific purposes (e.g., to increase salivation and flow of enzyme containing fluid from the intestinal wall into lumen or to kill <i>Clostridia</i> ); spice extracts (e.g., cinnamaldehyde) have well known antimicrobial or flavoring properties; oils and extracts typically from commonly used, well known flavoring products (e.g. capicum) (Stein and Kil, 2006; Steiner, 2007).

Humic substances	Organic matter in soil, seams, or deposits (e.g. peat) including major constituents humic acid, fulvic acid, and humin; typically contain iron, manganese, copper, and as well; may lower ammonia emissions from pig manure by 3-18% depending on fulvic and humic acid contents; used at 0.5% level in diet; may benefit live performance (Goihl, 2006).
25-Hydroxy-vitamin D3	Performed so first step in vitamin D3 conversion in liver is not necessary, then to kidneys for conversion of 1,25-dihydroxycholecalciferol; enhances calcium, phosphorus, and magnesium absorption; about 1.7 times the potency of regular vitamin D3; may improve bone mineralization, growth rate, and feed conversion ratio; same improvements can not be achieved by adding more regular vitamin D3 (Calabotta, 1997).
Ionophore coccidiostats	Until 2006, salinomycin was approved for pigs in the E.U. Salinomycin is widely used in the U.S. for broiler chickens. Salinomycin has activity against <i>Clostridium perfringens</i> (Watkins <i>et al.</i> , 1997; Elwinger <i>et al.</i> , 1998; Martel <i>et al.</i> , 2004). Narasin, another ionophore coccidiostat, can decrease necrotic enteritis even in the presence of a coccidial challenge in broilers (Ross Tech, 2005).
Mannan oligosaccharide sugars	Mannose and certain other sugars can attach to pathogens with Type 1 fimbriae and block intestinal colonization; yeast cell wall (MOS) also has this ability; about 4 kg yeast produces 1 kg MOS product; stimulates nonspecific immune response; reported improvements in several animal species for gain, feed conversion ratio, and livability, and in sows increased birth and weaning weight, and pre-weaning growth rate and survivability of piglets (Pettigrew, 2000; Pettigrew <i>et al.</i> , 2004).
Metabolic modifiers	Repartitioning of nutrients within pigs is possible by use of a metabolic modifier such as a beta-agonist (e.g. ractopamine); typically increases protein and reduces fat deposition resulting in more lean gain (Lundeen, 2002)
Omega-3 fatty acids	Omega-3 fatty acid containing ingredients may help modulate the immune response such as macrophage activity (Branton <i>et al.</i> , 1997; Korver and Klasing, 1997; Kelley, 2004).
Organic acids	Certain organic acids or blends are effective against pathogens in feed or drinking water; coating allows delivery of acids to intestinal tract in lower, yet effective, concentrations than with uncoated acids; synergistic effect on potency with some acid blends; liquid methionine analogue has some activity; formic acid is not approved for use in the U.S. (Partenan and Mroz, 1999).
Plasma protein; egg yolk antibodies	May provide antibodies to specific pathogens; porcine plasma to pigs; IgY in yolk after injecting immunogen(s) into hens; heat or enzymatic denaturation of protein antibodies is sometimes possible; plasma increases feed intake and growth of young pigs, with greater effect in unsanitary conditions (Owusu-Asiedu <i>et al.</i> , 2003).
Prebiotics	Chicory root inulin or other fructo-oligosaccharides provide food for beneficial bacteria but not some of the pathogens; lactic acid bacteria proliferate (Mountzouris <i>et al.</i> , 2006)
Sodium bicarbonate	Enhances invasion of coccidia into intestine for rapid early immunity; potentiates ionophores that use sodium; with salt, effective for heat stress (Hooge, 2003)
Vaccines	Live, attenuated, or killed vaccines for specific diseases (e.g., poultry coccidiosis, salmonella, and E. coli vaccines); or sow vaccine against <i>Clostridium perfringens</i> around mid-gestation and repeat to protect pigs in cleaner facilities and get heavier pigs with better livability at weaning (Schultz, 1994; Fairbrother <i>et al.</i> , 2005).

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## Designing the next generation of growth promoters

### *Where does one start the process?*

What are the possible sources of different compounds that would be acceptable? The first and best place to start is by looking through the scientific literature, especially searching for those empirical observations for enhanced growth performance which may not have been deemed attractive whilst the AGP's were approved for use. Other potential sources of information include related fields of nutrition and the discipline of traditional fermented functional foods for maintenance of gut health has provided many clues for the animal health industry. It is important at this point to determine what kind of response variable is the customer looking for? Are they looking for growth performance in general or are they looking to disease resistance?

### *Market Survey*

The dreaded word – Marketing! Despite some of our prejudices towards this word the field of market surveys and understanding of the marketplace needs is vital to any business. I am not talking about communications or advertising in this instance. There is no point in a group of corporate executives sitting around a boardroom thinking up the next additive product to release on the market if it will not be adopted. Consumer acceptance is very difficult to determine without having a reliable information source that is watching trends and preparing market surveys. Despite knowing the importance of marketing, many companies are ill prepared at the outset of a given project and often wait until after the product is developed to pour money into the communications component. Much of this money and effort (and often heartache!) could be saved by putting effort in at the beginning.

There are various ways of obtaining information about the market trends and customers immediate and future needs. The first line of information should come from the companies' internal salesforce and external consultants who face the customer on a daily basis. The second line can come from attendance at regional and international meetings and garnering the information through observation of what others are doing. Finally, the third method is to contract a Marketing company to prepare a survey of the marketplace and obtain the information directly. There are a number of advantages and disadvantages to each of the methods. The first two methods have the advantage of being cheapest but suffer from the fact that every other company can obtain the same information as you. The third method will give you unique information as you prime the Marketing company with your specific request and that information gathered and analyzed is for you alone. However, there are two main disadvantages, the first is that any survey will be subject to the usual biases:

- a) What questions were asked?
- b) Were they open or closed questions?
- c) Was there bias in the interpretation?
- d) Was there sufficient control in the questioning?
- e) Was the survey conducted on a regional or international level?
- f) Were the numbers of interviewees sufficient to get a good overview of the needs of the marketplace?
- g) What is the robustness of the survey and reliability of the results?
- h) Had the company predetermined the responses because they had a specific product in mind? (This would be a terrible waste of time and effort).

The second disadvantage is the price tag that is normally associated with such surveys, particularly when you want large numbers for low error and also when you start crossing borders and performing international surveys. For these two reasons the area of market surveys are often bypassed.

From these interviews or through the connections that the company has to the market it is usually possible to identify some early adopters to help with development – this is ideal! Early adopters can give you rapid feedback as to whether or not you have a product worth moving forward with and also possibly a test site once the product has been made in trial batch quantities. Moreover, if the surveys are comprehensive in their design it may be possible to identify a key group of interviewees that represent the majority of the original set. These key players may then be followed up with a second questionnaire that is more specific to a product that may have been putatively identified by management following discussion of the original dataset. These questions would be used to:

- a) Estimate what monetary value the end user places on a growth promoting product.
  - a. Is the market ready to pay more than the original AGP?
  - b. Is there a premium to be gained if the product is natural or organic?
  - c. If a microbial product, will the market accept from a GMO?
- b) Determine the adoption rate in the market place given a set price.

- c) Determine the potential market penetration be within the product category.
- d) Identify the best geographical region to test market release the product.

### Building the development team

For any product development project it is the work of a number of different people working together on a common goal. In small companies it often falls on the same person to perform many of the following roles, or tasks are shared by a few or perhaps they outsource the expertise to other more specialized groups. In larger companies there would be a number of specialized personnel that would perform a specific task and these individuals would have multiple projects at different stages of development. A list of those specialists that would be identified (not comprehensive):

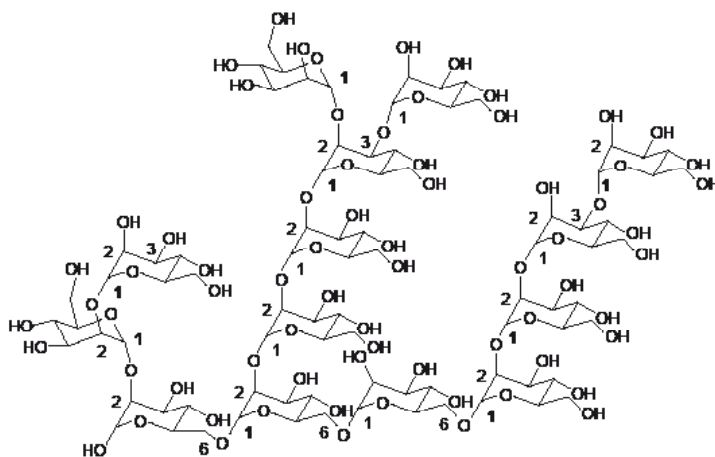
- Team Leader – Communications up and down the company, responsible for adhering to budget, coordination and timing of activities, report writing or collating. This would typically be an individual in Registration or Research Department.
- Research group – identifying who needs to be involved and when is the responsibility of the team leader and very much depends on the stage that the project is at. However, each of the members would be identified from the beginning and their input valued. For a project on developing a growth promoting product the following scientists would be included: animal scientists, analytical chemists, microbiologists and possibly an immunologist.
- Legal – Intellectual Property (IP) and Due Diligence are the hot buttons these days. It is not possible to get into this field without being aware of what's already got IP (to prevent being sued!) and also identify the possible risks (due diligence) of introducing a product into the marketplace. The questions being raised today about contaminants and possible toxic compounds make the issue of due diligence obligatory. What possible risks are involved if I introduce a product based on historical findings or risk analyses? The correct level of due diligence can prevent or at least demonstrate that the company minimized any possible introduction of risk to the animal feed/food chain.
- QA/QC – This is an often overlooked position on any R&D team but in fact possibly one of the most essential to include from the outset. The QA/QC team will take the product from the laboratory or developmental lab and introduce the product to the factory / processing plant through the outlining of Standard Operating Procedures (SOP) and implementing the various HACCP / ISO / cGMP procedures. In addition they need to have the time to set into place the testing procedures to ensure quality and conformity is being maintained throughout the production phase. Following manufacture the QC team needs to be able to assay for product activity and have established the limits of acceptability for quality parameters. These must conform to labeling and Product Specification sheets or parameters outlined in the submitted registration dossier, whichever is the strictest requirement.
- Production – The Production team representative will advise or provide feedback on issues pertaining to costs of production, availability of raw materials, availability of needed manufacturing equipment, scale up concerns and packaging issues. Many of these issues can be making or breaking of a product from the outset. Their expertise can counterbalance an often exuberant researcher that believes what happens in the lab can be produced in 100,000 tonne quantities! This is most definitely not the case.
- Registration department – does the product fit a known category or does the product need to undergo a special submission to be registered. Is the product already under the category Generally Recognized As Safe (GRAS) or will a self-affirmation GRAS application need to be submitted? What cost will this incur on the company? How many countries will require registration (nearly all these days)? Will one country accept the registration trials of another? The registration representative can save a lot of time and money in this regard.
- Senior management – If you don't have buy-in from the senior management board and the Chairman then the project will never be completed – fact of life. Senior management will be informed by the Team Leader and trusted to keep an eye on the budget.
- Consultant groups – This final group is generally introduced for specialist services that cannot be conducted in-house because of resource limitations. Wide range of specialties could include: registration, self-affirmation GRAS applications (for new product categories), toxicity testing, animal performance facilities (universities and research institutes) External marketing, Influencers (vets, research scientists, nutritionists, early adopters). These individuals often would not be privy to the entire project but just the component that involves their expertise.

## MRF – An example of an experimental product with anti-inflammatory properties

### What is MRF?

MRF is an experimental product under development, with its origins based on an existing product, Bio-Mos®, which has been through hundreds of studies. Many of these studies attempted to elucidate the mode of action of the product. The original product mode of action was based on the physical agglutination of enteric bacteria containing Type 1 fimbriae with the mannan component of Bio-Mos, thereby blocking their colonization to GI tract lectin receptors. Subsequent work demonstrated that there was a profound influence on the immune system when this product was fed to production animals.

MRF is a soluble extract from the yeast cell wall of *Saccharomyces cerevisiae* (Figure 5) whereby the mannoproteins are separated from the other cell wall components. The material is spray-dried by indirect heat at low temperature to prevent denaturation of the product. The product is not a single molecular structure but rather a collection of mannoproteins as described below. Yeast wall mannoproteins from *Saccharomyces cerevisiae* are highly glycosylated polypeptides, often 50-95% carbohydrate by weight, that form radially extending fibrillae at the outside of the cell wall (Lipke *et al.* 1998; Kapteyn *et al.* 1999). Many mannoproteins carry N-linked glycans with a core structure of Man10-14GlcNAc2-Asn structures very similar to mammalian high mannose N-glycan chains. “Outer chains” present on N-glycans consist of 50-200 additional  $\alpha$ -linked mannose units, with a long  $\alpha$ -1,6-linked backbone decorated with short  $\alpha$ -1,2 and  $\alpha$ -1,3-linked side chains. Until recently the identification of proteins in the cell wall has been hampered by the complex nature of the cell wall structure and its relative resistance to simple digestion and extraction. A novel method was developed to tag and identify cell surface proteins using a method based on treating intact cells with a membrane-impermeable biotinylation reagent that specifically reacts with free amino groups (Casanova *et al.* 1992). Using this method, the identity of approximately 20 cell wall-associated proteins was confirmed (Mrsa *et al.* 1997), although following a genomic approach greater than 40 have been predicted (Smits *et al.* 1999). Two distinct classes of cell wall proteins can be distinguished, GPI (glycosylphosphatidylinositol) proteins and PIR (proteins with internal repeats) proteins (Kapteyn *et al.* 1999). The GPI proteins are linked to other cell wall components through a remnant of their GPI anchor and  $\alpha$ 1,6-glucan cross-links the proteins to  $\alpha$ 1,3-glucan, an example is the  $\alpha$ -agglutinin protein. The PIR proteins are less well understood but in contrast to the GPI proteins, are not posttranslationally modified by addition of a GPI anchor, but are highly O-glycosylated (Mrsa *et al.* 1999). The mannoproteins determine the surface of the yeast cell and are responsible for the cells antigenic behavior. Their extraction in a functionally intact manner is relatively simple on a lab bench scale but proved to be rather difficult in the larger batch sizes.



**Figure 5.** Partial structure of the mannan from *Saccharomyces cerevisiae* showing the characteristic  $\alpha$ 1,2,  $\alpha$ 1,3 side chains attached to the  $\alpha$ 1,6 backbone of the polysaccharide.

### Mechanism of action of MRF

While the exact mechanisms have not been completely elucidated, significant evidence has been accumulated to propose MOS plays a multi-purpose role in immune modulation. Recently, there has been some evidence to suggest that MOS may play a role in suppression of the pro-inflammatory immune response. Ferket and co-workers (2002) induced an acute immune response in turkey poults by intraperitoneal injection of LPS from *Salmonella typhimurium* and measured fever response. Poults fed a diet containing MOS showed no fever response compared with the control (no additive) group, which experienced an increase of +0.4°C in body temperature. Greater control of the immune

response, particularly the fever response, can be beneficial to the host in terms of energy savings, maintaining feed intake and reducing stress, such as those responses invariably seen with the AGP's. Further studies demonstrated this to be a reproducible response when supplementing the diet with Bio-Mos®.

Following the experiments by Ferket *et al* (2002) it was decided to develop a product that would be specifically for suppression of the acute phase protein response (inflammatory response). The first step was to identify the active component or structure aligned to the mode of action. The extraction of this component from the yeast cell wall were performed on a lab scale and then a number of attempts were made to scale production to four tonnes (dry matter) batch size quantities in our facility in Serbia. In-house testing of final product, using broiler chicks, concentrated on estimating concentration for use in field as per dose titration experiments, growth performance parameters measured (ADG, ADFI, FCR), determination of possible toxicity (mortalities), quantification of select gut microbial populations, gut histology changes – morphometric and goblet cell measurements and quantification of the product in feed and faeces. A number of positive and negative controls were included alongside. The product was also tested in our salmonella challenge model for broiler chicks, to prevent colonization. With the exception of the salmonella exclusion study, where we could not achieve the same results as Bio-Mos® in terms of competitive exclusion, the results proved to the team that the product had a significant potential as a growth performance enhancer.

To investigate the immunomodulatory/anti-inflammatory properties of the MRF product it was important to work with an academic partner. Small lab scale samples were sent to be tested in the laboratory of Professor Frank Edens at North Carolina State University.

#### *Regulation of inflammatory responses and enhanced growth performance in broiler chickens.*

The following work to be described formed part of the graduate studies of Ms. Panthong Singabottra at NCSU under the direction of Prof. Frank Edens. The aims of Dr. Singabottra's research work included: 1) determining whether a mannan rich fraction (MRF), extracted from the cell wall of *Saccharomyces cerevisiae*, reduces inflammatory responses of chicken macrophage cell lines exposed to lipopolysaccharide (LPS); 2) identifying the mechanisms involved in the reduction of inflammatory responses of chicken macrophage cell lines exposed to LPS and MRF and 3) investigating whether MRF can reduce the inflammatory responses in vivo (Singabottra, 2006).

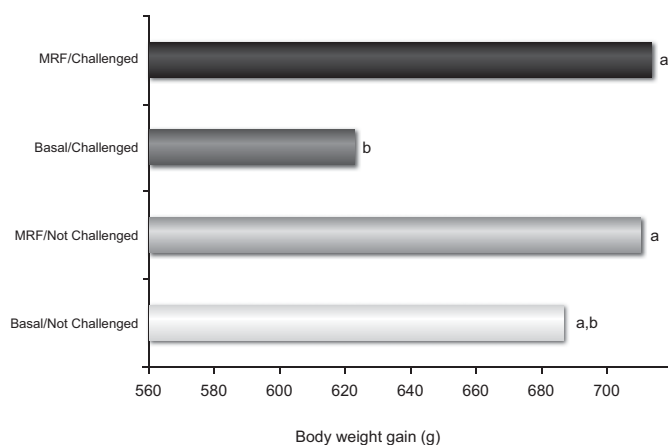
The first experiment investigated the effect MRF supplementation had on LPS-stimulated macrophage cell lines (MQ-NCSU and HTC). Nitrite production, IL-6 secretion, iNOS and IL-6 gene expression were quantified to determine the effect of MRF. By itself, MRF had no effect on nitrite or IL-6 secretion of either the MQ-NCSU or HTC cell lines. However, MRF significantly reduced the nitrite and IL-6 production of these LPS-stimulated two cell lines. Using RT-PCR to measure gene expression responses it was possible to determine that MRF down-regulated iNOS and IL-6 at the transcriptional level in LPS-stimulated macrophage cell lines. From these results it could not be determined at which point the MRF influenced control of the inflammatory cytokine production by macrophages, it may have a regulatory effect at the transcriptional level or by affecting TLR-4 events upstream to the LPS-signaling cascade as shown by less iNOS expression.

The second experiment was designed to investigate the effect MRF had on the signaling cascade via TLR-4 and NF- $\kappa$ B and elucidate the possible mechanism by which MRF reduces the inflammatory responses in chicken macrophage cell lines. MQ-NCSU cells were treated with medium only, LPS, MRF or LPS+MRF and cells were harvested at 0, 5 and 24 hours. The TLR-4 expression was analyzed by Western Blot analysis and NF- $\kappa$ B analyzed using an electrophoretic mobility shift assay (EMSA). TLR-4 expression of LPS-treated cells was significantly reduced in the presence of MRF at all time points in this experiment. This would suggest that the MRF is not having an effect on expression in this instance, at least at time = 0 h, because to down-regulate any protein expression, its transcription and translation processes have to be reversed. The immediate reduction in TLR-4 expression suggests an alternative interaction, possibly a binding mechanism, between the MRF and TLR-4. Thus, when the MRF is bound to the ligand binding site of TLR-4 it is no longer available or recognizable for LPS. Secondary confirmation comes from the fact that the monoclonal antibody used for the Western Blot assay could not bind to the TLR-4 until post 5h treatment. Before the 24h timepoint, de novo synthesized TLR-4 that is unbound to the MRF is being undetected. On the other hand, culturing MQ-NCSU cells in the presence of LPS activated NF- $\kappa$ B, which could be decreased by 20.2% through the addition of MRF as measured by EMSA. These results could explain the reduced activation of the inflammatory cytokine cascade in the presence of MRF, since triggering an inflammatory response by LPS requires TLR-4. These results are being subsequently confirmed in a second independent laboratory.

The third and final experiment in this series was an in vivo study, investigating the effect of supplementing MRF on the inflammatory response in broiler chickens under an LPS challenge. Two hundred one-day old Cobb 500 X Cobb 500 broiler chicks were randomly allocated into four groups in a completely randomized design. The four treatments were: 1) basal diet/ non-challenged; 2) MRF / non-challenged; 3) basal diet/ *E. coli* challenged; or 4) MRF/

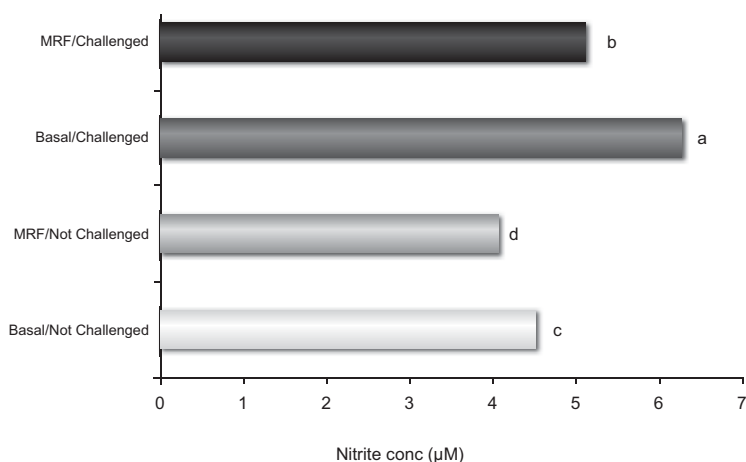
*E.coli* challenged. Birds were allowed *ad libitum* access to feed and water for the duration of the study. For birds in the challenge groups, 1ml of an enteropathogenic *E. coli* was administered by oral gavage. At 20 days, macrophages were isolated from the abdominal exudates from 10 birds/group. Cells from different birds in the same group were pooled and subjected to interleukin-6 (IL-6) and nitric oxide analyzes. At 1, 2 and 3 weeks of age the body weights of all birds were measured.

The performance of the birds was treatment related. The inclusion of MRF, challenged and non-challenged groups, in the diet led to numerically higher average body weight gains than the basal/ non-challenged group of birds after three weeks. The mean body weights of the basal/*E.coli* challenged birds was statistically lower than the mean body weights of the other three groups (Figure 6).



**Figure 6.** Mean body weight gains (g) of broiler chickens with and without an *E.coli* oral gavage challenge at day of hatch, with and without dietary supplementation of MRF. Means with a common superscript are not significantly different ( $P > 0.05$ ). (after Singabottra, 2005).

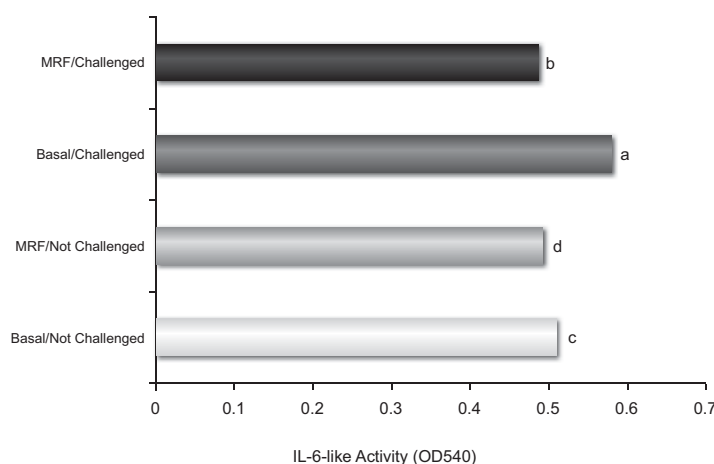
The MRF significantly reduced nitric oxide (measured as nitrate) production of macrophages from EPEC-challenged broiler chickens (Figure 7).



**Figure 7.** Nitrite production ( $\mu\text{M}$ ) of the abdominal exudates macrophage cells isolated from one of four treatment groups of broiler chickens with and without an *E.coli* oral gavage challenge at day of hatch, with and without dietary supplementation of MRF. Means with a common superscript are not significantly different ( $P > 0.05$ ). (after Singabottra, 2005).

MRF did not reduce IL-6-like activity of primary macrophages from *E.coli* challenged broiler chickens, but this may have been related to timing of sample collection. Three weeks may have been too late to observe this parameter. This statement can be supported by the fact that the primary chicken macrophages isolated from the chickens when stimulated by LPS had a significantly higher IL-6-like activity than cells from the other three treatments. The MRF treated cells in the presence of LPS demonstrated a significantly reduced IL-6-like activity (Figure 8).





**Figure 8.** *IL6-like activity of the primary chicken macrophage cells treated with medium, MRF, LPS, or MRF + LPS. Means with a common superscript are not significantly different ( $P > 0.05$ ). (after Singabottra, 2005).*

In conclusion, the in vitro study findings correlate well with the in vivo responses observed. The inclusion of MRF significantly reduced the effects of an LPS challenge in the broiler birds as evidenced by the lower nitric oxide levels produced in the macrophage and lower IL-6-like activity in the challenged primary chicken macrophages. In vivo these immunological changes demonstrated that supplementation of MRF in the diets of both *E.coli* challenged and non-challenged birds resulted in the improvement in average weight gain and total final weight of birds at the end of study. This would suggest that the dampening of the inflammatory response results in less energy being expended on immunological responses and instead making this energy available for growth (Humphrey and Klasing, 2003). This research finding suggests MRF to be a product with a similar immunomodulatory mechanism of action of the antibiotic growth promoters and therefore, has the potential to become a future replacement (Labro, 1998; Labro 2000; Niewold, 2007). Currently, MRF is being tested in pigs and larger scale studies are planned as of the time of writing this article.

## Conclusion

With the phasing out or banning of antibiotic growth promoters in the major food animal and poultry production markets around the world there has been intense activity to search for alternatives. The potential to maximize feed utilization, lower production costs and reduce environmental pollution in the animal production sector are of enormous economic benefit. As described the first part of this paper the focus of the search has been disparate with many different products being under consideration based on some form of empirical evidence. However, the results of studies performed on many of these 'alternatives' has been inconsistent, and led to confusion and frustration by many end users. To a large degree these alternatives work under certain conditions or when there is a low degree of microbial challenge in the system. However, when there is an immunological challenge on the animal many of these products cannot substitute for the former antibiotic growth promoters, either from a microbial management or anti-inflammatory standpoint. Hence, producers of growth promoters should focus more carefully on 'replacements' to AGPs. This is not as easy as it sounds as the mode of action of many of these compounds is still under dispute, 60 years after they were first used in animal feed as growth promoters! In the work that we have been doing recently in the field of anti-inflammatory mannan preparations we observe results similar to that of the AGPs with respect to growth performance. The data generated in the initial in vitro cell culture and broiler research studies indicates that there is merit in establishing large scale production studies to investigate MRF under field conditions, in pigs as well as broilers.

The identification of key biomarkers for inflammatory responses (cytokines, chemokines, toll-like receptors, etc) has facilitated the use of rapid screening of cell cultures to test candidate compounds in a matter of hours rather than weeks or months. A greater understanding of the regulatory mechanisms involved in the host intestinal immune defenses will greatly aide in the search for replacements to the AGPs. In the meanwhile, there are a number of alternatives that have been scientifically studied and known to give a growth performance response.

## Symposium conclusions

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There is little doubt that 'gut health' is a complex and multi-faceted area of pig science and production with a vast array of information and literature concerning the topic. The main purpose of this symposium was to inform and educate the audience in an area of pig science that is always in the press and attracts a considerable amount of attention. We hope that this symposium will set a platform for future discussion of 'gut health' in the context of the Australasian pig industry, and also assist in steering any future research in this general arena.

Important and rapid progress has been made in our understanding of this field particularly at the gastrointestinal tract level, with reference to the mechanisms underpinning the physiology, microbiota and localised immune system. Important concepts have emerged, such as the notion of stimulating/nullifying specific groups of bacteria to modify the gut environment. In the post-weaning period, where most attention has been focused, the compromised state of the young pig makes it an ideal candidate for the range of dietary products that might influence 'gut health', although significant work is still required in this area especially from the whole animal perspective. The paper by John Pluske and colleagues overviewed this general area of 'gut health' in the pig.

The paper by Colm Moran highlighted the documented modes of action of antibiotic feed additives as a rational starting point for the development of a replacement product. It is evident that comprehensive understanding must underpin any venture into this area of science; this is a fact made more obvious when one considers the many possible modes of action of the antibiotic feed additives and the possible combination of factors that can elicit a growth response and (or) protective influence against pathogenic microbiota, for example. It is clear that 'gut health' is a complex issue.

Ultimately though, the decision in practice to use a replacement dietary product (or products) in lieu of an antibiotic feed additive, where this is possible, will be distilled to similar factors that dictate the inclusion of other feed additives in a diet. These include price relative to effectiveness and efficacy of the product(s) over a broad range of conditions and circumstances in terms of growth performance, enteric disease control, or both.

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## Stimulating gut development with sodium butyrate to enhance litter viability and to reduce the post-weaning 'growth check'

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In piglets, the post-weaning 'growth check' is characterized by villous shortening, crypt elongation and reduced enzyme activities. As a result, the digestive and absorptive capacity of the small intestine after weaning is strongly reduced (Pluske *et al.*, 1995). Sodium butyrate (Na-butyrate) affects epithelial cell growth and differentiation, and increases the proliferation index in the intestinal crypts (Salminen *et al.*, 1998) and thereby reveals trophic effects on the gut mucosa. Galfi and Bokori (1990) were the first to show the positive influence of Na-butyrate on body weight gain, feed utilization and composition of intestinal microflora in piglets. The aim of this trial was to determine if the supplementation of Na-butyrate in the diet of lactating sows and post-weaning diets would improve litter viability and increase post-weaning growth rates.

One hundred and eight commercial F1 mixed parity sows ( $2.7 \pm 0.13$ ; mean  $\pm$  SE) were selected and individually housed in a farrowing shed. For three days before farrowing until day 25 of lactation the sows were fed either a standard lactating diet (Control) or a diet supplemented with 0.1% Na-Butyrate in the form of ADIMIX® Butyrate 98% (Admix®). Litter size was standardized within two days of birth and was similar between litters ( $10.4 \pm 0.1$ ; mean  $\pm$  SE). Weaned at 25 days of age, 120 male and 120 female weaners were randomly selected from each sow treatment, housed in pens of 10 piglets and continued on their respective dietary treatment from the day of weaning. The weaning diets with Na-butyrate were supplemented with Adimix® as follows: 0.2% until 7 days post weaning, then 0.15% 7-14 days post-weaning and 0.1% 14-21 days post-weaning. The data was analyzed by General Linear Model analysis of variance with the exception of piglet populations at day two and at weaning (Chi square).

This trial showed a numerical increase in the number of pigs weaned per sow with the addition of Na-butyrate as Adimix® to the lactation diet of 0.4 piglets per sow but this was not statistically significant (Table 1). There was no significant difference in weaning weight between treatments. Feeding Na-butyrate in the weaner diet significantly improved the growth rate and feed conversion, with a trend for improved average daily feed intake ( $P=0.068$ ) 0-20 days post-weaning. There were no treatment differences ( $P>0.05$ ) in losses or ill thrift after weaning. The use of Na-butyrate in the lactation diet and weaner diet can increase the number of pigs weaned and improve the growth performance of pigs post-weaning most likely due to a better utilization of nutrients.

**Table 1. Mean  $\pm$  SE of piglets from unsupplemented or Na-butyrate treatment on the number of pigs weaned and post-weaning performance 0 to 20 days**

	Control	Na-butyrate	Significance
Total piglets day 2	587	580	
Total piglets weaned	494	511	$P=0.051$ □ (3.84)
Piglet weight at weaning (kg)	$8.6 \pm 0.12$	$8.7 \pm 0.12$	NS
20-day weight (kg)	$14.0 \pm 0.20$	$15.15 \pm 0.22$	**
Rate of Gain (g/d)	$273 \pm 7.6$	$320 \pm 8.0$	**
Feed Conversion Ratio (g feed/g gain)	$1.45 \pm 0.021$	$1.34 \pm 0.022$	**
Daily Feed Intake (g/d)	$397 \pm 11$	$427 \pm 9$	NS

\*\* $P<0.01$ ; NS:  $P>0.05$  mean values not significantly different within rows.

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## Duodenal crypt depth of piglets at weaning is altered when a yeast extract is included in the lactating sow diet

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Nucleotides are the building blocks of nucleic acids which are required for new tissue growth. Including the yeast extract NuPro®, a product rich in nucleotides and glutamic acid, in the lactating sow diet may increase the amount of nucleotides in the milk and subsequently improve the development of the digestive tract – enabling piglets to better adapt to the many challenges faced after weaning. Therefore, it was hypothesized that the gut morphology of piglets from sows fed a lactation diet containing NuPro® would differ from those piglets from sows fed a control diet.

Twenty piglets were randomly selected at weaning from Large White x Landrace x Duroc sows that had either been fed a lactation diet containing 2% NuPro® or a control diet. The piglets were weaned at 21 days of age and did not receive creep feed before weaning. The piglets were euthanased (Na-pentobarbitone) and samples of the duodenum, jejunum and ileum collected and placed in formalin for 24 hours before being transferred to a 70% ethanol solution. The samples were then sectioned, mounted onto slides and stained with hematoxylin and eosin. About 10 measurements of the villous height and crypt depth were taken on each slide with a binocular light microscope and the average of these and the villous:crypt ratio determined. All data were analyzed by one-way analysis of variance using Genstat v.8.

The crypt depth tended to be greater ( $P=0.053$ ) and the villous to crypt ratio was significantly lower ( $P=0.037$ ) in the duodenum in piglets from sows fed the NuPro® diet compared to the control diet (Table 1). There were no differences between treatment groups in the villous height in the duodenum or in any measurement of other sections of the gut ( $P>0.05$ ). This study shows that duodenal morphology differs between piglets from sows fed either a NuPro® or control diet during lactation. This difference in morphology may help to partly explain why the progeny from sows fed 2% NuPro® during lactation in Moore *et al.* (2007) had an improved growth rate after weaning.

**Table 1. Small intestinal histology of piglets at weaning reared on sows fed a NuPro® or control diet in the lactation period (n=10)**

		Control	NuPro®	SED	p-value
<i>Duodenum</i>	Villous height (µm)	822	797	53.3	0.644
	Crypt depth (µm)	102	114	5.74	0.053
	Villous:crypt ratio	8.16	7.14	0.452	0.037
<i>Jejunum</i>	Villous height (µm)	768	725	44.9	0.351
	Crypt depth (µm)	101	102	4.57	0.906
	Villous:crypt ratio	7.70	7.28	0.577	0.475
<i>Ileum</i>	Villous height (µm)	472	444	29.9	0.356
	Crypt depth (µm)	105	101	4.58	0.428
	Villous:crypt ratio	4.62	4.53	0.339	0.790

Supported in part by Alltech Biotechnology Pty Ltd.

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# The effect of calcium formate and/or exposed yeast $\beta$ -glucans on growth performance of piglets under *Escherichia coli* challenge

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In the light of the current ban on antibiotic growth promoters in the European Union, several alternatives have been proposed. Among these, short chain organic acids (or salts thereof, e.g. Ca-formate) and yeast products are commonly used by stock feed producers in this region. Yeast products not only have nutritional value but can also have immune stimulatory effects and toxin-binding properties. Toxin-binding properties and binding of bacterial fimbriae are related to mannans in the yeast cell wall. Immune stimulation is due to  $\beta$ -1,3/1,6 glucans of the yeast cell wall, mainly modulating the non-specific immune system (Robertsen *et al.*, 1990). To expose the  $\beta$ -glucans, a proprietary extraction process is applied that removes the mannoprotein outer layers of the yeast cell wall. This results in a product like Fibosel<sup>®</sup>. The net outcome of immune stimulation should be a better health and an improved performance, especially under high infection pressure. It was hypothesized that a combination of Fibosel<sup>®</sup> and Ca-formate would be even more effective, because of their different modes of action.

In a trial with 72 individually housed weaned piglets ( $7.02 \pm 1.1$  kg) orally challenged with a pathogenic *E. coli*, the effects of feed additives on health and growth were studied. Ca-formate (10 kg/ton) alone or combined with glucans (Fibosel<sup>®</sup>, 150 ppm) were compared with avilamycin (40 ppm, Maxus<sup>®</sup>) and with a control feed without growth promoter or organic acids. Piglets received an oral *E. coli*-suspension on day five after weaning. For a period of 34 days average daily feed intake (ADFI), average daily gain (ADG) and feed efficiency (FE) were determined over weekly intervals, and diarrhoea incidence and severity were scored twice daily (Table 1). A general linear model procedure (SAS Inc) was used to estimate least-square means of the four different treatments. Weaning weight was used as a co-variable.

**Table 1. Overall effects of dietary treatments on growth performance and incidence of diarrhoea of piglets challenged with *E. coli* during days 5-34 post weaning**

	Start weight		ADFI		ADG		FE		Diar. inc. <sup>1</sup>
	(kg)	std	(g)	std	(g)	std	(g/g)	std	(%)
Control	6.98	1.06	407 A	95	286 a	87	0.69 a	0.11	36
Maxus <sup>®</sup>	6.73	1.16	526 B	140	397 b	115	0.75 b	0.06	27
Ca-formate	6.79	1.01	468 AB	112	350 b	84	0.75 b	0.07	25
Ca-fo+ Fibosel <sup>®</sup>	6.75	0.92	501 B	91	390 b	77	0.78 b	0.05	21

a,b = P<0.05; A,B = P<0.10. <sup>1</sup> incidence of severe watery diarrhoea.

As expected, variability in growth performance parameters was high within groups due to infection. Control piglets had a lower ADG, ADFI and consequently a lower FE compared to the other treatments. This was even more apparent during the initial post-challenge period of 14 days. During this period, body weight gain was improved over the control animals for avilamycin (90%), Ca-formate (40%) and the combination (70%). Ca-formate and even more so the combination of Ca-formate and Fibosel<sup>®</sup>, resulted in weight gains similar to the avilamycin supplemented group. No significant effects on the incidence of diarrhoea were observed (P>0.10). Organic acids and  $\beta$ -glucans appear to increase growth performance in piglets and are regarded as promising alternatives for antibiotic growth promoters in weaner pigs.

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# 8

## Pig growth and nutrition

## Separate and combined effects of oligofructose and inulin on post-weaning colibacillosis and weight gain: a preliminary study

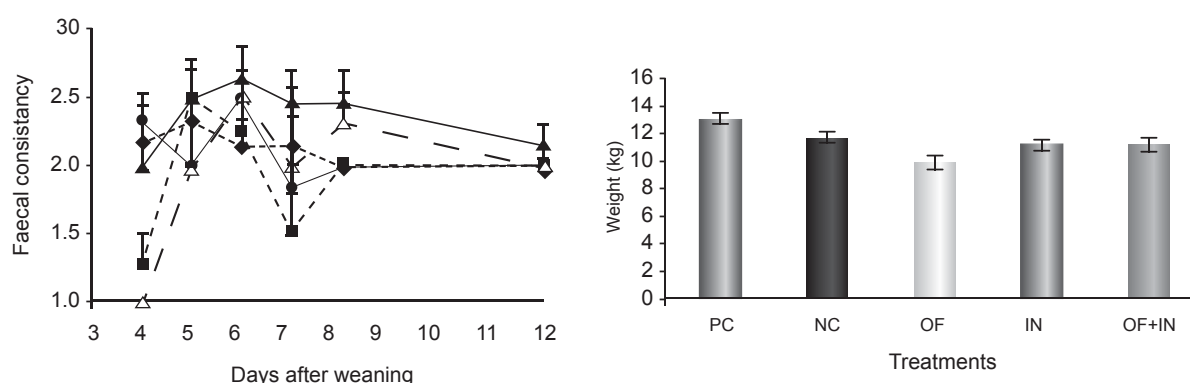
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Trends for a reduction in the use of dietary antibiotic growth promoters have caused increased interest in the use of alternative feed additives to maintain 'gut health' after weaning. Oligofructose and inulin are 'prebiotic' fructan-containing carbohydrates purported to enhance 'gut health' in newly-weaned pigs by altering microbial diversity (Konstantinov, *et al.*, 2003). However, their effects using a challenge model of post-weaning colibacillosis (PWC) have never been examined. The aim of the present study was to compare the separate and combined effects of oligofructose and inulin supplementation on the occurrence of diarrhoea and the weight performance in piglets experimentally challenged with enterotoxigenic *E. coli* (ETEC).

A total of 28 piglets ( $7.0 \pm 0.23$  kg; mean  $\pm$  SEM) weaned at 21 days of age were used in a completely randomized design with five treatments: 1) positive control (PC); 2) negative control (NC); 3) oligofructose (OF) at 40 g/kg of diet; 4) inulin (IN) at 40 g/kg of diet and 5) oligofructose and inulin combined (OF+IN) at 20 g/kg of diet each. The control diets (PC and NC) were identical wheat-based diets, with the experimental diets formulated from the control diets by substituting wheat with the appropriate amount of the prebiotic(s). All pigs, except PC, were inoculated with a  $\square$ -haemolytic strain of *E. coli* (O149, K91, K88) on days 3, 4, 5 and 6 after weaning. Faecal consistency was assessed daily as dry (1), moist (2) or diarrhoea (3). ETEC shedding was expressed as a percentage of the total *E. coli* population grown on 5% sheep blood agar plates. The final weight was measured on day 21. Treatment effects were evaluated using the GLM procedure in SPSS (SAS Inc v14.0) using treatment as the fixed variable and weaning weight as a covariate. Faecal consistency was analyzed as repeated measures analysis of variance.

The faeces of piglets fed OF were more ( $P < 0.05$ ) liquid than those of the PC, IN and OF+IN pigs (Figure 1). Pigs fed inulin had the lowest incidence of PWC ( $P = 0.008$ ) on day four, the day after inoculation started. Pigs fed OF showed a tendency ( $P = 0.061$ ) to have the lowest weight at the end of the trial (day 21). Furthermore, there was a negative correlation between total *E. coli* shedding and total weight gain ( $r = -0.58$ ,  $P = 0.001$ ) (data not shown). Based on this preliminary study, inulin has been selected for further investigations evaluating its effects specifically on PWC under experimental challenge conditions, but also on growth performance and diversity of the microbiota.



**Figure 1.** Effects of oligofructose, inulin, and oligofructose + inulin on faecal consistency and pig weight at 42 days of age

Supported by the ARC Linkage scheme, Danish Pig Production, WA Department of Agriculture and Food and Wandalup Farms.

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## Feeding a low protein amino-acid supplemented diet after weaning reduces incidence of post-weaning diarrhoea

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Post-weaning diarrhoea (PWD), a condition associated with proliferation of  $\beta$ -haemolytic strains of *Escherichia coli* in the small and large intestine, often occurs after weaning. Once attached to the small intestinal epithelium, these strains of *E. coli* can disrupt digestive and absorptive functions of the enterocytes by releasing both heat labile toxins (LT) and heat stable toxins (ST; variants STa and STb) that are responsible for hypersecretory diarrhoea (Pluske *et al.*, 2002). Numerous dietary strategies have been attempted to ameliorate the losses associated with PWD. Of these, feeding a lower-protein diet with supplementation of essential amino acids has been suggested because by-products of protein fermentation, such as ammonia and amines, are implicated in the aetiology of the condition (Aumaitre *et al.*, 1995). However feeding a lower-protein diet after weaning is associated with reductions in performance (Nyachoti *et al.*, 2006). In this study, we hypothesized that feeding a low protein diet for a short period of time after weaning would reduce PWD by reducing protein fermentation in the LI.

Seventy-two female pigs (Large White x Landrace) aged 21 days and weighing  $5.9 \pm 0.12$  kg (mean  $\pm$  SEM) were randomly allocated to six treatments based on weaning weight. The treatments were: high protein diet (24% CP) to day 14 post weaning (HP14); low protein amino-acid supplemented diet (18% CP) to either day 14 (LP14) or day 7 (LP7). Half of the pigs (n=12) per treatment were infected (I) with 3 mL, 8 mL and 8 mL (107 CFU/mL) of *E. coli* (serotype O149; K91; K88) at 72, 96 and 120 hours after arrival, respectively. Diet LP was fortified with lysine, methionine, tryptophan, threonine, isoleucine and valine, based on proposed ideal amino acid patterns. An intermediate diet (20.5% CP) was fed to pigs at the conclusion of each treatment. None of the diets contained antimicrobial compounds. Rectal swabs were scored according to the number of positive streaked sections (0-5) on days zero, five, seven, 10 and 14. Plasma urea nitrogen (PUN) was measured on day seven and 14. Diarrhoea index (DI) was recorded for the first 14 days. Repeated-measures analysis of variance (SPSS Inc v.14.0, SAS Inc) was used to analyze the results.

Infection increased *E. coli* shedding ( $P < 0.001$ ) and the DI ( $P < 0.001$ ) (Table 1). Piglets fed a low-protein diet both for seven and 14 days after weaning showed lower levels of PUN ( $P < 0.001$ ) and the DI ( $P < 0.001$ ) than piglets fed a high-protein diet. The effects of protein level on the decreased PUN and DI were independent of *E. coli* infection. These results support our hypothesis that feeding a lower-protein, amino-acid supplemented diet for a shorter period of time after weaning reduces PWD by reducing the protein load entering the hindgut of the young pig.

**Table 1. Effect of a low protein diet on shedding of  $\beta$ -haemolytic *E. coli*, PUN and DI<sup>2</sup>**

Infection Treatment	Non- infected			Infected			SEM	P-value		
	HP14	LP14	LP7	HP14	LP14	LP7		PL <sup>1</sup>	I	PLx I
<i>E. Coli</i> score	0.3	0.1	0.1	0.6	0.6	0.5	0.11	0.373	<0.001	0.490
PUN	5.2 <sup>a</sup>	2.1 <sup>c</sup>	3.4 <sup>b</sup>	5.6	2.2	3.9	0.59	<0.001	0.467	0.944
DI <sup>2</sup>	19.8 <sup>a</sup>	8.5 <sup>c</sup>	9.6 <sup>b</sup>	44.2 <sup>a</sup>	31.6 <sup>b</sup>	21.4 <sup>c</sup>	3.54	<0.001	<0.001	0.154

<sup>1</sup>Protein level; <sup>2</sup>Diarrhoea index: Proportion of days with diarrhoea with respect to total days (14 days) on trial.

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## Neonatal oxytocin administration and weaning onto a gruel based diet reduce weight loss at weaning and enhance gastric leptin expression

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Administering oxytocin to neonatal rats has positive long-term effects on growth and development (Uvnas-Moberg and Petersson, 2005). These effects include a reduction in the stress response to weaning, increased post-weaning feed intake and alterations in the expression of gastrointestinal (GI) hormones regulating feed intake (Uvnas-Moberg *et al.*, 1998; Sohlstrom *et al.*, 1999). Two GI hormones of importance in regulating feed intake are ghrelin and leptin, which have antagonistic actions. Ghrelin expression is increased in response to fasting and leptin expression increases rapidly in response to feed intake. Since weaning the piglet is associated with stress and growth restriction, this study examined whether oxytocin given to young pigs could reduce the extent of the post-weaning growth check, along with any associated changes in ghrelin and leptin expression.

A total of 240 piglets (Large White x Landrace) suckling 20 sows were injected daily with 1mg/kg of either oxytocin or saline s.c. from 0-14 days of age. All piglets were weaned at 21 days onto either a dry pelleted diet or a gruel based diet that was fed for the first week post-weaning, after which all piglets received a dry pelleted feed. On day 10, 21 and 28 piglets were slaughtered and the fundus of the stomach collected for gene expression analysis via real-time polymerase chain reaction. All data were analyzed by analysis of variance.

Oxytocin administration resulted in enhanced gastric leptin expression at day 10 ( $P=0.028$ ). Gastric leptin was unaltered in oxytocin administered piglets at weaning ( $P=0.547$ ). Gastric ghrelin expression was also unaltered at day 10 ( $P=0.528$ ) and at weaning ( $P=0.930$ ) by oxytocin administration. Neonatal administration of oxytocin partially alleviated the post-weaning lag in growth by reducing weight loss over the first two days post-weaning (-214 vs. -293 g/day,  $P=0.03$ ). This was associated with increased expression of both gastric ghrelin ( $P=0.016$ ) and gastric leptin ( $P=0.017$ ). However, no differences in feed intake were observed between oxytocin- and saline-injected piglets over the first two days post-weaning (369 vs. 374 g/day,  $P=0.70$ ). In addition, weaning onto a gruel-based diet was effective in promoting a higher rate of daily gain compared to piglets not offered gruel in the first week after weaning (124 v 76 g/day,  $P<0.001$ ). This was associated with increased feed intake (561 vs. 131 g/day,  $P<0.001$ ) and enhanced gastric leptin expression at day 28 ( $P=0.018$ ). Gastric ghrelin expression at day 28 was not influenced by gruel feeding ( $P=0.895$ ). Oxytocin administration and weaning onto a gruel-based diet are both effective means of enhancing gastric leptin expression and reducing weaning weight loss.

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## Pigs born light increase adipose tissue deposition following a restricted protein intake during early growth

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Compensatory growth (CG) responses following a period of reduced protein intake have been observed in pigs, although the degree of compensation can be variable. It is possible that fetal development in utero may account for some of this variation, with pigs born at low birth weights having fewer muscle fibers at birth than those born heavier (Hegarty and Allen, 1978; Handel and Stickland, 1984). As the number of muscle fibers is a major determinant of postnatal muscle growth, low birth weight pigs may not have the capacity to fully 'catch up' following a short period of protein restriction. As such, this study investigates whether birth weight will influence the CG response of gilts following a period of reduced protein intake during the weaner period.

Ninety-six gilts were selected at weaning based on birth weight and individually housed. Pigs were allocated to a 2 x 2 factorial experiment, with the respective factors being birth weight (light or heavy) and dietary lysine intake from 32 to 55 days of age (Control – 0.80 g lysine/MJ digestible energy (DE), or restricted – 0.64 g lysine/kg DE). Outside of the period of restriction all pigs received nutritionally adequate diets. Feed was offered *ad libitum* for the entire experimental period. Data were analyzed using REML.

**Table 1. Effect of birth weight and weaner protein intake on daily gain through to slaughter**

Measurement	Light		Heavy		SED	P-value		
	Control	Rest	Control	Rest		BW	Diet (D)	BW x D
Average daily gain (kg/d)								
32 – 55 days of age	0.40	0.37	0.59	0.49	0.027	<0.001	<0.001	0.065
55 – 153 days of age	0.85	0.95	1.02	1.01	0.040	<0.001	0.076	0.080
32 – 153 days of age	0.77	0.83	0.94	0.91	0.036	<0.001	0.349	0.070

At birth, the light birth weight pigs were 37% lighter than their heavier contemporaries (1.24 vs. 1.98 kg respectively,  $P < 0.001$ ). From 32 to 55 days of age the restricted pigs grew more slowly than the controls (0.42 vs. 0.50 kg/d respectively  $P < 0.001$ ) (Table 1), while feed intake was similar. During the post restriction period, 55 to 153 days of age, the restricted pigs tended to gain faster than the controls (0.98 vs. 0.93 kg/d respectively,  $P = 0.076$ ), particularly the light birth weight pigs. At 153 days of age the light birth weight pigs remained smaller than those born heavy (103.6 vs. 121.9 kg respectively,  $P < 0.001$ ). Interestingly, pigs born light and restricted in protein intake during the weaner period were 7.6% heavier than the light controls, while at commercial slaughter weights there was little difference between the two heavy birth weight treatment groups. Over the entire study the restricted pigs tended to gain more adipose tissue than the controls (218 vs. 197 g/d respectively,  $P = 0.059$ ), particularly the light birth weight pigs. Regardless of slaughter weight, gilts that were born light were fatter (+2.4% fat) than their heavier contemporaries. These data suggest that birth weight may influence the CG response, with compensatory weight gain in low birth weight pigs favouring adipose tissue deposition. As such, producers utilizing strategic periods of protein restriction may need to manage and market light birth weight pigs separately.

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# The effect of betaine supplementation on growth performance of piglets raised under sub-optimal management conditions

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Betaine is the trimethyl derivative of the amino acid glycine. As a by-product of sugarbeet processing, betaine is commercially available as a feed additive. Due to its chemical structure, various functions have been described for the betaine molecule. Increasingly, research on betaine as a feed additive is focusing on its osmo-protective properties in relation to gut functionality and health (Eklund *et al.*, 2005).

An experiment was conducted to test the effect of dietary <sup>TNI</sup>betain (Trouw Nutrition International; 96% betaine) supplementation in weaned piglets housed under suboptimal conditions. It was hypothesized that <sup>TNI</sup>betain may diminish diarrhoea problems and improve performance of weaned piglets because of its gut health promoting properties. In addition, it was speculated that its effects may be more pronounced if piglets are fed diets focused at a higher growth performance. These diets typically contain a higher balanced protein and net energy level.

Hypor Body piglets were blocked by gender and weaning weight and assigned to four dietary treatments during the first four weeks post-weaning, with 16 pens per treatment and three piglets per pen. Sub-optimal conditions were created by not cleaning the room before the start of the trial, introducing sow manure, lower ambient temperature settings and increasing dust concentration. The trial was set up as a 2x2 factorial experiment with <sup>TNI</sup>betain (0 and 0.2%) and diet specifications (low and high) as main effects. Feed and water were available *ad libitum*. No antibiotic growth promoters were used. Instead, diets contained a mixture of short and medium chain fatty acids and beta-glucans. A GLM procedure (SAS Inc v8.2) was used to estimate least-square means with block (entangled with gender), betaine and diet type as independent variables.

As intended, overall performance level was at a low level (Table 1). No interactions between main effects were observed ( $P > 0.10$ ). Average daily feed intake (ADFI) and average daily gain (ADG) did not differ between treatments. The high specification diet improved overall feed efficiency (FE) but also increased the incidence of diarrhoea (61 vs. 68%;  $P < 0.001$ ).

**Table 1 Overall effects of betaine (<sup>TNI</sup>betain) and diet specifications on growth performance of piglets (0-28 days post weaning)**

Treatment	<sup>TNI</sup> betain		Diet specs <sup>1</sup>		SEM	P-values		
	0%	0.2%	LP	HP		Betaine	Specs	Bet x Specs
ADG (g)	298	298	292	304	7	0.995	0.222	0.369
ADFI (g)	459	446	461	443	10	0.336	0.202	0.445
FE (g/g)	0.650	0.675	0.639	0.686	0.009	0.048	0.001	0.491

<sup>1</sup> LP and HP = low and high (growth) performance diets respectively. I.e. 18 vs. 20% balanced crude protein (day 0-28) and 1.27 vs. 1.33 (day 0-7) and 1.01 vs. 1.14 (day 7-28) g ileal dig. Lys/ MJ NE.

Supplementing <sup>TNI</sup>betain improved overall FE and reduced the incidence of diarrhoea in week 3 and 4 of the experiment (75 vs. 68%,  $P = 0.021$ ). The reported energy sparing and gut health promoting effects of betaine (Eklund *et al.*, 2005) are confirmed in this study.

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## Effects of dietary betaine on ileal and faecal digestibilities and intestinal microbial fermentation of crude fibre in piglets

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Dietary betaine supplementation has been shown to increase fibre digestibilities in piglets (Eklund *et al.*, 2006). In the current study we investigated the effects of betaine on ileal and faecal digestibilities and the microbial fermentation of crude fibre (CF) in weaned pigs.

Four five-week-old barrows (German Landrace x Piétrain, initial body weight (BW) of  $8.6 \pm 1.3$  kg) were surgically fitted with T-cannulas at the distal ileum. The basal diet consisted of barley, wheat and soybean meal (13.88 MJ DE/kg; 11 g/kg lysine; 37 g/kg CF). The basal diet was fed either alone (Control) or supplemented with 0.45 % anhydrous betaine (Betaine). The feed allowance was restricted to 4.5% of individual BW. Following seven days of adaptation to the experimental diets, faecal and ileal nutrient digestibilities and microbial parameters were assessed via four repeated measurements with two pigs per treatment resulting in a total of eight observations per treatment. The data were analyzed using the MIXED procedure of SAS (SAS Inc). Errors of repeated measurements on the same subject (animal within treatment) were assumed to be serially correlated.

Supplementing betaine improved ileal dry matter (DM) digestibility by 1.2% ( $P=0.030$ ), whereas faecal DM digestibility was not affected ( $P=1.0$ ) (Table 1). Moreover, supplementing betaine tended to increase ileal digestibilities by 6.5 ( $P=0.068$ ) and faecal CF digestibilities by 4.7 ( $P=0.117$ ) percentage units. This increase in microbial CF fermentation coincided with higher concentrations of diaminopimelic acid (DAP) as the bacterial marker in ileal digesta ( $P=0.117$ ) and faeces ( $P=0.004$ ), which suggests increased microbial growth along the digestive tract. Simultaneously, there was an increase in volatile fatty acid (VFA) concentrations in ileal digesta ( $P=0.008$ ), which confirms an increase in microbial fermentation activity due to betaine supplementation. Faecal concentrations of VFA were not affected ( $P=0.848$ ). In summary, these data suggest betaine has the potential to improve intestinal microbial fermentation.

**Table 1. Effect of betaine supplementation on ileal and faecal digestibilities and microbial fermentation of crude fibre**

Item		Control	Betaine	SEM	P-value
Dry matter digestibility (%)	ileal	68.8	70.0	0.29	0.030
	faecal	86.8	86.8	0.75	1.000
Crude fibre digestibility (%)	ileal	1.30	7.80	2.60	0.068
	faecal	37.9	42.6	1.94	0.117
Diaminopimelic acid (mg/kg DM)	ileal	43.7	56.8	4.70	0.117
	faecal	545	872	64.4	0.004
Volatile fatty acids (mmol/kg DM)	ileal	141	218	16.8	0.008
	faecal	468	413	178.3	0.848

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## Bone mineralization and phosphorus digestibility in weaned pigs fed diets containing thermostable phytase

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Exogenous microbial phytases are often added to commercial feeds to improve phytate digestibility and reduce phosphorus (P) excretion and other negative effects associated with ingesting phytate of plant origin (Selle *et al.*, 2006). However, to be effective *in vivo* it is important the exogenous phytase survives the feed processing phase, which often occurs under high temperatures. In this study we evaluated the thermostability and bioefficacy of a novel coated (C) bacteria-derived phytase (Phyzyme XP; 6-phytase, EC 3.1.3.26) in pigs fed corn-soybean meal-based basal diets. Individually-housed, 28-day-old male weaned pigs (Duroc x F1 Large White x Landrace), with a mean bodyweight of 8.2 kg, were allocated in a randomized complete block design to seven dietary treatments: 1) Positive Control ([PC-mash] 1.01% Ca and 0.4% available P); 2) Negative Control ([NC-mash] 0.83% Ca and 0.22% available P); 3) NC pelleted at 90°C; 4) NC plus 500 FTU/kg phytase-mash; 5) NC plus 500 FTU/kg C phytase-mash; 6) NC plus 500 FTU/kg C phytase pelleted at 80°C; and 7) NC plus 500 FTU/kg C phytase pelleted at 90°C. Basal PC-mash and NC-mash diets were formulated to similar nutrient contents, except for Ca and P. Chromic oxide was added to the diets as a marker and each diet was fed for 21 days. Bodyweight and feed intake were measured weekly and faecal grab samples were collected for two days to estimate digestibility. All pigs were euthanized on day 21 and the third metacarpal bone removed for bone mineralization measurements. Bone ash and bone strength are highly correlated and bone ash as an index of bone strength is a sensitive criterion for assessing efficacy (Onyango *et al.*, 2003). Data were analyzed using the GLM procedure of SAS (SAS Inc).

Pelleting had no effect on performance, enabling a comparison of the mash and pelleted results (Table 1). Ash weight was higher ( $P < 0.05$ ) for pigs fed the diets supplemented with PC and phytase than for those fed the NC diet. Digestibility of P was higher ( $P < 0.05$ ) for diets 1, 4, 6 and 7 than for diets 2, 3 and 5. No differences ( $P > 0.05$ ) were observed between pigs fed pelleted diets containing C-phytase and pigs fed the phytase diet. Reducing dietary P by 0.12% is recommended when 500 FTU/kg phytase is added. However, despite the NC diet containing almost half the phosphorus content of the PC diet (0.22 vs 0.40%), the bone mineralisation and animal performance of pigs fed the uncoated and coated phytase were equivalent. The results of this study demonstrated that the novel coated phytase maintained similar bioefficacy to the uncoated phytase when included in either mash or pelleted feed manufactured at up to 90°C.

**Table 1. Performance, phosphorus digestibility and bone mineralization in weaned pigs<sup>1</sup>**

Dietary treatment	ADG g/day	FCR	Phosphorus <sup>2</sup> (% ash)	Ash weight <sup>2</sup> (g)	Phosphorus digestibility <sup>3</sup> (%)
1. PC-mash	567 <sup>a</sup>	1.39 <sup>b</sup>	17.8 <sup>a</sup>	1.37 <sup>a</sup>	56.8 <sup>b</sup>
2. NC-mash	435 <sup>b</sup>	1.58 <sup>a</sup>	17.2 <sup>b</sup>	0.91 <sup>c</sup>	51.6 <sup>c</sup>
3. NC pelleted at 90°C	449 <sup>b</sup>	1.57 <sup>a</sup>	17.7 <sup>ab</sup>	0.91 <sup>c</sup>	47.9 <sup>c</sup>
4. NC + Phytase-mash	500 <sup>ab</sup>	1.44 <sup>ab</sup>	17.8 <sup>a</sup>	1.05 <sup>b</sup>	59.8 <sup>ab</sup>
5. NC + C phytase-mash	505 <sup>ab</sup>	1.48 <sup>ab</sup>	17.9 <sup>a</sup>	1.05 <sup>b</sup>	53.0 <sup>c</sup>
6. NC + C phytase pelleted at 80°C	530 <sup>ab</sup>	1.39 <sup>b</sup>	17.9 <sup>a</sup>	1.11 <sup>b</sup>	62.8 <sup>a</sup>
7. NC + C phytase pelleted at 90°C	520 <sup>ab</sup>	1.36 <sup>b</sup>	17.8 <sup>a</sup>	1.12 <sup>b</sup>	59.4 <sup>b</sup>
Pooled SEM	39.9	0.058	0.15	0.040	1.35

<sup>1</sup>Mean values determined from eight pigs per treatment; <sup>2</sup>Values obtained from third metacarpal bone defatted; <sup>3</sup>Total tract digestibility; <sup>a,b,c</sup>Means within a column with different superscript differ significantly ( $P < 0.05$ ).

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## Only severe gastric ulcers reduce performance in growing-finishing pigs

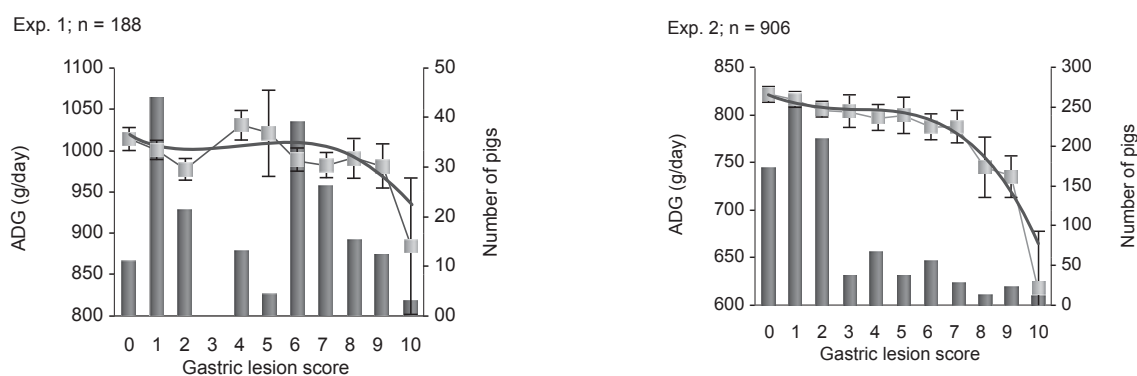
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Ulceration in the pars oesophageal region of the stomach of pigs is frequently registered at slaughter. Pigs with gastric lesions are reported to have lowered productivity (Ayles *et al.*, 1996), but other studies have found no significant effects on performance (Guise *et al.*, 1997). The objective of this study was to quantify the correlation between ulcer severity and growth performance in growing-finishing pigs.

Individual daily gain was compared with the degree of gastric ulceration at slaughter in two feeding experiments. In Exp. 1, the outcome of dietary inclusion of 10% alfalfa hay meal in a pelleted diet (2 mm pellets) was examined while in Exp. 2 the effect of coarser grinding in pelleted diets (3.5 mm pellets) on performance and gastric ulceration was studied. The pigs were in the experiments from about 30 kg until slaughter at around 100 kg. The stomachs from 188 pigs in Exp. 1 and 906 pigs in Exp. 2 were collected at the abattoir and lesions in the pars oesophagea were scored on a scale from zero to 10: zero being normal and 10 having severe gastric ulceration (Christensen, 1998). Influence of stomach score on average daily gain (ADG) was analyzed univariately in a normal linear model using the GLM procedure in SAS (SAS Inc v.9.13).

In Exp. 1, no significant ( $P=0.18$ ) effect of gastric ulceration on growth performance was detected, due to one of three pigs given a gastric score of 10 that grew at 1042 g/day (Figure 1). In Exp. 2, the number of animals was increased and a highly significant ( $P<0.001$ ) correlation between gastric ulceration score and growth was detected.



**Figure 1.** Influence of gastric lesions on average daily gain ( $\square$  mean  $\pm$  SEM) in growing-finishing pigs and number of pigs with a given score in two different experiments

Daily gain was only moderately affected until a gastric score of 6-7 equivalent to lesions and/or scars of above 0.5 cm<sup>2</sup>. Daily gain did not drop significantly until a score of 8-10 corresponding to lesions/scars above 5 cm<sup>2</sup> and contraction of the oesophageal opening. It is concluded that only severe gastric ulcers have a negative impact on growth rate in growing-finishing pigs.

This work was sponsored by Danish Pig Production.

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# 9

## Pig health and nutrition

## Colicin genes in commensal and enterotoxigenic *Escherichia coli* isolates from the gastrointestinal tracts of pigs

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The gastrointestinal tract (GIT) is made up of sections each performing a specific function. Within this GIT environment, there exists considerable diversity in gut microflora with bacteria competing with each other for nutrients as well as an ecological niche in each microhabitat. Commensal *E. coli* are normally the primary colonizers of the GIT and pathogenic *E. coli* must compete against this population to survive. Strategies used in this competition include more efficient assimilation of essential nutrients, better ability to bind to receptors on the luminal face of intestinal epithelial cells and the production of anti-microbial compounds, such as bacteriocins that may inhibit or kill other bacteria. Colicins are bacteriocins produced by *E. coli* and other members of the family Enterobacteriaceae such as *Citrobacter* and *Shigella*. The antimicrobial properties of colicins make them excellent candidates for use by commensal bacteria against pathogenic bacteria in the GIT. Therefore, it is important to assess whether commensal and pathogenic *E. coli* isolates differ in their ownership of colicin genes.

A multiplex polymerase chain reaction (PCR) was optimized and applied to survey the prevalence of eleven common colicin genes (A, B, D, E1, E2, E3, E6, E7, Ia-Ib, K, and M) in *E. coli* isolates. The study focused on 152 porcine commensal *E. coli* isolates obtained from different compartments of the GIT (duodenum, ileum, colon and faeces). In addition, 132 isolates from pigs post-weaning were included in the PCR analysis.

Five individual colicins (B, E1, E3, E7 and Ia-Ib) and ten colicin combinations (B/Ia-Ib, B/E1/M, B/E7/M, B/M, B/Ia-Ib/M, E1/E3, E1/E7, E1/E2/E3, E2/E7 and E2/Ia-Ib) were detected (Tables 1 and 2). About 2.9% of the porcine pathogenic isolates had at least one colicin gene with colicins B, E1 and E3 occurring at frequencies of 0.7%, 0.75% and 1.5% respectively while the dual colicin B/M occurred at a much higher frequency of 15.4%. In contrast, there was a significantly higher carriage of single and multiple colicin genes in porcine commensal *E. coli* isolates. Among the single colicins, colicins E1 and E7 were the most common while colicin B/M was the most frequent among the multiple colicin combinations. Furthermore, there appeared to be differences in the types of colicins found in commensal *E. coli* isolates recovered from different intestinal sections.

**Table 1. Frequency of single colicin genes in different intestinal sections of porcine commensal *E. coli* and in pathogenic *E. coli* isolates**

Colicins	Commensals				Pathogens (n=132)
	Duodenum (n=30)	Ileum (n=39)	Colon (n=39)	Faeces (n=44)	
A	0.0%	0.0%	0.0%	0.0%	0.0%
B	0.0%	0.0%	0.0%	0.0%	0.7%
D	0.0%	0.0%	0.0%	0.0%	0.0%
E1	0.0%	10.3%	10.3%	11.4%	0.7%
E2	0.0%	0.0%	0.0%	0.0%	0.0%
E3	0.0%	5.1%	0.0%	0.0%	1.5%
E6	0.0%	0.0%	0.0%	0.0%	0.0%
E7	3.3%	5.1%	12.8%	22.7%	0.0%
Ia - Ib	3.3%	10.3%	0.0%	0.0%	0.0%
K	3.3%	0.0%	0.0%	0.0%	0.0%
M	0.0%	2.6%	0.0%	0.0%	0.0%

**Table 2. Frequency of multiple colicin genes in different intestinal sections of porcine commensal *E. coli* and in pathogenic *E. coli* isolates**

Colicins	Commensals				Pathogens (n=132)
	Duodenum (n=30)	Ileum (n=39)	Colon (n=39)	Faeces (n=44)	
A	0.0%	0.0%	0.0%	0.0%	0.0%
B	0.0%	0.0%	0.0%	0.0%	0.7%
D	0.0%	0.0%	0.0%	0.0%	0.0%
E1	0.0%	10.3%	10.3%	11.4%	0.7%
E2	0.0%	0.0%	0.0%	0.0%	0.0%
E3	0.0%	5.1%	0.0%	0.0%	1.5%
E6	0.0%	0.0%	0.0%	0.0%	0.0%
E7	3.3%	5.1%	12.8%	22.7%	0.0%
Ia - Ib	3.3%	10.3%	0.0%	0.0%	0.0%
K	3.3%	0.0%	0.0%	0.0%	0.0%
M	0.0%	2.6%	0.0%	0.0%	0.0%

From these findings it can be concluded that 1) more commensal *E. coli* carry colicin genes than pathogenic *E. coli*; 2) commensal in different compartments use different colicin gene combinations and 3) pathogenic *E. coli* are very restricted in the types of colicin genes they carry. Therefore, pathogenic *E. coli* are at a selective disadvantage if they encounter commensal *E. coli* carrying the appropriate combinations of colicins in different GIT compartments.

This work was supported by the Pork CRC.



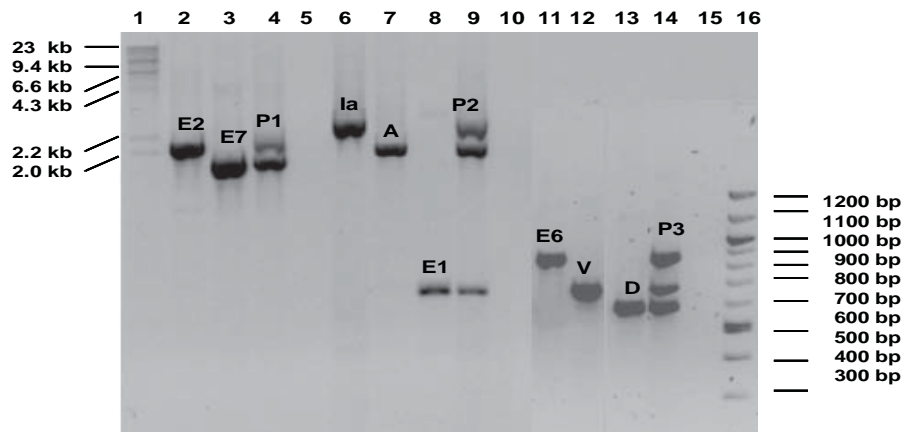
## Fitness genes of commensal and enterotoxigenic *Escherichia coli*

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Neonatal and post-weaning colibacillosis (NC/PWC) caused by enterotoxigenic *E. coli* (ETEC) strains can pose serious economic losses to the pig industry through reduced growth rate and increased mortality. Many theories abound as to why porcine ETEC, represented by serogroups O8, O9, O101, O120 (NC) and O8, O138, O139, O141, O147, O149, O157 (PWC) have evolved into pathogens. For instance, the presence of fimbriae and adhesins on the bacteria as well as corresponding matching receptors on the luminal face of intestinal epithelial cells provide an opportunity for colonization of both pathogenic and commensal strains. If the former dominates, then disease ensues. Maternal antibodies preventing ETEC attachment are required to protect the neonate against a background of increased stress from a relatively naïve and immature immune system. On the other hand, early colonization of the gastrointestinal tract (GIT) with non-pathogenic commensal organisms (a process known as probiosis) may provide a competitive barrier to colonization by pathogenic strains. Current understanding of the early gastrointestinal colonization dynamics between commensals and pathogens in neonatal pigs is limited. In this study, we hypothesised that fitness genes encoding colicins and microcins possessed by commensals are different from those possessed by ETEC. Colicins and microcins are peptides that display bacteriocidal activity against other microbes. If colicins and microcins produced by commensals are antagonistic against ETEC, then they may serve a very important role in intestinal prebiosis and prevention of disease.

The experimental strategy involved assembling a collection of porcine commensal *E. coli* and ETEC strains and screening the isolates with a gene panel consisting of eight colicin genes. The eight colicin genes (E2, E7, Ia, A, E1, E6, V, D) were separated into three multiplex pools (Figure 1).



**Figure 1.** Ethidium bromide-stained gel showing amplified bands corresponding to the eight colicin genes in the three multiplex pools – P1 (lane 4), P2 (lane 9) and P3 (lane 14)

Results showed that 35% of the commensals carried colicin genes compared to only 6% of pathogens. A second and more important finding was that commensal strains carried different colicin genes compared to pathogenic strains. This suggests that the right combination of commensal *E. coli* strains could potentially be used to achieve intestinal prebiosis in neonatal pigs and limit colonization by ETEC.

Supported in part by International Animal Health Products Pty Ltd.

## Phenotyping *Escherichia coli* antimicrobial resistances in pigs

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The use of antimicrobial agents in intensive animal production has increased public health concerns about the possible transfer of antimicrobial resistance genes (ARGs) via the food chain to human pathogens. Publicity on this subject can affect consumer confidence in animal products and raise ethical concerns about intensive systems of animal production. The pig industry is presently addressing these issues through a two phase survey on the use of antimicrobials in Australian pig production systems. The first phase involves an on-farm usage survey while the second requires formal sampling for phenotypic analysis of antimicrobial resistance. Such surveys have traditionally relied on faecal sampling and this work was established to determine if faecal sampling truly reflected the resistance profiles of the test organism, *E. coli*, throughout the gastrointestinal tract.

Three finisher pigs were selected from two different farms at an abattoir. Intestinal contents from the duodenum, ileum, caecum, colon and rectum were collected from euthanized animals and frozen in brain heart infusion broth containing 20% glycerol (-80°C). Suitably diluted aliquots were plated to yield 200-300 colonies. These were transferred to hydrophobic grid membrane filters (HGMP) and incubated overnight at 37°C on MacConkey agar (Master Grids). The master grids were then replicated onto four Muller-Hinton agar plates containing the following antimicrobials: ceftiofur (8 µg/ml), florfenicol (4 µg/ml), gentamicin (4 µg/ml) and ampicillin (32 µg/ml). The frequency of single and multiple resistances were determined using HGMPRES image analysis software.

The incidence of antimicrobial resistances for each intestinal compartment was averaged for all six pigs (Table 1). Resistance frequencies were similar between compartments and it was concluded that faecal samples are sufficiently representative of the pigs gut flora to be used for assessing phenotypic resistance in commensal *E. coli*.

**Table 1. Percentage of antimicrobial resistant *E. coli* in the different intestinal sections of pigs**

	Ceftio- fur	Florfen- icol	Genta- micin	Ampi- cilin	Cef/ Flo/ Gent/ Amp	Cef/ Gent/ Amp	Flo/ Gent/ Amp	Cef/ Amp	Flo/ Gent/ Amp	Flo/ Gent	Flo/ Amp	Gent/ Amp
Duodenum	2.2%	7.8%	7.4%	56.1%	0.0%	0.0%	0.1%	4.2%	0.0%	0.0%	3.1%	7.8%
Ileum	1.2%	4.6%	3.3%	53.6%	0.15	0.6%	0.8%	0.6%	0.1%	0.0%	4.2%	3.3%
Caecum	1.3%	18.5%	32.1%	66.5%	0.4%	0.0%	1.0%	0.2%	3.1%	0.4%	18.8%	29.3%
Colon	2.2%	10.6%	10.9%	59.5%	0.0%	0.8%	0.7%	1.1%	0.6%	0.0%	15.2%	10.3%
Rectum	0.8%	14.9%	9.8%	56.5%	0.0%	0.1%	0.0%	0.8%	3.1%	0.7%	12.7%	8.2%

A between-farm comparison of *E. coli* isolates resistant to various combinations of the antimicrobials showed a higher level of florfenicol and florfenicol/ampicillin resistance on Farm A while Farm B had more resistance to gentamicin, ampicillin and gentamicin/ampicillin (data not shown). A different resistance profile between farms probably reflected the different antimicrobial usage patterns and demonstrates the importance of the dual survey method in assessing both declared and actual usage of antimicrobials on-farm.

Supported by Australian Pork Limited and International Animal Health Products Pty Ltd.

## Lactation performance of sows after enhancing their gut microflora through a dietary nutraceutical

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Biomim<sup>®</sup> PEP is a phytogetic feed additive based on the combination of essential oils, dried plant extracts and fructo-oligosaccharides and is designed to improve feed palatability and digestion. The phytogetic compounds have a pronounced effect on the gut microflora and digestive processes of the sow. We hypothesized that Biomim<sup>®</sup> PEP would increase feed intake of sows during lactation, leading to an increased weaning weight. As Biomim<sup>®</sup> PEP stimulates beneficial bacteria, which may reduce pathogen, we also expected pre-weaning mortality to be lower.

One hundred and twenty Large White x Landrace sows of mixed parity were housed in farrowing crates and allocated to two treatments at the start of the experimental period, with 30 sows selected per treatment per week for two weeks. Treatments were assigned as Control lactation diets (CL) and Control + 0.2% Nutraceutical (PEP). All treatments contained 100 ppm monensin sodium and contained 14 MJ DE/kg and 0.9 g lysine. Sows were moved into the farrowing house on day 8.5±4 (mean±SD) and fed 2.7 kg/day of their treatment diet. The day after farrowing, all sows were offered their treatment diet to appetite for the remainder of lactation. Sow feed intakes were recorded from farrowing to weaning. The number of piglets born and born live was recorded for each sow. Litters requiring fostering were weighed and fostered on day one within treatment groups. Litter size and weight at weaning, treatment and mortalities were recorded. The average weaning age was 24 days for the CL and 25 days for the PEP treated animals. Six sows and their litters from each treatment were removed from the analysis due to production requirements. Performance data were analyzed using General Linear Model analysis of variance (SPSS Inc v14).

There was no difference in the farrowing results between the treatments, which was expected given the short period of time sows were supplemented with Biomim<sup>®</sup> PEP before farrowing (Table 1). While sows supplemented with Biomim<sup>®</sup> PEP had a tendency to have litters of a higher weaning weight, which was significant for sows greater than parity 3 (P=0.013), feed intake did not increase as expected. In addition there was no difference in the pre-weaning mortality after day two or the litter size weaned. Although monensin sodium was used in both diets and is known for its positive influence on gut health and performance, differences in growth performance were numerically better when sows were fed Biomim<sup>®</sup> PEP. Presumably, this effect is due to increased lactation performance of sows. We expect that differences in growth performance and piglet mortality could have been significant without antibiotics.

**Table 1. Performance data for sows fed either a control diet or the control diet supplemented with Biomim<sup>®</sup> PEP at 2kg/t for the entire lactation period**

	Litter size Day 2	Average weight (kg)	Litter size weaned	Av. Wean weight (kg) <sup>1</sup>	Weight gain (kg/day) <sup>1</sup>	Average sow intake (kg/day)
CL <sup>2</sup>	11.0	1.6	9.0	6.8	0.219	5.2
PEP <sup>3</sup>	11.3	1.6	9.1	7.3	0.231	5.2
P-value <sup>4</sup>	NS	NS	NS	NS	NS	NS
SEM	0.106	0.026	0.169	0.125	0.004	0.054

<sup>1</sup>Wean age taken as a covariant in wean weight and rate of gain analysis; <sup>2</sup>Control diet (no Biomim PEP); <sup>3</sup>PEP – Control diet with Biomim PEP;

<sup>4</sup>NS not significant

## Association between lactation, feed intake and lifetime reproductive performance of sows

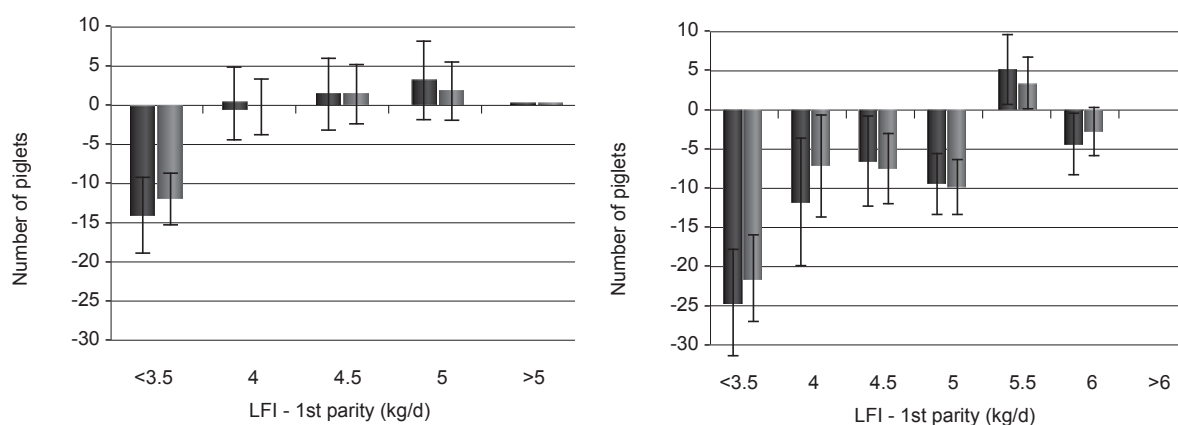
S. Hermesch and R.M. Jones

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Low feed intake during lactation (LFI) has been associated with impaired sow performance for several reproductive traits (Eissen *et al.*, 1999). In this study we aimed to establish whether there was an association between LFI and sow lifetime performance.

We compared the lifetime performance of 250 gilts and 202 second-parity sows with data on their LFI and total number of piglets born (LNBA) and weaned (LWEAN). The mean LFI was 4.3 and 5.4 kg/day-1 for the first and second parity. Sows had their first litter between May 2002 and April 2004 allowing them to complete eight parities by November 2006. The model for LNBA and LWEAN, developed using Proc GLM (SAS Inc v.9.2), included sow breed (three levels) and LFI grouped in 0.5 kg/day classes, ranging from less than 3.5 kg/day to above 5 kg/day for gilts and above 6 kg/day for second parity sows. About one fifth (19.2%) of gilts had a LFI below 3.5 kg/day. The first four LFI classes (<5.0 kg/day) represented 4.5, 3.0, 7.4 and 17.3% of second parity sows, respectively.

Solutions for individual LFI classes were expressed relative to the class with the highest LFI (Figure 1). Lifetime performance of sows was 14.2 (LNBA) and 11.7 (LWEAN) piglets lower for gilts with a LFI below 3.5 kg/day compared to the highest LFI class. There were no significant differences between LFI classes for LNBA or LWEAN once gilts ate more than 3.5 kg/day. A LFI below 3.5 kg/day in the second parity reduced LNBA and LWEAN by 24.7 and 21.5 piglets compared to the highest LFI class. The trend of impaired lifetime performance continued until 5 kg/day LFI in the second parity. No information was found in the literature about these associations. However, Williams (1998) outlined how the modern sow genotype is more susceptible to under nutrition during lactation, which subsequently can affect reproductive performance. About 20% of first and 30% of second parity sows had reduced lifetime performance due to low LFI, highlighting the need to increase LFI in sows.



**Figure 1.** Solutions for total number of piglets born alive (first column) and weaned (second column) over sow lifetime and lactation feed intake (LFI) during the first and second parity

T. and S. Neuendorf provided the data. Analyses were funded by Australian Pork Limited project 2133. \*AGBU is a joint venture of NSW Department of Primary Industries and the University of New England.

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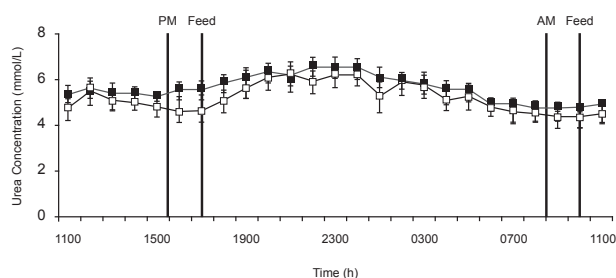
## The effect of feeding level on growth, plasma non-esterified fatty acid and urea levels in finisher pigs

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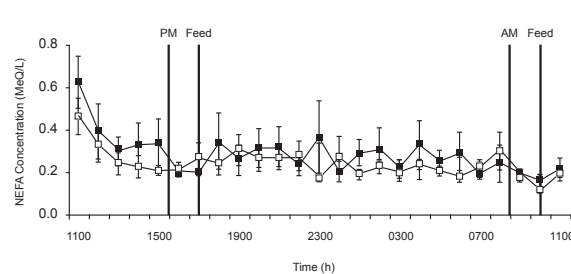
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Scrimgeour *et al.* (2006) found increased basal concentrations of insulin in pigs fed *ad libitum* compared to a similar quantity of feed offered in two time periods per day (0900-1000h and 1600-1700h). Interval feeding may therefore be associated with improved insulin sensitivity expressed as an improvement in feed:gain and decreased carcass fatness. However in our previous study the treatments were applied sequentially using the same animals and were therefore confounded with time. The objective of this study was to compare interval and *ad libitum* feeding in similar animals at the same time.

Twenty male hybrid (mainly Large White x Landrace) pigs were allocated to individual grower pens at  $70 \pm 4.58$  kg (mean  $\pm$  SD) live weight. The pigs were maintained at  $23 \pm 1^\circ\text{C}$  and a 12 hour light period (0600–1800 h) in two air spaces with two rooms per air space. Pigs were fed a commercial grower diet estimated to contain 10 g available lysine and 13.5 MJ digestible energy per kg. Each pig was fed either *ad libitum* (control) or a similar amount offered in two 90 minute feeding periods (0900-1030 h and 1600-1730 h) per day. Each feeding regime was replicated in separate rooms in both air spaces. Feed was offered to maintain about 2 kg in each trough and residues were recorded daily while body weights were recorded weekly. Water was provided using nipple drinkers. Pigs were entrained to these two feeding schedules for 28 days. On day 29, cannulae were inserted into the external jugular vein of each pig via the ear vein. Serial blood samples were collected hourly for 24 hours from day 30. Plasma urea (Figure 1) and non-esterified fatty acids (NEFA) (Figure 2) concentrations were determined enzymatically as indices of lipid and protein metabolism respectively. A linear mixed model was fitted to the data using a REML procedure in Genstat.



**Figure 1.** Plasma urea concentrations in finisher pigs fed *ad libitum* (□) or *phasic* (○) regimes



**Figure 2.** Plasma NEFA concentrations in finisher pigs fed *ad libitum* (□) or *phasic* (○) regimes

There was no evidence found in this study to support the hypothesis that interval feeding improves live performance or alters fat or protein metabolism. No significant differences were found in daily gain ( $1.38 \pm 0.13$  kg and  $1.39 \pm 0.13$  kg (mean  $\pm$  SD)), daily feed intake ( $3.33 \pm 0.39$  kg and  $3.14 \pm 0.72$  kg) or feed:gain ( $2.44 \pm 0.13$  kg and  $2.32 \pm 0.10$  kg) for *ad libitum* and interval-fed animals respectively. There were no significant differences in circulating plasma urea and NEFA concentrations when interval-fed animals were compared to pigs fed *ad libitum*. This result suggests that pigs fed at discrete intervals daily are able to adapt to this feeding pattern and consume similar quantities of feed over three hours to that offered to animals fed *ad libitum* over 24 hours. Body fat and protein metabolism appear to be unaffected by feeding regime. However, a measure of insulin sensitivity will be important in interpreting these data.

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## Low levels of copper and zinc proteinates maintain a normal mineral status in growing and finishing pigs

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A significant reduction in the levels of copper (Cu) (90%) and zinc (Zn) (40%) in the faeces of growing pigs was achieved without affecting pig growth when the inclusion level (IL) of Cu in the diet decreased from 50 ppm to 0 ppm Cu, and Zn from 80 to 40 ppm, both in the Bioplex® form (Hernández *et al.*, 2007). However it is important to establish if these low mineral levels enabled a normal mineral status to be maintained in the pigs. During digestion, minerals interact with each other and also with digesta components (e.g. phytate), which reduces the amount of each mineral that is absorbed. However it is likely that such interaction is less when minerals are supplied in the organic form due to the protection offered by the amino acid or peptides to which the mineral is chelated during manufacturing (Fairweather-Tait, 1996). In this study we examined the effect of feeding increasing IL of Cu together with low (treatments 1-4) or high (treatments 5-8) IL of Zn in the Bioplex® form on the status of biochemical markers of Cu, Zn and Fe in growing pigs.

The experiment was designed as a 2x4 factorial arrangement of treatments, with the respective factors being two IL of Bioplex® Zn (40 and 80 ppm) and four of Bioplex® Cu (0, 10, 30 and 50 ppm). A control treatment provided sulphates at levels of Cu and Zn similar to the high Bioplex® treatment. The study used 216 Large White x Landrace pigs from 25-107 kg live weight (LW) housed in three pens of eight pigs/treatment. Pigs were fed *ad libitum*. Blood samples were collected from the same random sub-sample of four pigs/pen on days zero, 21 and 49 of the experiment while samples of liver and bone were collected at slaughter (minimum of 104 kg LW). Analysis of variance, using the pig as a unit, was used to examine the main effects and all interactions on Cu, Zn and Fe in plasma and haemoglobin (Hb) and Cu content of red blood cells (RBC Cu) and Cu, Zn and Fe levels in liver and Zn in bone. Blood samples collected on days 21 and 49 were analyzed using repeated measures analysis of variance with blood levels on day zero as covariates.

Haemoglobin and plasma Fe levels were similar between treatments ( $P>0.05$ ), while Cu ( $P=0.006$ ) and Zn ( $P=0.011$ ) levels increased with the IL of the minerals in the diet. Copper content in RBC decreased as the IL of Zn in the diet increased ( $P=0.006$ ) (Figure 1). Hepatic Cu also increased with Cu IL ( $P=0.003$ ), but at 0 ppm IL it was at the lower end of what is considered normal (Figure 2). The concentration of Zn in bone was within the normal range and decreased as the IL of Cu increased from 0 to 50 ppm ( $P=0.043$ ). The results indicated that there were interactions between Cu and Zn at the levels studied. Although Hb and plasma Fe levels were within the normal range in pigs fed the 0 ppm IL Cu diet, storage of Cu in liver approached marginal levels. Since Cu is involved in Fe transport, a grower diet supplemented with at least 10 ppm Cu and 40 ppm Zn would be a safer recommendation.

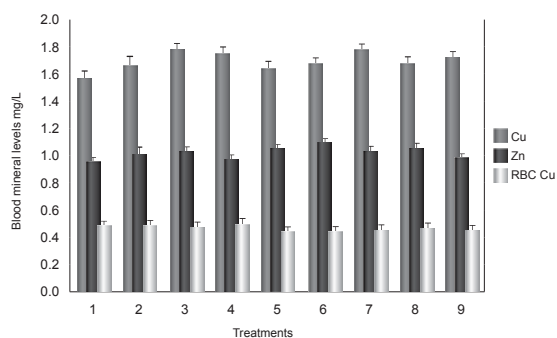


Figure 1. Blood mineral levels ( $\pm$ SEM)

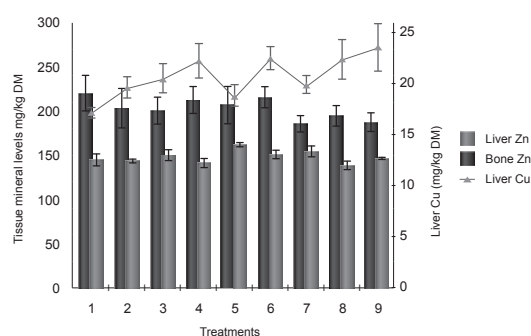


Figure 2. Tissue mineral levels ( $\pm$ SEM)

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## Effect of graded dietary tryptophan levels on [1-<sup>13</sup>C]leucine oxidation and nitrogen balance in growing pigs

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Tryptophan (Trp) is an indispensable amino acid (AA) that is incorporated into body proteins as muscle, but also in functional proteins like serotonin. Low dietary levels can limit animal performance and metabolism. Dietary Trp is known to be a limiting factor of serotonin synthesis. In this pilot study we aimed to estimate the animal requirements of Trp based on the three different methodologies: serum serotonin, nitrogen (N) balance and Indicator AA Oxidation (IAAO). Since IAAO and protein deposition are inversely correlated with the dietary concentration of a limiting AA, the oxidation of [1-<sup>13</sup>C]leucine as an indicator of the protein synthesis level and the N deposition were measured and compared in the pigs.

Six groups of pigs were fed six equivalent diets based on wheat, corn and soy bean meal with graded concentrations of standardized ileal digestible (SID) Trp (1.33, 1.51, 1.67, 1.82, 2.00, 2.16 g/kg feed, 88% DM) and 10.31 g/kg feed SID lysine for a period of three weeks beginning at 54 days of age (DOA). The daily feed intake was increased up to 85 g DM/kg<sup>0.75</sup> BW and was kept constant during the last 10 days of the experiment. The mean intercalated BW±SD of total 41 castrated male German Landrace pigs was 24.8±2.8 to 26.8±3.1 kg (unequal class allocation). In the tracer study 22 pigs were continuously fed from 1200 hours via half-hourly small meals over five hours. At 67, 68 and 69 DOA a 10-hour primed continuous i.v. infusion of [1-<sup>13</sup>C]leucine in 0.9% NaCl solution was started at 0800 hours (prime: 5.25 µmol/kg BW, infusion rate: 3.5 µmol/kg BW\*h) via a jugular catheter. Plasma was sampled at 1500 hours during the fed steady state to determine α-[1-<sup>13</sup>C]ketoisocaproic acid enrichment, which is considered to represent the enrichment of the intracellular leucine pool (Matthews *et al.*, 1982). At 71 DOA the serotonin concentration was analyzed in fasted serum. The N balance of the pigs was determined between 70 to 76 DOA.

Serum serotonin levels increased linearly from 600 to 1000 µg/L, parallel to the increase of Trp intake per kg<sup>0.75</sup>BW/day ( $y=3.14x + 264$ ,  $r^2=0.114$ ). The N deposition increased linearly from 1200 to 1550 mg/kg<sup>0.75</sup>BW ( $y=2.72x + 887$ ; slope  $P=0.0002$ ). Using a two-phase linear regression crossover analysis according to Robbins *et al.*, (1979) the N deposition intersected at 211 mg Trp/kg<sup>0.75</sup>BW intake and ≈ 1480 mg deposited N/kg<sup>0.75</sup>BW. This intersection point corresponded to a daily intake of 2.13 g SID Trp and to a concentration of 2.16 g SID Trp/kg feed and resulted in a Trp:Lys ratio of 0.21. The <sup>13</sup>C-leucine oxidation rate linearly decreased from 3 to 2 mmol/hour ( $y=-1.09x + 5.18$ ;  $r^2=0.116$ ; slope  $P=0.120$ ) but no inflection point could be calculated. The results suggest that the Trp need of the experimental pigs was about the highest Trp concentration tested.

The results also indicate that the Trp minimal need corresponded to a SID Trp:Lys ratio of 0.21. Additional Trp concentration levels higher than 2.45 g SID Trp/kg DM should be tested to allow reliable intersection point calculations and to validate our results.

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## Straw as bedding or in feed increases unsaturated fats in belly fat

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Pigs with access to straw either as bedding and/or via their diet deposit less fat in their belly than pigs without access to straw (Trezona *et al.*, 2007). There are at least two possible mechanisms for this. First, if pigs consume straw in addition to their food the energy density of their total intake may be less and this may depress their voluntary energy intake and lower the deposition of fat. Second, if pigs lie on straw that insulates them better they may respond by depositing less fat in the areas that are in contact with the ground. If less fat is deposited because endogenous synthesis is reduced then it is likely that the fatty acid (FA) profile of the fat will change and become less saturated.

Ninety-six Large White x Landrace female pigs were stratified by live weight ( $16.1 \pm 0.26$  kg) at eight weeks of age into groups of six and housed in commercial grower-finisher pens within a naturally ventilated shed. The experiment was a 2x2 factorial design with two dietary treatments, i) CD (grower and finisher diets) and ii) SD (grower-finisher diets fortified with 10% wheat straw) (calculated analyses were as reported by Trezona *et al.*, 2007), and two floor treatments: i) CF: partially-slatted concrete floor and ii) SF: straw bedding as flooring (~15 cm thick). At 24 weeks of age pigs were slaughtered at a commercial abattoir. Belly fat was collected at the ventral midline from the hot carcass of 12 pigs per treatment and stored at  $-80^{\circ}\text{C}$  until FA profiles were determined via gas chromatography. Data were analysed by two-way ANOVA (Genstat v8).

**Table 1. Effect of straw, as bedding and in the diet, on the fatty acid profile of belly fat in 24-week-old gilts (see text for explanation of treatment acronyms)**

Fatty acid (%)	SD-CF	SD-SF	CD-CF	CD-SF	SEMa	P		
						Diet	Floor	D*F
Myristic (C14:0)	2.0 <sup>b</sup>	1.9 <sup>a</sup>	1.9 <sup>a</sup>	2.0 <sup>b</sup>	0.04	0.949	0.693	0.049
Palmitic (C16:0)	25.0	24.4	24.8	24.8	0.31	0.796	0.374	0.424
Palmitoleic (C16:1)	3.0	3.1	3.0	3.1	0.11	0.689	0.283	0.946
Stearic (C18:0)	14.0	13.0	14.2	13.5	0.43	0.444	0.062	0.766
Oleic (C18:1)	31.6 <sup>a</sup>	32.5 <sup>b</sup>	32.8 <sup>b</sup>	33.0 <sup>b</sup>	0.30	0.007	0.063	0.252
Linoleic (C18:2)	18.1 <sup>bc</sup>	18.7 <sup>c</sup>	17.2 <sup>a</sup>	17.5 <sup>ab</sup>	0.46	0.029	0.362	0.819
□-Linolenic (C18:3 n3)	1.9 <sup>ab</sup>	2.0 <sup>b</sup>	1.8 <sup>a</sup>	1.8 <sup>a</sup>	0.05	0.049	0.416	0.863
□-Linolenic (C18:3n6)	0.078	0.071	0.066	0.066	0.005	0.086	0.486	0.486
Saturated:Unsaturated	0.734	0.685	0.733	0.710	0.0181	0.520	0.055	0.485

<sup>a</sup>SEM = pooled standard error of mean

The lower percentage of stearic acid ( $P = 0.06$ ), the higher percentage of oleic acid ( $P = 0.06$ ) and the lower ratio of total saturated to unsaturated FA ( $P = 0.05$ ) all suggest that pigs bedded on straw deposit more unsaturated FA. When pigs were given straw mixed into their diet unsaturated FA in the belly also increased as shown by the higher levels of oleic ( $P = 0.007$ ), linoleic ( $P = 0.03$ ) and linolenic acids ( $P = 0.05$ ). The finding that straw provided either as bedding or consumed directly by pigs changes the FA profile in belly fat is important for the Australian pork export market because in Asia the belly is a premium cut. While increasing the levels of unsaturated fat in the belly may help in the advertising of healthy pork, it may also be a disadvantage because higher levels of unsaturated FA are associated with softer fat that can be more prone to off flavours because of rancidity.

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# 10

## Feed ingredients and milling

## Grain and plant protein types fed to weaned piglets influence the apparent digestibility of carbohydrates and crude protein when measured at the terminal ileum

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Diets based on cooked white rice fed to weaned piglets have a higher apparent ileal digestibility of starch than diets based on wheat (Pluske *et al.*, 2007). The diets based on cooked white rice have used predominately animal sources of protein, however in Europe these are banned or excluded by retailer's specifications (except for milk proteins), and plant proteins are widely used instead. This study examined the interactive effects of cereal types and plant protein types on the apparent ileal digestibility of protein and carbohydrates to test the proposition that suitable sources of plant protein could ensure high digestibility coefficients in the small intestine.

Sixteen post-weaned pigs were surgically fitted with a post-valve T-caecum cannula on day 12 after weaning and allowed to recover in individual cages. Four non-medicated diets based on ingredients available in France were examined in a 2x2 factorial arrangement to assess the apparent ileal digestibility (AID) of carbohydrates and crude protein (CP). Diets differed according to grain source (GS) - extruded rice in a wheat and barley-based (RWB) diet compared to wheat and barley alone (WB) and protein sources (PS) - diversified refined protein concentrates from soybean, potato and fishmeal (DIV<sub>prot</sub>) compared to soybean meal and full-fat extruded soybeans (SB<sub>prot</sub>). Digesta collection commenced 12 days after surgery. Diets were fed for a four day adaptation period followed by a three day collection period for 12 hours per day. Diets included 0.2% chromic oxide as an indigestible marker. Total carbohydrate (CHO) was calculated by subtracting CP and fat to organic matter. Data were first analyzed including the effect of diet, series of collection and their interaction. As the latter was not significant, data were reanalyzed using repeated measures analysis of variance with GS and PS as main effects, and their interaction.

There was a trend (P=0.07) for the AID of CP to be higher in pigs fed DIV<sub>prot</sub> than SB<sub>prot</sub> (Table 1). Both the GS and PS influenced the AID of CHO (P<0.001). Pigs fed extruded rice at the expense of some wheat and barley (diet RWB) showed a higher (P<0.001) AID of CHO, and the use of DIV<sub>prot</sub> rather than SB<sub>prot</sub> showed a higher AID (P<0.001). Pigs fed the DIV<sub>prot</sub> showed a greater AID of sugars (P=0.07). Our data show that grain types and plant protein sources fed to weaned piglets alter the flow of CHO and CP into the large intestine. This, in turn, might have ramifications for the expression of post-weaning colibacillosis in diets not containing antimicrobials (Heo *et al.*, 2007).

**Table 1. Influence of protein source (PS) and grain source (GS) on apparent ileal digestibility (AID) of crude protein (CP) and total carbohydrates (CHO) after weaning**

PS	DIV <sub>prot</sub>		SB <sub>prot</sub>		RSD <sup>1</sup>	Significance <sup>2</sup>
	RWB	WB	RWB	WB		
Piglet BW, kg	14.0	13.5	14.5	13.7	2.0	-
AID CP, %	81.2	80.9	78.0	77.1	5.6	PS (P=0.07)
AID CHO, %	81.1	76.5	75.9	70.3	2.2	GS***, PS***
AID starch, %	99.7	99.4	99.5	99.3	2.3	GS (P=0.07), PS (P=0.14)
AID sugars, %	71.6	72.2	56.1	53.8	7.7	PS***

<sup>1</sup>RSD: residual standard deviation; <sup>2</sup>\*\*\*P<0.001.

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## Sorghum type significantly influences weaner pig growth performance

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In Australia about 35% of growing pigs are fed diets containing sorghum, however they do not perform as consistently as those on wheat-based diets. Although the general nutritional value of sorghum is known the nutrient composition of the feed can vary widely due to differences in cultivar, locality and plant nutrition (Kondos and Foale, 1983). We tested the hypothesis that voluntary food intake and growth of weaner pigs would vary when offered diets composed of different sorghum cultivars or the same variety sourced from different regions.

The experimental weaner diets contained 65% of one of the test sorghum grains and highly digestible raw ingredients. The diets were formulated to contain 14.5 MJ DE/kg and available lysine content of 0.85 g/MJ DE. The 10 sorghum grains were used in a random block design consisting of 200 (10 grains x 20 pigs) individual male weaner pigs (QAF genotype). Initial pig weights were statistically the same (range 7.85-8.17 kg). Pigs were placed in individual crates for 28 days. After a pre-treatment of five days on a common nursery diet, pigs were fed the test diets and water *ad libitum* for a further 21 days. Body weight and feed intake were measured on days 0 and 14 after the initial five-day period. Initial pig weight was used as a co-variant in the analyses.

Sorghum variety produced a significant effect on FCR ( $P=0.004$ ) and daily gain ( $P=0.021$ ) (Table 1). There was no effect of sorghum on feed intake ( $P=0.664$ ). Overall there was a difference between the highest and lowest FCR and daily gain of 17.4% and 18.2% respectively. Interestingly, the best performing sorghum was Liberty, which is the only white/yellow sorghum represented. The two varieties of Buster from different areas (QLD and NSW) generated a 10.5% difference ( $P<0.15$ ) in daily gain. Further research on causes of the variability is warranted.

**Table 1. Effects of 10 sorghum varieties on the growth performance of male weaner pigs**

Sorghum variety	21-day weight	Daily Gain (kg/d)	FCR	Feed intake (kg/d)
MR Maxi	16.00	0.383 <sup>ab</sup>	1.32 <sup>abc</sup>	0.501
Liberty	16.11	0.390 <sup>a</sup>	1.28 <sup>c</sup>	0.498
Pacer	16.46	0.396 <sup>a</sup>	1.30 <sup>bc</sup>	0.519
MR43	15.36	0.342 <sup>ab</sup>	1.37 <sup>bc</sup>	0.474
Buster (QLD)	15.15	0.341 <sup>ab</sup>	1.40 <sup>abc</sup>	0.471
Allora Grain	15.37	0.334 <sup>ab</sup>	1.46 <sup>ab</sup>	0.481
Venture	15.77	0.367 <sup>ab</sup>	1.46 <sup>ab</sup>	0.516
Buster (NSW)	15.87	0.381 <sup>ab</sup>	1.31 <sup>bc</sup>	0.505
Red 1	14.87	0.324 <sup>b</sup>	1.55 <sup>a</sup>	0.481
Red 2	16.00	0.388 <sup>ab</sup>	1.31 <sup>bc</sup>	0.528
P value	0.021	0.035	0.004	0.664

<sup>abc</sup> Values in a column with different superscripts are significantly different ( $P<0.05$ ).

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## Evaluation of mandelup lupin (*Lupinus angustifolius* L.) at different inclusion levels and in response to enzyme supplementation for grower/finisher pigs

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Despite lupins (*Lupinus angustifolius*) being an economical plant protein ingredient to feed growing and finishing pigs in Australia, the presence of anti-nutritional factors such as oligosaccharides and non-starch polysaccharides has generally restricted high inclusion levels in diets. New cultivars of Australian sweet lupins (ASL) have been released in Western Australia, but these have been bred mainly for improved disease resistance, drought resistance and yield. While these breeding criteria may also have changed nutritional characteristics, no research has investigated the nutritional adequacy of recent cultivars of ASL when incorporated at high levels in diets for grower/finisher pigs. The aims of this experiment were to 1) evaluate the optimum inclusion level for grower/finisher pigs of the current major variety of ASL (cv. Mandelup) and 2) examine the effect of a multi-enzyme preparation on the performance of grower and finisher pigs fed the lupin-based diets. The enzyme treatment was included to assess any potential negative effects of feeding high levels of Mandelup to pigs.

Two hundred and twenty-four (Large White x Landrace, initial body weight 27.2 kg±0.22) male pigs were used in a completely randomized block design having eight experimental treatments, with 28 pigs (seven pigs/pen) allocated to each. The experiment was a 4x2 factorial design with the respective factors being lupin inclusion level (200, 250, 300 and 350 g/kg) and multi-enzyme supplementation (±200 g/tonne Allzyme® SSF, Alltech Biotechnology Pty Ltd). The enzyme contained minimum activities of 100 U 1,4- $\alpha$ -xylanase, 30 U  $\alpha$ -amylase, 200 U  $\alpha$ -glucanase, 700 U protease, 40 U cellulase, 4000 U pectinase, and 300 U phytase per g product. All diets were formulated to contain equal amounts of ileal digestible amino acids and the same ileal digestible lysine to DE ratio. Lupins were progressively substituted for soybean meal based on equivalent ileal digestible amino acid contents between test diets. Pigs were fed grower, finisher and pre-sale diets between 27-50 kg, 50-75 kg and 75-107 kg, respectively. The GLM procedure of SPSS (SPSS Inc) was used for statistical evaluation of the results with pen as the experimental unit.

**Table 1. Effects of lupin inclusion level (L) and multi-enzyme supplementation (E) on the performance of grower/finisher pigs and days to slaughter (DTS) between 27 to 107 kg**

	Lupin inclusion (g/kg)				Enzyme		SEM	Main effects			Lupin effects	
	200	250	300	350	-	+		L	E	LxE	Linear	Quadratic
ADG (kg/day)	1.00	1.01	1.01	1.03	1.01	1.01	0.008	0.694	0.602	0.791	0.266	0.892
VFI (kg/day)	2.72	2.74	2.76	2.68	2.69	2.76	0.020	0.505	0.079	0.322	0.484	0.237
FCR (kg/kg)	2.73	2.71	2.74	2.62	2.68	2.72	0.027	0.459	0.447	0.751	0.221	0.379
DTS (days)	77.8	76.7	77.4	76.3	77.4	76.6	0.65	0.871	0.588	0.883	0.535	0.964

Including up to 350 g/kg lupin seed in the grower, finisher and pre-sale diets did not depress growth, feed intake or feed conversion ratio (FCR). Diets formulated to contain 350 g/kg lupins with adjustment of ileal digestible essential amino acids supported daily gains of over 1 kg with 2.7 kg/kg FCR between 27 kg to 107 kg live weight. Consequently, days to reach 107 kg were similar regardless of the lupin concentration in the experimental diets. Supplementation of the multi-enzyme preparation did not improve production, although supplementation of the enzyme tended to increase feed intake without increasing growth rate during the pre-sale period (3.44 vs. 3.28, P=0.054). The result suggests that the current variety (Mandelup) of lupin can be used in grower and finisher diets up to 350 g/kg without compromising growth of pigs.

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## Digestibility of energy and amino acids in pearl millet hybrids fed to growing pigs

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Pearl millet (*Pennisetum glaucum*) hybrids developed by the Queensland Department of Primary Industries and Fisheries have relatively high metabolizable energy and protein values (poultry studies), tolerate heat and drought and generally out-yield sorghum in areas of low rainfall and lighter soils (Singh, 2005). While some studies have examined the value of the grain for poultry, no research has been done on the grain's value in pig diets. In this study we hypothesized that the nutritional value of pearl millet would be superior to that of sorghum in pig diets. We determined the ileal digestibility of energy and amino acids in pig diets based on two hybrids of pearl millet and compared these to sorghum and two standard feed wheat grains. The cereals were the sole sources of protein in the diets. Five male Large White grower pigs (35-40 kg live weight) were used in a 5x5 Latin Square experimental design.

**Table 1. Mean ileal digestibility values of two pearl millet- (PM), one sorghum- (S) and two wheat-based (W) diets fed to grower Large White pigs**

	Diet						± s.e.d.	P
	PM 1	PM 2	S	W 1	W 2			
Gross energy (GE)	0.83	0.82	0.80	0.76	0.76	0.015	< 0.001	
Nitrogen (N)	0.79	0.74	0.73	0.85	0.83	0.027	< 0.001	
Fat	0.73	0.78	0.58	0.61	0.67	0.086	0.163	
Starch	0.97	0.98	0.95	0.97	0.98	0.013	0.253	
Histidine	0.86	0.83	0.76	0.89	0.87	0.014	< 0.001	
Isoleucine	0.89	0.87	0.85	0.89	0.86	0.014	0.040	
Leucine	0.91	0.89	0.89	0.90	0.87	0.012	0.066	
Valine	0.88	0.85	0.83	0.86	0.83	0.014	0.016	
Lysine	0.88	0.87	0.82	0.87	0.84	0.021	0.057	
Methionine	0.93	0.92	0.88	0.91	0.88	0.013	0.008	
Threonine	0.80	0.76	0.76	0.81	0.79	0.024	0.100	
Phenylalanine	0.90	0.88	0.88	0.92	0.89	0.011	0.014	
Tryptophan	0.65	0.61	0.74	0.79	0.76	0.027	< 0.001	

The average gross energy (GE) content of the pearl millet diets (17.5 MJ/kg) was higher than the sorghum- (17.1 MJ/kg) and wheat-based (17.3 MJ/kg) diets, due to the higher average fat content in the millet (3% fat) than the sorghum and wheat ( $\leq 1\%$ ) diets (Table 1). Values of gross energy digestibility for the millets were higher than for the other grains. The millets had higher nitrogen (N) digestibility than sorghum but lower than that of wheat. Within diets, most essential amino acids (EAA) had digestibility values of  $\geq 80\%$  except threonine and tryptophan. Most EAA digestibility values were higher in the millet diets, particularly PM 1, than in the sorghum diets, except for tryptophan. Nutritionally, the millet based diets appear to be superior to the sorghum based diet. Of the two millet hybrids, PM1 would be better nutritionally.

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## Evaluation of pearl millet as a pig feed ingredient

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Pearl millet (*Pennisetum glaucum*) (PM) is a food grain grown across the semi-arid tropics of Africa and Asia. It is more drought and heat tolerant than sorghum and will out-yield sorghum in areas with low rainfall and lighter soils. The new short-season pearl millet hybrids developed in Australia mature in 85 days, which is significantly earlier than current sorghum hybrids (Lawrence *et al.*, 2007). It offers potential for greater use in Australia's livestock industries as an energy source.

We evaluated the inclusion level of two PM hybrids (PM1 and PM2) in diets of male grower (20-50 kg) and finisher pigs (50-90 kg). The control diet contained 60% sorghum (no PM), with the sorghum component replaced by either PM1 or PM2 at 25, 50, or 100% in the treatment diets, giving a total of seven treatments. Thirty-five individually housed Large White male pigs were used in a completely randomized block design. Diets were formulated to 14 and 13 MJ DE/kg and 0.63 g and 0.55 g available lysine/MJ DE for growers and finishers respectively. Pearl millet nutrient composition was based on the same grain batches as used in the digestibility experiment reported by Teleni *et al.*, (2007).

Results indicated that there were no significant differences between the diets in terms of ADG, DFI and FCR in grower and finisher phases and the P2 measurement taken at 90 kg liveweight (Table 1). Pigs on PM2 diets appeared to perform better than those on PM1, especially in the finisher phase. The costs of the PM-based diets were much lower than sorghum diet (\$297, \$281 and \$275 for grower diets and \$265, \$262 and \$256 for finisher diets respectively for sorghum, and PM1 100 and PM2 100 diets) based on sorghum and PM costs being equal. Pearl millet may be an acceptable substitute to sorghum and does have potential for superior feed cost advantage.

**Table 1. Effect of graded levels of pearl millet (PM) on the average daily gain (ADG), daily feed intake (DFI), feed conversion ratio (FCR) and P2 backfat of pigs growing from 20-90 kg**

Diets	Grower 20-50kg			Finisher 50-90kg			P2 backfat (mm)
	ADG (kg/day)	DFI (kg/day)	FCR	ADG (kg/day)	DFI (kg/day)	FCR (feed/gain)	
Control	1.022	2.063	1.934	1.123	2.708	2.127	11.6
PM1 25	1.033	1.971	1.956	1.073	2.577	2.120	11.0
PM1 50	1.033	1.944	1.993	1.011	2.557	2.246	11.6
PM1 100	1.043	1.963	1.981	1.066	2.620	2.157	11.0
PM2 25	1.109	2.185	2.090	1.210	2.902	2.103	12.8
PM2 50	1.007	1.921	2.050	1.103	2.684	2.092	11.3
PM2 100	1.090	2.125	2.054	1.206	2.837	2.065	11.2
LSD (0.05)	0.1214	0.2934	0.1752	0.2425	0.4352	0.2401	2.23

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## Symposium - current issues in feed milling

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### Introduction

Over the past 20 years the Australasian Pig Science Association (APSA) conference has grown to be recognized internationally as one of the premier world pig events. As such, the proceedings of the APSA conference can be used as a reliable indicator of the development of our understanding of many subject areas, not the least being nutrition. There have been many valuable APSA contributions to the understanding of what nutrients are needed by pigs, how nutrients can be supplied to pigs from different feedstuffs, how pigs will respond to these nutrients and approaches to modelling and modulating the responses. Successful application of nutritional knowledge depends on pigs actually consuming the nutrients in the correct amounts, and APSA has also provided many contributions to the literature to assist better industry understanding of optimal feeder design, stocking density, water management and other factors influencing feed intake. However, one topic that has not been explored in any detail in the past 20 years is the role that feed manufacturing plays in pork production. In simple terms, having the appropriate shed and feeder design and the correctly formulated diet utilizing all the latest digestibility and intake enhancers is of little benefit if the ration has not been adequately and appropriately prepared. The present symposium seeks to address some of the current issues facing feed manufacturers and to show how better managing these issues could reduce feed production costs and improve animal performance and food and feed safety.

The first paper in this symposium (Behnke, 2007) identifies the issues surrounding cross transference of raw materials in the feed production process. Cross transference of compounds such as medications or restricted animal materials like meat meal into non-intended feeds is a worldwide issue with very few countries being incident free. As seen with the well-publicized fire retardant and dioxin issues in recent years, there can be devastating effects on the meat industry if food and feed safety are compromised. Behnke (2007) also explores the methodology behind mixing accuracy. While the on-farm and commercial mixers generally assume that mixing is a precise process, how good is it really and what would the consequences be of getting it only a 'little bit wrong?'

The majority of commentators on global trends have indicated that the demand for crop products such as grain and oilseeds for human consumption and/or biofuel production is expected to increase significantly through to 2020 and beyond. Unless there is a marked shift in the profitability of the livestock industries, these industries will be unable to compete for raw materials and will become increasingly reliant on fractionated or co-products. The second paper in the symposium (Zilstra and Beltranena, 2007) discusses some of the opportunities the feed industry has in using these fractionated products and possible ramifications for the feed manufacturing sector.

The third paper in the symposium (Gannon, 2007) picks up on the themes of variability in, and variety of, raw materials that can be used by the Australian pork industry. Recent research programs have identified some of the characteristics of raw materials that impact on animal performance and this paper discusses how these characteristics can also impact on feed manufacturing. Some of the 'behind the scenes' activities of feed manufacturers are revealed and a number of opportunities for improving the efficiency of feed manufacturing, and therefore reducing costs, are identified.

# Issues in feed manufacturing: a US perspective

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## Abstract

Feed manufacturers around the world face many unique local challenges. However, issues of cross transference or ingredient carryover and assessing nutrient homogeneity seem to be common around the globe. This paper describes the risks associated with cross transference and then identifies the causes of carryover. Also identified are the processes and procedures involved in sequencing, flushing and equipment clean-out that can be used to reduce cross transference and the management and operational aspects that need to be addressed. The second part of the paper deals with the importance of nutrient uniformity and the methods available to test for this. Having the precise knowledge of the nutrient requirements of an animal is negated if the feed is not mixed effectively and the paper identifies the importance of choosing the correct marker for the situations being tested.

## Introduction

While every area of the world involved in animal agriculture has its unique problems and perspectives regarding stock feed production, there are many generic issues for which there are often common solutions. Of the many generic issues, avoiding cross-transference of feed additives and testing for nutrient homogeneity are likely the most important.

Cross transference or ingredient carryover is a form of feed contamination that results when the substance in question is transferred (carried) from an acceptable location or feed to an unacceptable location or feed. Carryover of an animal drug can occur during feed processing, handling or delivery.

This paper deals with the US feed manufacturing industry and outlines procedures and policies which, when properly used, will avoid unsafe carryover. The US Food and Drug Administration's (FDA) Good Manufacturing Practices (GMP) provide guidance for medicated feed manufacturers to ensure their products meet the required identity, strength and quality standards with respect to their drug content. These regulations stipulate that adequate procedures be established and followed for all equipment used in the production and distribution of medicated feeds to avoid unsafe contamination of medicated and non-medicated feeds.

Carryover of a Category II drug (one which requires a withdrawal period) into a finishing ration may result in a residue problem in meat animals and/or milk or eggs. Producers experiencing this problem in their market animals may incur significant financial loss. Carryover of either a Category I drug (one which does not require a withdrawal time) or a Category II drug into a batch of feed intended for an off-label species may create serious problems. For example, carryover of an ionophore from cattle feed to horse feed may kill the horse.

The intent of this paper is to provide feed manufacturers, whether a commercial mill operator or an on-farm feed manufacturer, information that will help them avoid drug carryover. This information should help feed processors improve their product quality, reduce the likelihood of feed contamination, and help ensure safe meat, milk, and eggs destined for human consumption.

## Risk assessment

It is the responsibility of the feed processor (whether commercial or on-farm) to ensure that the correct amount of the desired drug is properly incorporated and that no cross-contamination of an unwanted/unspecified drug or other contaminant are present in that feed. Whether the manufacturing system is simple or complex, it is possible to avoid drug carryover by following the GMP.

The type of drug, number of species and feed delivery system determine the degree of risk associated with drug carryover. Feed processors, who manufacture products for one species and, using only a few drugs, experience the least amount of risk associated with cross-contamination and tissue residue.

The individual who uses a Category II drug can avoid carryover and tissue residue problems by using separate delivery systems for these feeds and by sequencing, flushing, and cleaning feed processing equipment. Mills that produce feed for multiple species experience an increased risk of cross-contamination by medicated articles (either Category I or II) than mills manufacturing products for one species. When changing feed rations from one species to



the next the risk of cross-contamination is minimized through sequencing, flushing, and clean-out of feed processing and delivery equipment.

It is an accepted fact that some level of carryover is unavoidable regardless of how clean the system is maintained or how carefully employees follow cleanout procedures. It is this point at which science, regulators and manufacturers must converge to create an understanding, based on risk-assessment, as to what level of contamination is acceptable. If the consensus is that there is no acceptable level of contamination for a specific compound or ingredient (e.g. animal protein in ruminant feeds), the only alternative is to totally remove the compound or ingredient from the mill and avoid its use altogether.

In most cases, scientific discovery has been our ally. However, as the technology of detection and assay precession has improved, zero is no longer zero. In 1958, an amendment to the US Food, Drug and Cosmetic Act was passed that set the tolerance for any known carcinogen at zero. At that time, detection limits were generally in the parts-per-thousand or, at most, parts-per-million. Today, scientists can easily measure certain compounds in the parts-per-trillion and even higher in some cases. As a result of scientific advancement, and based on a valid human health risk-assessment, the tolerance for some known carcinogens (e.g. Aflatoxin) has been set at a level that is easily measured but that will not result in a significant risk to human or animal health.

### Causes for carryover

Drug carryover may occur for many reasons as outlined in Table 1. Significant amounts of the drugs or medicated feed may remain in the production system and contaminate the following batches of feed. It may occur in one piece of equipment, or it may result from a combination of residues throughout the entire system.

**Table 1. Sources of carryover**

<b>Equipment</b>	<b>Mode of carryover</b>
Dust system	Delayed return of dust to production line Excessive pickup of drug and carrier Hang-up (electrostatic or moisture)
Mixer	Residual feed remaining in mixer Build-up of material on ribbons and walls Electrostatic hang-up on walls and top Leaking mixer gate (not fully closed)
Surge bin	Incomplete clean-out Electrostatic or moisture hang-up
Conveyors	Same as surge bin
Elevators	Residual feed remaining in buckets and boot Electrostatic or moisture hang-up
Bins	Bridging Residual feed from incomplete cleanout
Bulk truck	Error in bin chart records Incomplete clean-out Bridging and hang-up

When the source of carryover is known, corrective action can be taken and adjusting the equipment will often markedly relieve the problem. Repairs, remodelling or replacement of worn components may be necessary. Common corrective measures for carryover are listed in Table 2.

**Table 2. Some common corrective actions for carryover**

<b>Mode of carryover</b>	<b>Corrective actions</b>
1. Electrostatic hang-up	Ground wire to affected equipment Purchase non-electrostatic form of premix Use liquid ingredient to control dust Vibrators to shake hang-up loose
2. Delayed or extended dust return	Adjust air velocity at collection points Allow more time for dust to clear system Use liquid ingredient to reduce dustiness Collect and discard dust following production of medicated feeds (or retain for next run of like medicated feed) Remodel dust system
3. Mixer residues	Adjust ribbons or paddles Install plastic 'wipers' on ribbons Install air sweep jets for cleaning Remodel discharge for more complete cleanout Add drug when mixer is 1/2 to 3/4 full (may affect mixing time required for good mix)
4. Surge bin, conveyor residues	Adjust for more complete cleanout Remodel bin or discharge
5. Elevator residues	Adjust bucket clearance in boot (if possible) Install air sweep jets Remodel boot for more complete cleanout
6. Bin residues	Manual inspection and cleaning when changing kind of feed stored Install vibrator or air sweep jets
7. Pellet mill and dryer residues	Flush blender and dies Adjust dryer for more complete cleanout
8. Entire system	Use production scheduling Allow time between kinds of feed for manual cleaning of system Use 'flush' material - about 5% of mixer capacity, but not less than 200 lbs. (should be established by actual tests)
9. Bulk truck	Establish cleanout procedure for truck Require a sample from the first product discharged at point of delivery Analyze delivery samples randomly and let driver know that samples are being analyzed

### **Sequencing, flushing, and equipment clean-out**

When working with a drug that requires a withdrawal time before the meat animal goes to market or when manufacturing and delivering feed for several animal species, one must be careful to follow the label and use sensible practices to avoid drug cross-contamination. The GMP's state that adequate procedures shall be established and used for all equipment used in the production and distribution of medicated feeds to avoid unsafe contamination of medicated and non-medicated feed. Three techniques to avoid cross-contamination include sequencing, flushing and equipment clean-out.

#### *Sequencing*

The ordering sequence in which feed rations are processed and delivered determines the likelihood of drug carryover and tissue residue. It is an excellent practice to schedule the production of all medicated feeds having the same drug(s) in sequence with the higher levels first and ending with a low level. This sequence should be followed by a non-medicated feed for the same animal.

Individuals manufacturing feed for a single species such as swine, in which a withdrawal drug is fed to young animals, should generally mix feed in the following order: nursery ration containing the withdrawal drug, sow feed, grower, and finishing ration. Place cull sows in the finishing pen for an appropriate amount of time prior to sending them to market if this sequence is followed. When using a sequencing pattern to avoid cross-contamination, it is imperative that feed production records are kept and are detailed enough to denote the last batch/ration. Otherwise, the sequencing pattern could be violated by the next individual preparing feed.

In most feed mills, sequencing feed will reduce carryover enough to eliminate the potential for tissue residue. However, sequencing may not reduce carryover to a sufficiently low level if maintenance or design problems exist in the mill as described in Table 2.

### *Flushing*

Flushing involves using a safe ingredient, usually ground grain, and moving a quantity through the system to flush out any medicated feed that remains. The objective of flushing is simply to dilute the offending compound to the point that it is no longer of concern or danger to a given animal. The amount of flush material depends on the system (about 5-10 percent of mixer capacity) but should not be less than 100 kg of ground grain. Once the material has passed through the feed processing/conveying system, it must be stored in a separate bin for use in an identical medicated ration.

### *Single vs. Multiple Flushes*

Common practice is that a single flush batch of 5-10% of the mixer capacity is used. However, on certain occasions such as when a feed is made with an exceptionally high level of a medicated feed additive, a single flush may not suffice. If, for example, a production run of a feedlot supplement containing 2000 g/t Monensin® is completed, to be followed by the manufacture of a horse feed, a single flush would be far too risky. The following example is used to make the point:

Assume the following condition:

1. Batch size 4000 kg
2. Mixer residual after complete discharge 50 kg
3. Additive (PhuPhumicin - a 'make-believe medication') 2000g/t (8000g/4000 kg batch)

#### **Scenario A. No flushing at all**

50 kg residual with 100 g PhuPhumicin  
+4000 kg fresh ingredients  
 4050 kg total with 100 g PhuPhumicin (**0.0247 g/kg**)

#### **Scenario B. A single flush of 10% of the batch size (400 kg)**

50 kg residual with 100 g PhuPhumicin (2 g/kg)  
+400 kg flush  
 450 kg material with 100 g PhuPhumicin (0.222 g/kg)

The next batch manufactured will be:

50 kg residual with 0.222 g/kg or 11.11 g/50 kg  
+4000 kg non-medicated ingredients  
 4050 kg feed with 11.11 g PhuPhumicin (**0.00274 g/kg**)

**This is a 9 X reduction in concentration with a single flush compared to no flush**

Scenario C. Two flushes of 5% each (total of 10% of the batch size)

1st flush:

50 kg residual with 100 g PhuPhumicin 2 g/kg

+ 200 kg flush

250 kg feed with 100 g (0.4 g/kg PhuPhumicin)

2nd flush:

50 kg residual with 20 g PhuPhumicin (0.4g/ kg)

+200 kg flush

250 kg feed with 20 g PhuPhumicin (0.08 g/kg)

50 kg residual with 4.0 g PhuPhumicin

+ 4000 kg non-medicated ingredients

= 4050 kg feed with 4.0 grams PhuPhumicin (0.000988 g/kg)

**This is a 2.8 X reduction from the ‘single flush’ scenario and a 25 X reduction from the ‘no flush’ scenario.**

The point of the above example is that, with the same amount of flush material a substantial reduction in additive concentration can be obtained with two flushes compared to a single flush and almost any flush is better than no flush at all.

#### *Equipment clean-out*

Equipment clean-out is the least used, but potentially most effective, method of avoiding drug carryover during feed processing and delivery. Cleaning the mixer, conveying system, pellet cooler, and sack-off bin or delivery truck between runs to remove residual feed is recommended under high risk situations. These may include the following: working with a high potency form of a drug (making premixes); sequencing cannot be incorporated into the production schedule; feed processing systems have large carryovers between batches (e.g. portable grinder-mixers); or when physical properties of drugs are such that sequencing and flushing is not sufficient to prevent carryover.

The GMP's stipulate that all equipment shall be designed, constructed, installed, and maintained so as to facilitate inspection and use of clean-out procedures. Scheduled cleaning of mixers is required where liquid ingredients (molasses or fat) are added to the feed ration in the mixer.

#### **Operational issues and mill construction**

The objective of any feed enterprise is to design, manage and operate feed production facilities to control biological, physical and chemical hazards that may pose a threat to either animal feed or human food safety. A major concern for animal agriculture today is the perception that feed can somehow contribute to food safety issues. In fact, the US Center for Disease Control has made the statement that ‘...contaminated animal feed could actually be an important source of salmonella and other causes of human illness’. While this concept has not been proven and is certainly not a common route of human infection, there is the perception that we, in animal agriculture, could and should do more to protect the world food supply through design, management and operation of feed milling facilities.

As raw materials are turned into feed, they are combined with other ingredients or medications to form a complete diet for the target animal. At every stage of the process, there is the possibility of unintentional mistakes or even intentional adulteration or contamination. Our job is to take every step necessary to limit or eliminate even the possibility of this happening. This is partially done by creating internal and external programs such as QA/QC, GMP's, HACCP programs and various regulatory policies that, when followed, will prevent safety and contamination issues in feeds. However, if the feed mill design and construction is defective or if management fails to provide sufficient oversight, contamination may still occur and not be caught by the programs listed above.

## Operational and management

As mentioned earlier, this paper is being written from a US perspective and, at this time, most of the regulatory oversight is provided by following federally mandated Current Good Manufacturing Practices (CGMP's). GMP's mandate the minimum safe feed practices for the use of drugs in medicated feeds. They have been developed over nearly a half-century and are dynamic rather than static in nature. As new additives and/or equipment come on line, the GMP's are modified to accommodate those changes.

For a feed mill to use certain classes and strengths of feed additives (drugs), the mill must be licensed by the federal government and is subject to periodic inspections. The GMP regulations require compliance in the following areas:

- Personnel

Ensuring that employees are trained and understand the steps necessary to avoid carryover and feed adulteration is the most critical issue in avoiding problems in this area.

- Buildings

All physical facilities used in the receipt, use, storage and distribution must be designed, constructed and maintained in a manner that ensures the safe use of feed additives.

- Equipment

All equipment, including scales, used in the manufacture of medicated feed must be of proper design and operated in a manner that will not result in adulteration of feeds.

- Laboratory Controls

Periodic assay of medicated feeds are to be conducted to provide a measure of manufacturing performance.

## Facility design and operation

In many instances, the design of a feed mill and/or the selection or condition of specific equipment makes cross-contamination or unintended contamination unavoidable. To minimize or avoid unsafe contamination, it is necessary to determine where in the mill the situation might occur. The starting point for this self-diagnosis is a complete and up-to-date flow diagram of the mill that identifies potential areas of concern. Likely areas include the bulk receiving system, bin bottoms, gates, conveyors, elevator boots, processing equipment and even lubricants.

The following is a brief discussion of some of the more common areas in a mill where cross-contamination is likely to occur.

### Batching system

If an automated batching system is used, the 'medicated feed interlock' system should be active. The ability to change or override the interlock system should be limited to senior management to avoid having carryover issues due to lack of understanding on the part of an employee. In all cases medicated and non-medicated feeds are manufactured in the same facility, a pre-approved sequencing chart should be available to the batch operator.

### Mixers

The 'Drop-Bottom' mixer design is the most effective way to avoid residual feeds being carried into the next batch of feed. If a drop bottom mixer is not available, careful attention should be paid to flushing procedures and sequencing. The clearance between the mixer reel and the tub should be adjusted and maintained to the minimum practical distance. Mixers should be tested at least annually to assure that expected additive uniformity is being met at the end of the specified mixing time.

### Pelleting system design

In many US feed mills, attention is closely paid to flushing the mixer and mash conveying equipment but little if any attention is paid to flushing the pelleting system. In fact, it is common practice to withhold 500-1000 kg of grain from the last batch of a production run, then weigh out the appropriate weight of grain as an added batch, mix it as if it were a single batch of feed and convey the grain to the pellet mill mash bin. While this undoubtedly flushes the mixing and conveying system, it is difficult to see how this flushes the pelleting system.

In most cases, the pellet mash bin is equipped with a pneumatic vibrator to assure that there is no mash remaining in the mash bin at the completion of a run. The conditioner is the most likely component where significant residual feed can be found. By sending a flush through at the end of a pelleting run, carryover should be avoided.

Special attention should be paid to the fines return system if the pellets are being cleaned before being conveyed to the load-out system or bagging operation. If scalper fines are being continuously returned to the pellet mill, it will take a long time to completely finish a run. The recommended procedure is to by-pass the scalper once the mash supply bin is empty. This way, residual fines will not be recycled to the pellet mill and a clean ending point will be reached.

### Other probable points of cross-contamination

Special attention should be paid to elevator boots, spouting, screw and drag conveyors and flow gates. They should be of proper design and should be maintained in such a way that carryover will be avoided.

A lot of thought is being given to controlling microbial contamination in feeds. Of course, the most practical way to accomplish control is with thermal processing and pelleting, with proper controls, is effective. Temperatures must reach in excess of 80°C for a minimum of 30-40 seconds for efficient thermal control. Caution should be used in examining down-stream routing to ensure that recontamination cannot occur.

Another way of obtaining microbial control is with chemicals such as propionic acid or formaldehyde preparations. Both of these compounds are approved for use in numerous rations in the US.

### Nutrient uniformity and uniformity testing

Nutrient uniformity is critical for nearly all animals if optimum growth and performance is to be obtained. Uniformity is especially important for the young animal or small animals consuming a small daily ration. Beumer (1991) indicated that mix uniformity is one of the critical quality control points in feed manufacturing. Concerns for creating a uniform mix would include the following: 1) nutritional over-fortification by the nutritionist (Wicker and Poole, 1991); 2) regulatory aspects (Muirhead, 2006); and 3) animal performance (McCoy *et al.*, 1994).

While testing feed to determine if uniformity has been achieved seems intuitive and is, in fact, a common practice, there is little agreement in the industry regarding exactly what marker to test for and how the testing should be accomplished. From the animal performance standpoint, the only place that uniformity is really important is in the bunk or feeder in front of the animal. Yet, nutrient uniformity is seldom established at that location but is most often determined at the mixer. This is based on the idea that feed must, at the least, begin its journey as a uniform blend if there is to be any hope of uniformity at the point of consumption.

In testing a feed for uniformity, a great deal of thought must go into selecting the marker that is to be used to determine uniformity. Post *et al.* (1966) stated that the criteria for marker selection should include the following: 1) avoid using a marker in which the variation will not affect animal performance (e.g. Vitamin A); 2) select a marker whose physical properties are similar to the major ingredients in the feed (particle size and density); 3) do not utilize a characteristic shared by the majority of other ingredients in the feed (e.g. protein, ash etc); and 4) analytical assay variability for the tested marker must be less than the target mixer variability (e.g. target CV=10%, assay CV=5%).

A recent study (Clark *et al.*, in press) reported on the evaluation of several markers to determine mix uniformity. Treatment diet was a corn-soybean meal based diet formulated for broiler starter chicks. Dietary nutrients or tracer evaluated included the following: 1) DL-Methionine; 2) L-Lysine; 3) Crude Protein; 4) fine mixing salt; 5) Phosphorous; 6) Manganese; 7) Dyed iron particles (MicroTracer® red and blue); 8) Roxarsone and 9) Semduramicin. Diets were mixed using a 500 kg capacity double ribbon mixer for 0.5, 2.5 and 5.0 minutes. The mixer was stopped and ten samples obtained at each mix time using a grain probe.

Overall, from 0.5-5.0 minutes mix time, all markers showed a numerical improvement in nutrient uniformity. However, there were substantial differences in the effectiveness of certain markers to show improvement in uniformity with time. For example, crude protein and phosphorous uniformity showed little in the way of improvement because many of the ingredients contribute to the pool of each nutrient confounding the results. Common nutrients such as protein, calcium, phosphorous and the like should never be used as markers for uniformity evaluation. The synthetic amino acids DL-Methionine and L-Lysine were the only markers that showed a reliable improvement in uniformity with extended mix time (23.86 to 9.47%). Of the two feed additives, Roxarsone and Semduramicin, only Semduramicin approached the target CV of 10% (CV=11.23%) at the end of the 5.0 minutes mix time. Roxarsone variability was still above 25% CV after 5.0 minutes mix time. In both cases, the high CV is likely due to the inherent assay variability for each additive.

The marker chosen for mix uniformity studies can significantly influence the outcome of the study. The entire area of study regarding the effect of nutrient uniformity on animal performance and the evaluation of nutrient uniformity in blended stock feeds has been a bit of an orphan in animal agriculture for decades. In formulating a diet, one of the basic assumptions made by nutritionists is that the manufacturing process is perfect and that every mouthful consumed by the target animal perfectly represent the intended nutrient profile in the formula. In many cases, this is far from the truth.

## Summary

The objective of this paper was to discuss the risk of unsafe carryover of animal feed additives and to describe ways in which the problem could be avoided. Medicated feeds are important to animal health and growth promotion. Avoiding drug carryover during feed processing and delivery is essential when using medicated articles/feeds. Procedures to avoid cross-contamination between feed batches include assessing the risk and potential causes for drug carryover, preventative maintenance of feed processing equipment and correct use of sequencing, flushing, and equipment clean-out procedures. Ingredients can segregate during feed handling and delivery.

The answer to most cases of cross-contamination lies in the design, operation and management of our feed mills. However, without proper employee training and supervision, contamination and carryover are likely to happen. We all know of situations where a disgruntled employee has tried to get back at a manager through what amounts to an act of terrorism. Selecting proper equipment and training employees to understand the regulatory requirements and operational policies will avoid most unsafe contamination.

While nutrient uniformity and mixer evaluation are known to be critical aspects of any feed manufacturing enterprise, there is little agreement on how uniformity should be evaluated and even to the extent nutrient uniformity is important to various species and ages of animals. Some markers commonly used to describe uniformity, such as protein and calcium, are totally worthless while others that are extremely good at measuring uniformity are simply too expensive to consider. This should be an active area of research for animal scientists.

# New frontier in processing: ingredient fractionation

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## Abstract

Ingredient fractionation, or the separation of crop products into value added co-products, has been practised for many centuries as is the case with oil extraction. More recently, it has been identified that seeds of plant crops also contain carbohydrates and proteins that can have value in a range of food and industrial applications. Consequently, more co-products are becoming available that may have advantages in swine feeding. This paper discusses the rationale behind the production of co-products from oats, canola, flax, barley, field pea, wheat and wheat and corn-dried distillers-grains plus solubles (DDGS). The issues and opportunities for these co-products for swine feeding are then identified, with particular focus on DDGS.

## Introduction

The province of Alberta has a strategy to develop a bio-based economy (AARI, 2003). The fractionation of crop products into value-added co-products is a key component. Seeds of plant crops contain some of the three main macronutrients: carbohydrates - divided into sugars, starch and non-starch polysaccharides (NSP) - protein and oil (fat). Some of these crops have been fractionated commercially for centuries, usually to acquire the oil. Canola and flax oil are present examples of processing crops for oil in western Canada. Ingredient tables list a large array of ingredient fractions that can be used in the feed industry (e.g. CVB, 1994). Commodity fractions of ingredients produced in western Canada include canola meal, wheat by-products, oat groats, sugar beet pulp and the newest feedstuff: distiller's dried grain with solubles (DDGS).

In this summary, the current state of knowledge regarding ingredient fractionation relative to the feed industry is described for the most important crops in western Canada. These crops are canola, flax, oats, barley, wheat, and field peas. Canola and flax are oil seeds containing large amounts of fat and small amounts of starch; oats, barley and wheat are cereals containing large amounts of carbohydrates, mostly starch, and small amounts of protein; field peas are pulses containing protein and starch (CVB, 1994) (Table 1).

**Table 1. Concentrations of starch (+ sugars), NSP, protein, and fat (% as is; adapted from CVB, 1994) of 7 selected feed ingredients grown in western Canada**

Crop	Starch	NSP	Protein	Fat
Oats	39	31	11	5
Canola	4	24	20	41
Flax	9	21	22	35
Barley	54	18	11	2
Field peas	47	14	23	1
Wheat	61	10	12	2

Most examples of high-value ingredient fractions in swine production include fractions that can be incorporated in diets for weaned pigs ensuring a rapid increase in voluntary feed intake of digestible nutrients, thereby stimulating a successful transition from sow milk to a dry diet. Prime examples of established high-value, plant-based ingredient fractions that are used in the feed industry include canola oil, oat groats, and soy protein isolates and concentrates. Other examples may include fractionated canola meal, peas and flax in the near future. However, examples ensuring that the extra diet costs are captured in the marketplace in the fetched price for pork are currently scarce.

Most ingredient fractions that are used in the feed industry come from a second approach of building value-added processing into animal nutrition. An ingredient is fractionated, and a minimum of one fraction is used for human or industrial application, while the remaining co-product goes into the feed market; DDGS is a prime example in this category. Information on ingredient fractionation combined with feeding all resulting individual fractions to a single animal species is scarce. This paper therefore is focussed on describing the potential for value-added ingredient processing for animal nutrition for the most important seeds crops in western Canada, and in particular DDGS.



## Value-added processing

A range of technologies exists to fractionate ingredients. There are two categories: 1) an up-front fractionation process allowing further processing of individual ingredient fractions and 2) a process on the entire ingredient that separates one fraction from the ingredient. Examples of category 1 include dry and wet milling and air classification, etc. Examples of category 2 include current ethanol production procedures and oil extraction from canola. Dry separation techniques (dry milling/air classification) are particularly useful for the production of protein-rich fractions from non-oilseed legumes, such as peas (Dijkstra *et al.*, 2003). The advantages of dry over wet separation techniques are lower costs and the absence of effluents. However, wet-processing techniques may result in fractions containing a higher protein concentration. Category 1 and 2 processes can be combined for one ingredient. For example, soybeans are first fractionated into soy oil and soybean meal. Subsequently, soybean meal is fractionated into protein concentrates and isolates, and more purified fractions such as isoflavones (Potter, 1998).

## Oats

Oats contain a large amount of NSP that are non-digestible by swine. Most of these NSP are included in the oat hull, and mechanically dehulling of the oats therefore results in highly digestible groats and less-digestible hulls (Patience *et al.*, 1995). Oat groats are used widely in starter diets, because they are palatable, contain little fibre, and are highly digestible (Lin *et al.*, 1987). Because of the removal of the fibrous husk, oat groats contain 7% oil (Asp *et al.*, 1992). Naked oats are a natural variant of oats resulting in most of the hull being removed easily (Morris, 1990), resulting in most of the hull and thus the indigestible fibre fraction being removed. However, naked oats have not gained widespread adoption to compete against oat groats, in part due to issue related to storage related to rancidity of the oil.

Despite its high reputation as an ingredient for starter diets, few peer-reviewed articles exist showing a benefit of oat groats inclusion. In diets containing 45% dried skim milk, oat groats and corn as basal grain sources resulted in similar growth performance of starter pigs for two weeks following 21 days weaning (Mahan and Newton, 1993). Similarly, replacement of wheat with 25% (domestic) oat groats resulted in similar performance in four week old starter pigs for five weeks (Thacker and Sosulski, 1994). Finally, regular and high-oil oat groats improved growth performance in three week old starter pigs for four weeks compared to wheat-based but not corn-based diets (Zijlstra *et al.*, 2002a). These results seem to indicate that beneficial effects of oat groats may be limited to the immediate post-weaning phase.

Oat hulls contain a fibre fraction that is resistant to digestion and fermentation (Zervas and Zijlstra, 2002). Oat hulls are used occasionally in sow diets as a fibre source to prevent constipation. Oat hulls are not considered as a regular feed ingredient for grower-finisher pigs. In contrast, the  $\beta$ -glucans contained in oat groats have a high digestibility (Bach Knudsen *et al.*, 1993). The  $\beta$ -glucans extracted from the groats (endosperm/bran) may impact intestinal bacteria populations or act as immuno-stimulator (Yun *et al.*, 2003), indicating that the NSP contained in oats may have functional characteristics benefiting animal health.

## Canola

Within the history of agriculture of Western Canada, canola is one of its premier success stories. The creation of canola from rapeseed through crop breeding by Downey and Stefansson (Rakow, 2000) resulted, after fractionation, in oil and meal fractions that both could be fed to domestic animal species. At the canola crusher, canola seed is processed into canola oil and canola meal using solvent extraction. Although canola meal fetches a premium price in the dairy feed market, canola meal is sold at a discount in other feed markets. The oil is further purified into edible canola oil, or remains feed grade. Following purification, the canola oil is used as a human food, although the oil fraction might also be converted into bio-diesel (Zou and Atkinson, 2003) or bio-plastics (Narine, 2003). Recently, canola press-cake has been introduced, a product that is generated following cold-pressing of canola. More residual oil and thus energy remains in the cake than in canola meal.

Limitations for optimum use of the canola meal remain (Bell, 1993). The NSP, phytate, and other compounds may limit nutrient digestibility of canola meal by non-ruminants. The hull of canola seed is high in poorly digestible NSP, and partial mechanical tail-end dehulling will reduce NSP content and increase protein content of canola meal (De Lange *et al.*, 1998). As a result, energy digestibility and DE content of dehulled canola meal will be increased for swine.

Further diversification for canola can be achieved by, for example, fractionating the meal to a variety of feed products. Proprietary technology to achieve this feat has been developed by MCN Bioproducts Inc (MCN, 2004). A main product stream will be a high protein fraction that can be used as a (partial) replacement of fish diet in the high value diets for the commercial fish industry. In a similar process, physical, enzymatic and chemical processing of commercial canola meal results in a canola protein fraction isolate that is low in NSP and phytate and thus has a high nutrient digestibility for fish (Mwachireya *et al.*, 1999) and swine.

## Flax

More recently, fractionation of flax is getting increasing attention. The use of flax oil for its health benefits was started centuries ago. The omega-3 fatty acid contained in flax seed and thus fractionated flax oil can be included in the diet, and thereby be concentrated either in meat products such as pork (Romans *et al.*, 1995). Swine research with flax has primarily focused on changing fatty acid profile of pork by using dietary flax seed, and recently on improving the nutrient digestibility of flax (Htoo *et al.*, 2007). The health benefits of omega-3 fatty acid have been described in numerous reviews (e.g. Covington 2004).

Apart from its benefits via changes in fatty acid profile, flax seed may have other desirable digestible nutrient and functional characteristics for swine. However, the total effect of flax seed or individual flax fractions on intestinal microbial populations has not been well described (Bhatty, 1993). Flax seed contains 20% hull, 40% oil and 28% dietary fibre (Flax Council, 2002), and both fibre and fat fractions may change the intestinal microbial populations directly due to the anti-microbial properties of flax fractions or by serving as a substrate for intestinal bacteria, or indirectly through modulations of intestinal inflammatory responses.

Flax fractions including  $\alpha$ -linolenic acid (Lee *et al.*, 2002), lignans (Pauletti *et al.*, 2000) and cyanogenic glycosides (Osborne, 1996) have broad anti-microbial activity against bacteria and fungi present in the small intestine. These products are not present in significant concentrations in other feed ingredients used in swine diets suggesting that flax may exhibit antibiotic like activities. Flax also contains 10% mucilage, which is a water-soluble carbohydrate that can be removed by hot-water extraction (Bhatty, 1993). Mucilage will increase the viscosity of digesta in the small intestine causing a reduction in nutrient digestibility (Garden-Robinson, 1994).

Results from our laboratory indicate that flax fractions, in particular flax oil and hulls are indeed capable to change intestinal microbial populations (Smith *et al.*, 2004). The use of flax seed or its fractions as an antibiotic replacement might thus appeal to all swine producers.

## Barley

The main value-added processing for barley has been malting and brewing, resulting in the production of beer and brewer's grain. Similar to oats, most of the barley NSP is contained in the barley hull. However, the hull content is less in barley and the NSP contained in the barley hull appear more digestible than oat hull NSP. Still, barley NSP is negatively related to energy digestibility in pigs (Fairbairn *et al.*, 1999). Hull-less barley is a natural variant of barley resulting in most of the hull being removed easily during harvest (Aherne, 1990), resulting in most of the hull and thus the less fibre fraction being removed. Hull-less barley acreage has declined recently, because yield of hull-less barley has been consistently lower than for covered barley, partly due to the removal of the hull, and the lack of a substantial price difference between wheat and barley to overcome the yield reduction. Finally, the hull remains of the field and value of the hull fibre can thus not be captured.

Similar to oats, barley could be pearled using processing to produce a highly digestible barley kernel by removing the less digestible hull. Dehulling of barley by the techniques used for oat dehulling is difficult (Hoseney, 1994). Commercially, pearling of barley is solely performed for human food application to access the functional characteristics of the barley kernel (Edney *et al.*, 2002). Recently, fractions of the barley hull (i.e. pearling by-products) gained interest for human food applications. Some pearling flour fractions (from later stages of pearling) contain high amount of  $\beta$ -glucans that can be enriched using milling and sieving, and included in functional pastas (Marconi, 2000). Finally,  $\beta$ -glucans have been extracted from barley using patented technology (Temelli and Vasanthan, 2003), to enable food application for the  $\beta$ -glucan concentrates. Similar to oat  $\beta$ -glucans, barley  $\beta$ -glucans might provide health benefits for animals (Wang *et al.*, 1992). The wet fractionation process used to generate  $\beta$ -glucans from barley and oat also generates starch concentrates (Table 2) that may have application in the swine industry (Johnson *et al.*, 2007).

**Table 2. Energy characteristics of hull-less barley and oat grain and starch concentrates<sup>1</sup>**

Item	Hull-less barley		Hull-less oats	
	Grain	Starch concentrate	Grain	Starch concentrate
Energy digestibility, %				
Ileal, %			89.0 <sup>ab</sup>	90.2 <sup>ab</sup>
	86.4 <sup>b</sup>	91.4 <sup>a</sup>		
Total tract, %	94.8	96.0	97.3	96.6
DE, Mcal/kg DM	4.45	4.44	4.83	4.74
NE, Mcal/kg DM <sup>2</sup>	3.37	3.51	3.60	3.63

<sup>1</sup>Source: Johnson *et al.* (2007); <sup>2</sup>NE = 0.700 x DE + 1.61 X EE + 0.48 x Starch - 0.91 x CP - 0.83 x ADF

### Field peas

The field pea is an interesting crop for ingredient fractionation. Among the pulse crops, fractionation of field peas has a strong tradition but not as strong as the main competing legume and cereal: soybeans and corn. In western Canada, field peas have been fractionated commercially into protein, starch, and NSP fractions (Parrheim, 2004), all of which are commercially valuable for food processing.

In pigs, pea starch is slightly less digestible by the end of the small intestine, but similar in digestibility at the end of the total tract than wheat. However, the rate of pea starch digestion is lower than wheat (Fledderus *et al.*, 2003). Pea starch thus has a lower glycaemic index than wheat or barley. Pulses including faba bean in general have a lower rate of starch digestion than cereal grains. Following extrusion, difference in kinetics of nutrient digestion remain (Wierenga *et al.*, in press). Legume starches are different from cereal and potato starches, and possess some improved functional properties. For example, pea starches are more soluble and swell less than cereal starches (Wang *et al.*, 1998). The most important starch characteristics for animal nutrition are total and rate of digestion.

**Table 3. *In vivo* starch digestion kinetics of extruded diets containing wheat or faba bean starch fed to weaned pigs<sup>1</sup>**

Starch digestibility (%)	Wheat	Wheat and zero tannin faba bean starch	Zero tannin faba bean starch	Pooled SEM
Duodenum	80.66 a	79.23 a	73.07 b	1.23
Jejunum	91.75	89.03	91.64	0.84
Ileum	95.79 A	95.04 AB	93.04 B	0.62
Total tract	98.17 b	98.45 a	98.37 a	0.06

<sup>1</sup>Source: Wierenga *et al.* (in press).

Pea protein is well digested by pigs. However, amino acid digestibility appears lower than for soybean meal (Mariscal-Landin *et al.*, 2002). The predominant protein fraction in peas is the globulin fraction, and albumins are the secondary fraction. Peas also contain anti-nutritional factors that may interfere with protein digestion, including trypsin inhibitors, lectins, tannins,  $\alpha$ -galactosides and alkaloids.

Pea NSP are concentrated in the pea hulls. For more than a decade, peas have been identified to be a rich source of fermentable NSP that produce a large intestine fermentation pattern of potential health benefit (Goodlad and Mathers, 1990). Compared to wheat and oat bran NSP, pea NSP has less bulking capacity and does not reduce passage rate as much (Hansen *et al.*, 1992). Pea hulls have a high cellulose content. Apart from the pea hull NSP, the NSP contained in the cotyledons appear to increase endogenous N losses without affecting protein digestion (Leterme *et al.*, 1998). These inner pea NSP have a high water-holding capacity. Generally, high solubility, swelling and water-holding capacity of NSP are related to high fermentation and development of microbial populations. Indeed, the pea cotyledon has a higher water-holding capacity than the pea hull (Canibe and Bach-Knudsen, 2002) and results in a higher production of volatile fatty acids.

Similar to canola meal, peas can be dehulled and the protein concentrated to create plant-based proteins to replace fishmeal in aquaculture. Dehulling peas improves energy and protein digestibility of peas in diets for silver perch, and protein concentrates had the protein digestibility (97%), suggesting that pea fraction may be excellent protein sources for some fish species (Booth *et al.*, 2001).

## Wheat

The starch content in wheat is highest among small cereals. Thus, wheat has been the main feedstock for ethanol production in western Canada. Increased wheat NSP is related to reduced energy digestibility (Zijlstra *et al.*, 1999). Although wheat contains less hull than oats and barley, removal of the hull and therefore the NSP contained in the hull using dehulling will increase the energy digestibility and DE content of wheat (Zijlstra *et al.*, 2002b). However, attractive (human) markets for high quality wheat bran will be needed to offset the cost of ingredient fractionation.

Wheat gluten is being fractionated for specific cooking purposes for human markets. However, the proteins included in the gluten have gained attention due to being a contributing factor to celiac disease in humans (Mowat, 2003). Celiac disease causes damage to the epithelium of the small intestine, leading to mal-absorption of nutrients. The information regarding the protein fraction of wheat suggests that ingredient fraction that may have benefits for most individuals might have a detrimental effect for some.

Wheat co-products from dry milling for flour production are gaining increasing attention in the swine industry as an opportunity to reduce feed costs. These by-products are generally available at a reduced cost. However, much research will have to be completed to characterize and improve the nutritional value of the by-products (Nortey *et al.*, 2007a, 2007b). These co-products have a high content of NSP and phytate and are therefore prime candidates for feed processing including enzyme supplementation to improve energy digestibility.

## Wheat and corn DDGS

For ethanol production, the wheat feedstock is ground and undergoes the fermentation process. The ethanol is extracted and the remaining by-product (i.e. distiller's grain) is either fed directly to cattle or dried together with the thin solubles to create DDGS. Wheat thin stillage as a liquid and thin stillage and wet wheat distillers' grains are good sources of nutrients for ruminants (Mustafa *et al.*, 2000). For swine, wheat DDGS is an alternative ingredient with a similar digestible profile of critical nutrients as wheat (Widyaratne and Zijlstra, 2007). Corn DDGS is the main co-product from ethanol production in the US.

**Table 4. Chemical characteristics of wheat, and corn and wheat DDGS (% DM)<sup>1</sup>**

Variable	Wheat	Corn DDGS	Wheat DDGS
Moisture	11.8	11.8	8.1
Crude protein	19.8	30.3	44.5
Non-protein nitrogen	4.6	5.4	10.2
Crude fat	1.8	12.8	2.9
Ash	2.1	4.8	5.3
Acid detergent fibre	2.7	14.6	21.1
Neutral detergent fibre	9.4	31.2	30.3
<b>Amino acid</b>			
Lysine	0.52	0.83	0.72
Methionine	0.32	0.61	0.69
Threonine	0.54	1.09	1.28
Tryptophan	0.23	0.23	0.44

<sup>1</sup>Source: Widyaratne and Zijlstra (2007).

The nutrient content of DDGS reflects the starch removed during ethanol processing. The content of measured characteristics was for the wheat sample than the two DDGS samples (Table 3), except for phytate (Table 4). The crude protein content was highest for wheat DDGS. The non-protein nitrogen content of wheat DDGS was higher than for corn DDGS, indicating heat damage during the drying process. The crude fat content was highest for corn DDGS. The fibre content was similar between the DDGS samples, and was higher than in wheat. Similar to CP content, the amino contents was highest for wheat DDGS, with the exception of lysine (Table 4). With the exception of lysine, the AA content in wheat DDGS doubled that of wheat.

Among the DDGS samples, wheat DDGS had the highest total P content and corn DDGS had the highest intact phytate (IP6) content (Table 5). Wheat did not contain any of the lower forms of phytate (IP2, IP3, IP4 and IP5). In addition to phytate, wheat DDGS contained all lower inositol phosphate forms of phytate; corn DDGS and

wheat/corn DDGS did not contain IP2. Wheat DDGS had the highest soluble and total NSP content, but corn DDGS had the highest insoluble NSP content (Table 4). The arabinoxylan were higher in wheat DDGS than corn DDGS.

**Table 5. Phosphorus, inositol phosphates and NSP including constituent sugar profile of wheat, and corn and wheat DDGS (% DM)<sup>1</sup>**

Variable	Wheat	Corn DDGS	Wheat DDGS
Phosphorus	0.40	0.86	1.10
<b>Inositol phosphates</b>			
Inositol diphosphate (IP2)	ND	ND	0.08
Inositol triphosphate (IP3)	ND	0.09	0.09
Inositol quadruphosphate (IP4)	ND	0.19	0.28
Inositol pentaphosphate (IP5)	ND	0.45	0.64
Phytate (IP6)	1.39	0.92	0.81
<b>Total NSP</b>			
Soluble	2.15	1.39	7.76
Insoluble	7.57	17.85	15.13
Total	9.72	19.24	22.89
<b>Arabinoxylan</b>			
Soluble	1.71	0.5	4.66
Insoluble	4.03	9.92	8.29
Total	5.74	10.42	12.95

<sup>1</sup>Source: Widyaratne and Zijlstra (2007). ND = Not detected

The apparent ileal and total-tract digestibility of energy did not differ between DDGS samples (Table 6). However, energy digestibility was higher for the wheat than for the DDGS samples ( $P < 0.05$ ). The high DE content of corn DDGS reflects the high fat content, and corn DDGS may have the same energy content as the corn of origin (Pedersen *et al.*, 2007). The total-tract digestibility of P was 40% higher for DDGS than the wheat ( $P < 0.05$ ). The standardized ileal digestibility (SID) of Lys was lower for DDGS than wheat. However, the content of SID Lys was higher in corn DDGS than wheat DDGS, providing further evidence that the analyzed wheat DDGS sample was heat damaged during fermentation or drying. Variability in digestible nutrient content is one of the main concerns for the feeding of co-products including corn DDGS to swine (Pedersen *et al.*, 2007). With increasing inclusion of wheat and corn DDGS into swine diets, equivalent performance can not be achieved, in part due to a depressed feed intake and likely in part due to diets not being equivalent in NE content (Thacker, 2006; Whitney *et al.*, 2006).

**Table 6. Phosphorus, inositol phosphates and NSP including constituent sugar profile of wheat, and corn and wheat DDGS (% DM)<sup>1</sup>**

Variable	Wheat	Corn DDGS	Wheat DDGS	Pooled SEM
Energy digestibility, %				
Ileum	71.8	67.3	65.6	1.63
Total tract	84.8 a	78.7 b	77.4 b	1.39
DE content, kcal/kg DM	3807 b	4292 a	4019 b	73.4
SID Lys digestibility, %	78.3 a	66.6 b	64.1 b	2.08
SID Lys content, %DM	0.41 c	0.55 a	0.46 b	0.02
P digestibility, %	14.8 b	55.5 a	53.0 a	4.15
Digestible P content, %DM	0.06 b	0.48 a	0.59 a	0.04

<sup>1</sup>Source: Widyaratne and Zijlstra (2007).

### Functional characteristics of fractions

The main fractions possess functional characteristics that differ among ingredients. These characteristics are extremely important for food production but may also play a role in animal nutrition. Some examples are given.

- Starch

Even though ileal starch digestibility may be similar and total-tract starch digestibility may be identical among the ingredients, rate of starch digestion will be different (Fledderus *et al.*, 2003). The rate of starch digestion influences the glycaemic index of the feed, and this index may impact protein deposition. The ratio of amylase to amylopectin may be an important determinant of starch nutritional characteristics.

- NSP

The NSP can be characterized into soluble and insoluble fractions. Each fraction contains some unique properties, but the soluble fraction is expected to interact more with rate of digestion and absorption, passage rate and microbial populations (De Lange, 2000; Owusu-Asiedu *et al.*, 2006). Previous work in our laboratory has shown that the carbohydrate and protein fractions of feed ingredients may exert significant influence on bacterial populations in the small intestine of pigs (Drew *et al.*, 2002).

- Protein

The protein and gluten fraction is obviously critical to supply digestible amino acids. However, protein also contains antigenic properties that may be important factors for food allergies and hypersensitivity reactions in the intestinal tract. Dietary protein may also affect intestinal health by modulation of the intestinal mucus layer (Montagne *et al.*, 2004).

- Fat

Fat provides the highest amount of gross, digestible, and net energy per gram of mass. Ingestion of fatty acids will lead to their distribution to virtually every cell in the body with effects on membrane composition and function, eicosanoid synthesis, cellular signalling and regulation of gene expression (Benatti *et al.*, 2004) and thereby among other affecting the immune system.

### Conclusion

In North America, the long-term tradition of swine feeding is mostly based on simple grinding of cereal grains and grain sales being targeted towards livestock feeding is gradually changing. Crop producers especially embrace the current status of excellent marketing opportunities of the crops due to the increase in value-added processing, especially ethanol production. Ingredient fractionation opens the door to the feeding of a wide range of co-products in swine feeds, with a large range in nutrient characteristics. The feeding of co-products will become more of a norm, and especially the feeding of DDGS to swine will become prevalent in western Canada. Although ingredients can be fractionated successfully for the ethanol industry and for animal nutrition, especially for high margin market such as fish, most ingredients are fractionated for the human food supply or industrial processes.

# Challenges facing feed manufacturers: an Australian perspective

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## Abstract

Australian pork producers often have access to a wide variety of raw materials from which diets can be formulated. In many cases the raw materials themselves are inherently variable with respect to their nutrient composition. The chemical composition of these raw materials is generally well described, but often the chemical descriptions of the energy sources give no indication of the digestible energy (DE) that a pig will be able to extract from that energy source or how that raw material will perform during the feed manufacturing process. This overall variability raises significant issues for animal nutritionists but also poses particular problems for feed manufacturers. This paper describes the various processes raw materials undergo in their transformation into a feed, with a focus on pelleting, and the key drivers of the efficiencies in these processes. In many cases conflicts can arise between the nutritionist's requirements for the feed and the ability of the feed manufacturer to maximize the performance of the mill. By exploring why these conflicts arise, the paper identifies some key areas where recognition and understanding of the profit and performance drivers of feed manufacturers by nutritionists and pork producers could result in mutual benefits.

## Introduction

Due to the relatively small size of the industry in relation to its geographic spread, combined with the raw material choices on offer, feeding the Australian pig herd provides many challenges for nutritionists and feed manufacturers. While the pig herd is small by world standards at approximately 318,000 sows (Australian Pork Limited, 2006) and assuming an average feed intake of 5.4 t per year for each sow and her progeny, the industry requires approximately 1.6 million tonnes of feed per year. Unlike the United States, there is not a corn-soy belt that provides bountiful amounts of raw materials in close proximity to the pig growing areas. Unfortunately and, unlike most of Europe, imported grains do not provide a ready source of cost effective raw materials. Consequently, pork producers in Australia have to draw from a diverse basket of local and imported raw materials depending on the season and their location (Table 1).

**Table 1. Possible protein and energy ingredients in pig feeds in Australia**

Wheat	Sunflower meal	Skim milk powder	DDGS from sorghum
Oats	Canola meal	Dried buttermilk	DDGS from wheat
Groats	Cottonseed meal	Full cream milk powder	DDGS from millrun
Barley	Soybean meal	Cheese powder	Wheat germ
Triticale	Safflower meal	Lactose powder	Dextrose
Sorghum	Linseed meal	Whey	Sugar
Corn	Copra meal	Millrun	Ground oat meal flour
Rice	Palm kernel meal	Wheat bran	Rice hulls
Lupins	Peanut meal	Wheat pollard	Lupin hulls
Faba beans	Linseed	Rice pollard	Oat hulls
Mung beans	Peanuts	Rice bran	Cottonseed hulls
Chick peas	Tick beans	Wheat gluten	Almond hulls
Field peas	Corn/soya extruded	Pea pollard	Soybean hulls
Meat meal	Full fat soybean meal	Chick pea offal	Sunflower hulls
Blood meal	Soy lti flour	Hominy meal	Peanut shell
Fishmeal	Corn gluten meal	Oat flour	Peanut hulls
Poultry offal meal	Corn gluten	Oat starch	Lucerne meal
Malt	Vegetable oil	Oat pollard	Millet
Tallow			

Understanding the challenges of how best to utilize many of these raw materials was the basis of the symposium led by Black (1997) and discussions of this nature led to the formation of the Premium Grains for Livestock Project (PGLP), a joint project funded by Grains Research and Development Corporation, Australian Pork Limited, Meat and Livestock Australia, Australian Egg Corporation Limited, Rural Industries Research and Development Corporation – Chicken Meat and Ridley AgriProducts Pty Ltd. A key finding from the PGLP from an animal perspective was that when the same known grain samples were fed to broilers, layers and pigs, distinct differences were observed in the available energy between the grains as well as between the species (Table 2).

**Table 2. Range obtained in the Premium Grains for Livestock Program for the available energy content of cereal grains following digestion by different animal types (from Black *et al.*, 2006)**

Available Energy content (MJ/kg DM)					
Animal Type	Wheat	Barley <sup>2</sup>	Oats <sup>2</sup>	Triticale	Sorghum
Sheep	12.7-13.7	11.6-13.9	11.2-15.7	12.3-13.4	13.6-14.3
Cattle	12.2-13.1	12.2-13.2	10.8-13.4	12.9-13.2	10.2-13.2
Pigs	12.4-15.0	10.6-14.7	-	12.3-16.5	15.5-16.6
Broilers	12.4-15.6	11.2-13.7	12.6-14.6	11.0-14.6	15.2-16.5
Layers	13.7-17.1	11.0-14.8	12.7-16.4	11.6-14.4	15.5-16.3

<sup>1</sup>Metabolisable Energy for ruminants, Digestible Energy (DE) for Pigs and Apparent Metabolisable Energy for poultry; <sup>2</sup>Naked grain samples included

From these cross species observations, PGLP was able to identify potential reasons for these differences but more importantly develop Near Infra-Red Spectroscopy (NIRS) equations that facilitated rapid, quantitative estimates of faecal available energy content of parcels of grain. Although currently the feed industry still trades raw materials on the basis of a National Agricultural Commodity Marketers Association (NACMA) standard which bears no direct resemblance to the parameters of interest for nutritionists (Table 3), commercial release of these NIRS equations is occurring and should change the dynamic. Furthermore, the equations have also been given to plant breeders and are being used in their selection programs so the hope is that in the future, grains with more relevance to the feed industry will be grown. While the equation for faecal DE in pigs advanced the industry a long way, PGLP also found that grains could have high faecal DE, but were low in DE intake and vice versa. The Pork Cooperative Research Centre (Pork CRC) is continuing to invest in this technology and the industry looks forward to the release of robust equations for both parameters in the not too distant future.

**Table 3. Differences between a nutritionist's needs for a raw material and the National Agricultural Commodities Marketing Association (NACMA) standards**

What nutritionists formulate on:	What grains are traded on (NACMA Standards)
Energy – MJ (DE or ME for each species)	Being that grain
Available amino acids e.g. lysine, methionine, threonine	Protein (sometimes)
Fat and or fatty acid content	Moisture
Fibre	Test weight
Available minerals	Total Admixture
Phytate level	Foreign material
Pelletability indices	Screenings/ Trash
Palatability factors	Defective grains
	Seed contaminants
	Other contaminants

The PGLP also identified changes in grains under different processing conditions and these observations provide feed manufacturers with some significant opportunities to enhance the efficiency and effectiveness of the processes used in feed preparation. The rest of this paper discusses the processes involved in feed production and how these processes may positively and negatively interact with animal performance and how these contradictions may be addressed.



## Overview of feed manufacturing and pelleting

Most feed manufacturing processes, whether commercial or home mixed can be represented by the schematic in Figure 1. The essential components of all pig feed preparation are that raw materials are received in either bulk or bags, the grains are further processed by either grinding or rolling and the processed grains are then mixed with other raw materials, perhaps including liquid additions such as tallows, oils, molasses etc, and before being distributed to the pigs. In the majority of situations in Australia the feed is also pelleted. There is no reported figure of the proportion of mash: pelleted feed in Australia but the author estimates that at least 65-70% of the feed is pelleted. There are few recent Australian references on the advantages of pelleting over mash as the advantages seem to be well understood in the industry and most reports are anecdotal and consist of on-farm trials. However there are a number North American reports detailing the advantages of pelleting of stockfeeds such as Wondra *et al.* (1995), Behnke (1996) and Vande Ginste and De Schrijvert (1998). The reasons for the advantage of pellets over mash generally given include:

Pelleting improves feed conversion ratio (FCR) by 5-10% through:

- Increased nutrient availability due to starch gelatinisation and particle size reduction;
- Reduction in physical feed wastage; and
- Improved ration consistency.

Pelleting improves growth rate by 5-10% through:

- Increased feed intake resulting from the increase in bulk density of the feed;
- Decreased dustiness of the feed which improves palatability and respiratory health;
- Increased digestibility from the thermal processing; and
- Improved ration consistency.

Pelleting improves the physical form of the feed. The form of the feed remains the same independent of ration composition and therefore feed delivery systems and feeders do not have to be adjusted as frequently.

Pelleting provides some control over pathogenic micro-organisms, especially Salmonella.

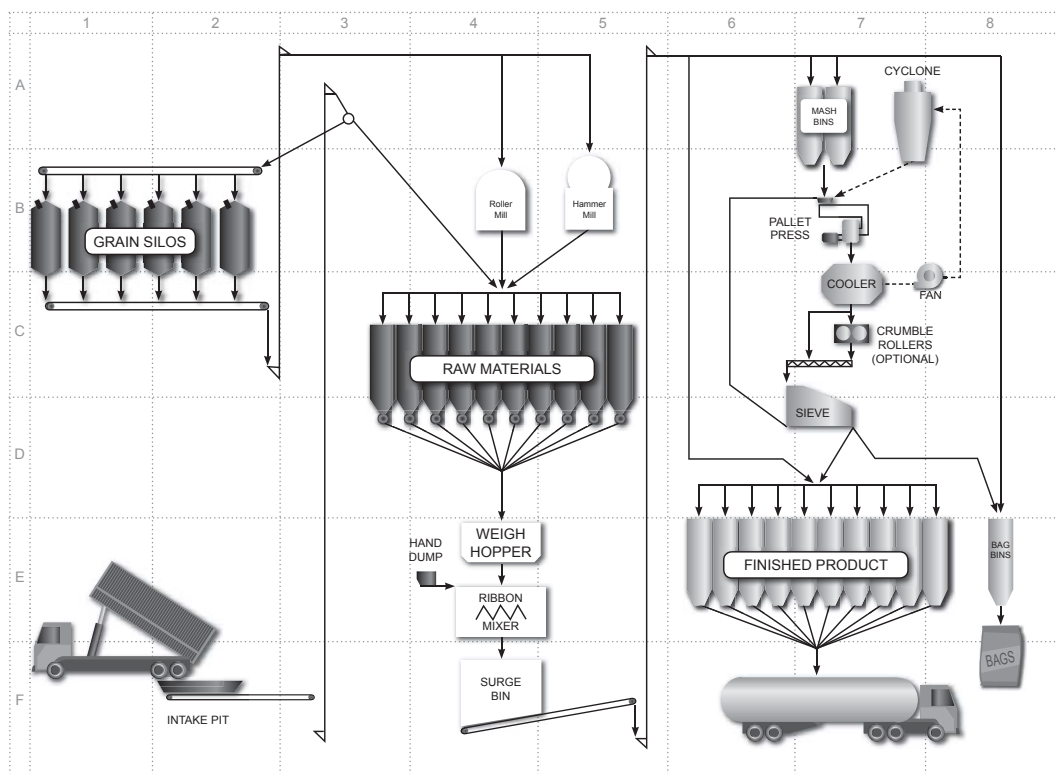


Figure 1. Schematic of feed manufacturing process

Pelleting a feed does not automatically guarantee all these benefits and it is well recognized by pig producers that a well prepared meal can result in better animal performance than a poorly presented pelleted diet. This observation has also been verified experimentally in the US by Schell and van Heugten (1998) who reported a significant linear depression in feed conversion efficiency (FCR) as fines increased from 2.5-40%. Stark *et al.* (1994) conducted a series of experiments with meal and high fines diets for weaning and finishing pigs. For weaners they observed that pelleting diets for nursery pigs improved gain:feed by 12-15% over the meal control and that fines concentrations of 25-30% decreased gain/feed by 3-4%. In finishing pigs where diets with up to 60% fines were fed, pellet fines did not affect ADG but gain/feed tended to decrease as the amount of fines increased.

Pelleting is a process that applies moisture, pressure and heat to a meal to condition or soften components of the ration such as the proteins and carbohydrates so that they can be compressed into a dense mass and shaped to conform to the die. There will be some increase in temperature of the meal as it moves through the pellet die due to frictional forces but most of the conditioning should occur before the die. When the heat and moisture is removed in the cooling process the pellet retains its shape and density and becomes durable enough to withstand transport and movement through feeder systems. The cooking or gelatinization of the carbohydrate is an important part of the pelleting process as it is the gelatinized carbohydrates that provide the glue which binds many of the other feed ingredients together. The temperature at which starch gelatinizes plays a key role in the effectiveness of pelleting and different raw materials have different gelatinization temperatures (Table 4). It is well known that sorghum and maize diets are more difficult to pellet and the data in Table 4 gives an insight into why this occurs. As this data refers to isolated starches, when it is considered that sorghum starch is bound in a protein matrix the temperature needed to achieve gelatinization in the normal feed environment is expected to be much greater. This means that more energy (= cost) would have to be expended in order to get the sorghum starch to the right temperature to achieve effective gelatinization compared to a wheat based diet.

**Table 4. Gelatinization temperatures of isolated various starches (from Black *et al.*, 2006)**

Grain	Gelatinization Temperature (°C)
Sorghum	74.5
Wheat	69.4
Maize	72.4

Conditioning is achieved using dry, saturated steam which imparts heat and moisture rapidly to the meal. Fine grinding of grain increases the surface area of the particles and therefore allows rapid transfer of heat; conversely larger particle sizes will not respond well and result in poor pellets. Additionally, if the particle size is too large, fracture points may develop which will lead to irregular pellet size and increased fines. It is generally considered by the industry that a mean particle size less than 600 µm is desirable for optimal pelleting.

The desired minimum pelleting temperature for most feeds in Australia is 85°C which gives good gelatinization and microbial control. As a general rule, when steam is applied, for each 1% of moisture in the steam the temperature of the meal is raised by about 14°C. However if the moisture of the meal goes above about 18% the pellet press will choke and the production run will have to be stopped, cleared and then restarted which interferes with operational efficiencies. The ideal moisture content of the meal in the conditioner is therefore 16-17%. Generally in pelleting, about 5% of moisture from steam is used. The moisture content of most grains is about 11% although sorghum is typically about 12.5% moisture. Therefore, if pelleting a wheat based meal and assuming a normal ambient temperature of 20°C, if 5% of moisture as steam is applied this is equivalent to raising the meal temperature by 70°C (5% moisture @ 14°C/% moisture = 70°C) and the meal will reach a final temperature of 90°C and an ideal moisture content of 16%. With a sorghum based meal under the same operating conditions, the moisture content would be 17.5% which is close to choking the press and yet from the data in Table 4 gelatinisation has probably not fully occurred. If the temperature of the grain is lower than 20°C as would be common in most Australian locations during winter, more heat will be required to achieve the same gelatinization.

The way a feed and ingredient will respond to the application of moisture, pressure and heat is dependent on seven factors: the protein content, density, fat content, fibre level, texture, carbohydrate content and moisture. For example, as one of the aims of pelleting is to increase the density of the feed, generally feed ingredients that are more dense to start with will pellet better, such as using soybean meal versus wheat bran. Also, as pelleting requires some degree of compression, some fat is useful in the ration as it provides lubrication. However, too much fat and the meal will slide easily through the die without proper pellet formation. A further complication, which is of concern for the feed miller and often not considered by the livestock producer, is the abrasiveness of the feed ingredients on the

equipment, especially the pellet die. When all of these factors are considered, most feed manufacturers have developed over the years a ranking for feed ingredients according to their pelletability and abrasiveness. In some instances, numerical values can be applied to these ranking and used within the linear programming model used to formulate the diets. Some indicative rankings for pelletability and abrasiveness for common ingredients are shown in Table 5.

**Table 5. Indicative pelletability and abrasiveness ranking for common feed ingredients (from Macbain, 1979)**

<b>Ingredient</b>	<b>Pelletability ranking</b>	<b>Abrasiveness ranking</b>
Barley	Medium	Medium
Blood meal	Low	Low
Distillers grains	Low	Med
Fishmeal	Med	Med
Tallow	Low	Low
Rice bran	Low	High
Soybean meal	High	Low
Wheat bran	Low	Low
Wheat	High	Low
Whey - dried	Low	Low

### Measuring pellet quality

As indicated above, good pellet formation is dependent on uniform particle size of the meal and effective gelatinization of the starch in the ingredients. Pellet quality is generally measured by Pellet Durability Index (PDI) and Hardness. A range of different apparatuses are available to measure PDI, a common one being the Holmen pellet tester (Borregaard Lignotech Ltd, Hull, UK). However, all of the apparatuses involve movement of a weighed amount of pellets for a prescribed period of time in a shaker mechanism designed to simulate damage the pellet would endure in transport, delivery and movement through the feeding system to the point of consumption by the pig. The damaged pellets are passed over an appropriate size sieve and the proportion of fines estimated. Hardness is measured on a single pellet using a spring loaded gauge and is designed to measure the individual breaking strain of a pellet. While generally hardness shows a positive correlation with PDI, it is possible to have hard pellets of low durability and vice versa. The PDI is the main quality indicator sought by livestock producers and it is difficult to recommend an industry standard PDI as pellet quality means different things to different producers. For example, each pig production system has different feed delivery systems which range from a chute directly above the feeder to many hundreds of metres of centreless augers or cable systems. Furthermore the feeder design itself can play an important role in how relevant PDI is. For example liquid feeding systems may favour less durable feed than producers using single-space, dry feeders. As will be discussed later, feed mills strive for the highest possible PDI within a given process. However there is generally a negative correlation between PDI and the economic drivers of a feed mill which are throughput rate and energy consumption. That is, maximal pellet quality can generally only be achieved at slower throughput rates and with higher energy use.

### Factors affecting pellet quality

Reimer (in Hancock, 1998) identified the importance of a number of factors that impact on pellet quality (Figure 2). There have been significant improvements in the design of pellet conditioners and pellet mills in recent years with manufacturers aiming to maximize the efficiency of the transfer of heat and moisture to the meal and improving energy efficiencies. However, pellet mills and conditioners are expensive capital items that have long service lives (there are many pellet mills in use that are over 30 years old) and therefore these new technologies are difficult to implement in Australia unless new feedmills are built or existing plants are upgraded. Consequently, the short to medium term focus for feed mills on improving pellet quality is through optimizing the process of particle size reduction and more effective characterization of the raw materials used in the formulation. This latter point was investigated within the PGLP and is being further developed within the Pork CRC. One study conducted by Ridley Agriproducts Pty Ltd in the PGLP utilized a test pellet mill that was capable of pelleting small (<5 kg) batches. A number of different grains that had been fully characterized within other investigations of PGLP were processed through the test peller to observe the effects of different grain type on pellet quality. The results of this study (Figure 3) show that wide differences in pellet durability exist between and within grains. These observations will be further explored within the Pork CRC.

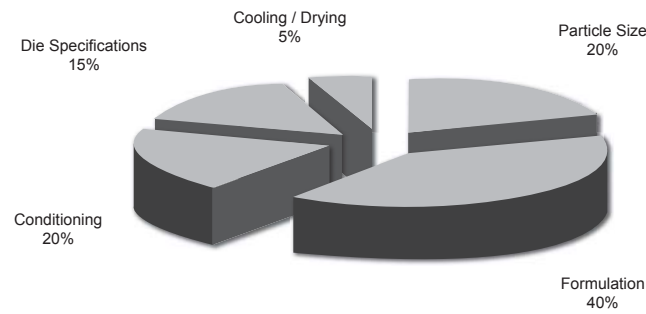


Figure 2. Factors contributing to pellet quality (from Reimer, 1992)

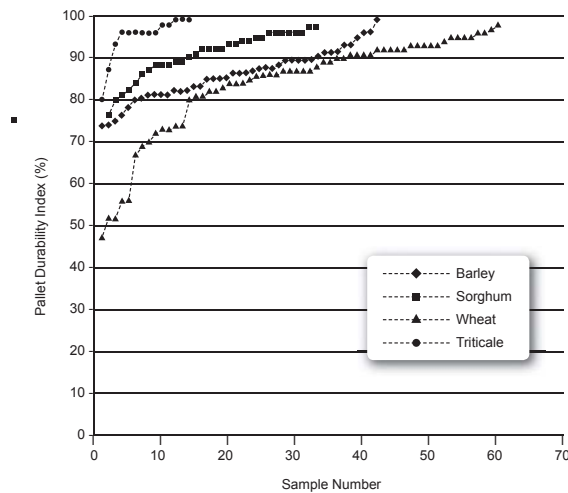


Figure 3. Range in pellet durability of different grains

There is a perception in some areas of the pig industry that ‘grains are grains’ and that a value in a database for a nutrient within a raw material is a true and repeatable representation of reality. The outcomes from PGLP have gone a long way to highlighting these concerns. There have been significant advances in our understanding of performance responses in pigs to different nutrients, as has been showcased at past APSA conferences and the application of pig modelling software such as AUSPIG (Black *et al.*, 1986). The challenge for the industry is to develop rapid and reliable means of estimating nutrient content in the wide variety of raw material in use in Australia (Table 1), and the effect that variability has on the whole pork production process, including feed manufacture. To further highlight how variable raw materials are, Figures 4 and Figures 5 indicate the variability that can be observed in a simple nutrient analyzed by wet chemistry in two raw materials used extensively in pork production. Samples were sourced by Ridley AgriProducts Pty Ltd from commercial suppliers and/or at mill receipt points.

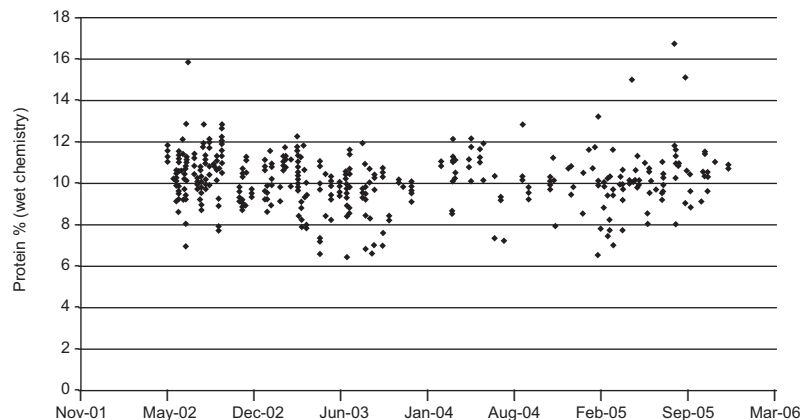
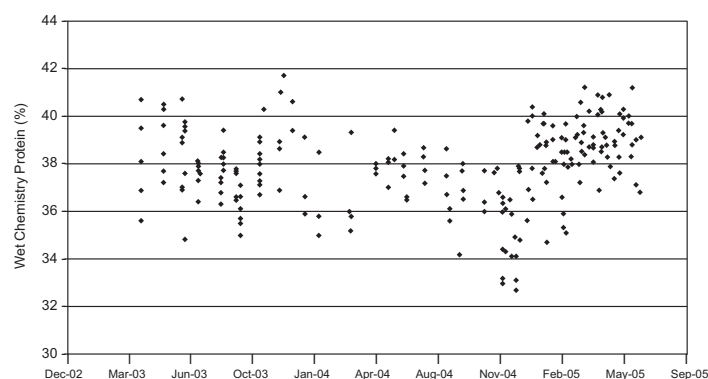


Figure 4. Variation in sorghum protein over time



**Figure 5.** *Variation in canola meal protein over time*

While the data presented (Figures 4 and 5) indicate that seasonal trends in protein can occur and therefore can be managed through appropriate testing, in both sorghum and canola meal the variation in protein within a week is as great as the variation observed within a year. The Australian Oilseed Federation (AOF) has recently completed a case study on canola meal variation (AOF, 2006) and outcomes from this work should go a long way to identifying the variation within and between oilseed crushing plants which will allow nutritionists to more accurately compensate for variation.

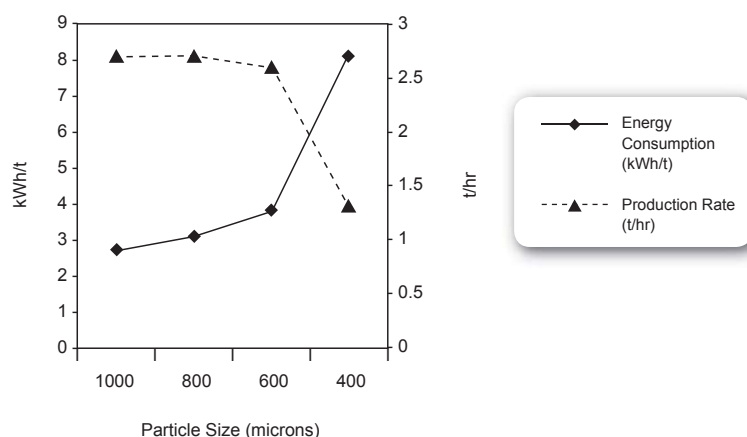
### Grain processing methods

It is well understood that some form of grain processing is required before grain can be fed to pigs. There are several types of grain processing available in Australia (Table 6).

**Table 6. Summary of the common grain processing conditions available in Australia**

Process	Brief explanation
Dry rolling	A process where grain is squashed between two metal cylinders or rolls. The gap between the rolls is adjustable for processing different sized grains and for achieving different coarseness of the end product. Limited heat is imparted to the grain and therefore limited gelatinization of the starch occurs.
Hammer milling	Size reduction is achieved by metal arms or 'hammers' rapidly revolving within a steel case which the grain is fed into. The action of the hammers grind the grain, pushing it through a metal screen. The size of the grind can be adjusted by choosing different sized screens. Some heating occurs which may cause slight gelatinization.
Steam pelleting	This is the most common form of heat processing of grains fed to pigs in Australia. It involves conditioning or softening the complete mixed feed with steam and then using a series of rollers to push the meal through holes in a pellet die to shape the pellet. Meal temperatures can be up to about 95oC and gelatinization is significant.
Steam flaking	Most commonly used in cattle feeding, this process involves treating whole grains in a sealed chest for a short period of time and then rolling or 'flaking' the product between two rolls to obtain a thin product. Grain temperature can be up to about 115°C and gelatinization is significant.
Expanding	Often used in combination with pelleting, this process involves conveying the meal quickly along a barrel with resistance applied at the outlet. The resistance causes pressure and shearing and in combination with steam, the meal can reach temperatures up to about 140oC. After the meal leaves the outlet, the pressure rapidly returns to atmospheric which causes the feed to expand in volume and the temperature to drop. Gelatinization can be complete.
Extrusion	This method is commonly used in the preparation of aquaculture and pet foods and in processing ingredients for subsequent use in stock feeds such as grains and protein sources. Similar to expansion, in this process heat and pressure are achieved by passing the meal through a barrel with increasing restrictions. As the meal passes through the restriction 'expansion' of the meal also occurs. The meal can reach temperatures up to about 180oC and gelatinization is complete.

All of these grain processing techniques rely on energy (gas and/or electricity). The cost of the process is governed by 1) the energy required to process the grain and 2) the throughput rate. This relationship was clearly shown by Wondra (1995) and reproduced in Figure 7 in relation to hammer milling corn. Energy consumption and production rate were not severely impacted until grinding to achieve a particle size  $<800\ \mu\text{m}$  was required. As mentioned previously, a particle size  $<600\ \mu\text{m}$  is desirable for pelleting. Processes such as extrusion and micronizing require great amounts of energy and have low throughput rates and as a consequence are often uneconomic for use in general feed manufacturing.



**Figure 7.** Relationship between desired grind size and energy consumption (kWh/t) and production rate (t/h)

Each of the processes described in Table 6 results in a different amount of gelatinization. Black *et al* (2006) reported on studies where the effects of different gelatinization processes on sorghum were conducted (Table 7).

**Table 7. Effects of different processing methods on the *in vitro* enzyme digestion of starch (from Black *et al.*, 2006)**

Treatment	<i>In vitro</i> enzyme digestion of starch (% starch)
Unprocessed normal sorghum	23-43
Unprocessed waxy sorghum	42-56
Fine grinding (1 mm screen)	42
Protein extraction (58% protein removed)	49
Urea ensiling (5 months)	43
Steeping (24 hr to 30% moisture)	27
Steeping and 21 day anaerobic ensiling	27
Germination (5 days)	35
Germination + 16 d anaerobic ensiling	43
Steam flaking	70-84
Grinding and steam pelleting	54-64
Extrusion	89-94
Microwaving	70-82
Cooking (5-10 mins $> 85^{\circ}\text{C}$ )	90

While there are clear differences in the ability of a process to affect the digestion of starch *in vitro*, the limited *in vivo* studies in pigs to date and wide range of processing techniques involved have not been able to identify a benefit and/or economic justification for processing sorghum above pelleting temperatures (J. Black: pers. comm.). There is a clear financial benefit to be derived for the pig industry if the digestibility of energy from grains can be enhanced and this work is being continued within the Pork CRC. In case studies reported by Spragg (2007), depending on the raw material base available, an extra MJ of energy for pigs can be worth between \$5-15/t of finished feed. While the future outcome may be that grain processing *per se* may be limited in its ability to improve energy utilization, the studies are providing significant amounts of information on what is happening at the cellular level due to processing. This may provide opportunities for exogenous treatments to be applied in feed manufacturing such as the use of enzymes and or surfactants.

The pig industry has gained a greater understanding in recent years of the influence of non-starch polysaccharides (NSP) in cereals when fed to pigs. The content of NSP in cereal grains is shown in Table 8 (M. Choct: pers. comm.) and it is clear that arabino-xylans and b-glucans are the primary NSP's of concern. Fortunately enzyme manufacturers have been able to develop cost effective b-glucanases and xylanases to assist in breaking down these NSP to the point where it is now routine to include these enzymes or mixtures into pig diets. The challenge for the feed manufacturer and nutritionist is to be able to rapidly identify the level of NSP in raw materials. They must assess whether an enzyme response would be achieved in both the feed prior to processing as well as the pig, what is the appropriate enzyme or mixture to use and when in the feed manufacturing process it should be added.

**Table 8. Non-starch polysaccharide content of selected cereals (% dry matter basis) (M. Choct: pers. comm.)**

Cereal	Arabino-xylan	$\beta$ -Glucan	Cellulose	Others	Total
Wheat	8.1	0.8	2	0.5	11.4
Barley	7.9	4.3	3.9	0.6	16.7
Rye	8.9	2	1.5	0.8	13.2
Triticale	10.8	1.7	2.5	1.5	16.3
Sorghum	2.1	0.2	2.2	0.3	4.8
Corn	5.2	0	2.5	0.3	8.1

### Aligning feed manufacturing processes with animal performance

The previous discussion has highlighted the exceptional level of understanding pig nutritionists and feed manufacturers have about their respective processes. As with any process there are Key Performance Indicators (KPIs) which allow the level of success to be monitored and improved. Readers of APSA proceedings are generally well versed in the KPIs for the pig industry that relate to growth and feed conversion efficiency or reproductive performance. They are probably less familiar with the KPIs for the feed manufacturing sector (Table 9). As with the drivers of grain processing and pelleting, the key drivers in feed manufacture are related to volume, energy use and throughput. While the energy required to produce feed is a variable cost and can be calculated for a given feed, production rate largely affects fixed costs. If production rate is decreased, all the fixed costs such as repairs, maintenance and depreciation are spread over fewer tonnes and the cost of feed increases. If there is no spare production capacity and production rate is significantly slowed existing feed orders may not be able to be met and the growth of the feed manufacturing business is constrained. For business with spare production capacity, there may be no detrimental effect on supply of existing feed orders. However, increased labour costs through overtime allowances may be incurred and growth of the feed business may again be constrained.

**Table 9. Typical key performance indicators likely to be used in feed manufacturing**

Tonnes produced – mash	Average pellet durability
Tonnes produced - pellets	Ordinary mill man hours worked
Total tonnes produced	Overtime mill man hours worked
Production hours - pelleting	% overtime/ordinary hours
Production hours total	Sick leave hours
Total man hours - manufacturing	Compensation hours
Total man hours - supervision & other	Rework (tonnes)
Total man hours	Maintenance hours O/T
Downtime - planned	Tonnes/hour overall
Downtime - unplanned	Tonnes/hour pelleted
% downtime	Overall kWh/tonne
kWh consumed for week	Man hours per tonne - overall
Gas consumed for week	Man hours per tonne - manufacturing
Total production runs per week	Man hours per tonne - supervision
Formula changes per hour	% pellets
Average run time in minutes	% mash
Average run size (batches)	Tonnes/hour/kW installed
Tonnes per die	

Balancing the number of diets required for a piggery and the feed formulation needs as directed by the nutritionist, overlaid with the requirements for medications and other additives as directed by veterinary surgeons and interspersed with meeting the requirements for load size and frequency requested by the freight company responsible for feed delivery represents some unique challenges for the feed manufacturer. Feed manufacturers are also part of a larger supply chain involving delivery of raw materials from both domestic and international origins and they are subjected to occasional failures in these supply chains as well as unscheduled mill closures due to breakdowns. This can create problems downstream. While the cost of feed per tonne is a significant input into pork production, the majority of the cost of the feed is in raw material charges. Accordingly, the feedmill manager has to balance inventory to improve working capital without constraining feed production.

Recognition of the benefits that phase feeding and or separate sex feeding brings to the efficiency of pork production has led to an increase in the number of diets an individual farrow-to-finish operation requires. Table 10 identifies the typical feed requirements for different size piggeries adopting a relatively basic phase feeding program. With the restructuring of the pig industry in recent years, there are few properties where all pigs are housed on the one site with each class of pig being fed out of a single silo. With multi-shed production, producers have also been able to target medication programs and in some cases different marketing options which require different additives in their feeds. In a feed mill environment, it is the total feed ration, with medication and additives included, that is considered to be the feed for scheduling, clean-out and run size determination. However, many producers consider the product code or feed name to be the feed. For example, a feed called Smith Grower, with no medication needs to be batched and processed completely separate to Smith Grower with 1 kg/t of medication. This reduces run size and mill production efficiencies, particularly on the issue of sequencing (Behnke, 2007: this symposium).

**Table 10. Typical weekly feed requirements for 100 and 1000 sow piggeries on a basic phase feeding program**

Feed type	% of total feed needs	Weekly feed requirement/100 sows	Weekly feed requirement/1000 sows
Starter	3	0.3	3
Weaner	10	1	10
Grower	30	3	30
Finisher	35	3.5	35
Lactation	11	1.1	11
Gestation	11	1.1	11
Total	100	10	100

Creep, starter and weaner feeds generally pose the most problems for feed manufacturers in terms of run size and throughput. As there is an absolute requirement for these feeds to be highly digestible and palatable these feeds are manufactured using milk based products and other raw materials that can be damaged at high temperatures. Consequently, they are pelleted at lower temperatures and because they should be used as fresh as possible, they are ordered in small run sizes.

From this brief scenario it is obvious how on-farm production needs can inadvertently complicate feed manufacturing efficiencies, particularly for producers with smaller pig numbers. While the vet and nutritionist recognize these limitations and work with the feed miller to improve efficiencies, feed orders are controlled by the piggery manager in relation to daily on-farm needs. In many cases, neither the vet nor nutritionist is aware of daily operations of the piggery. The formation of producer groups by geographic region and/or pig genetic base, where a number of producers band together to increase their collective buying power for feed and piggery consumables, has led to increased efficiencies for feed manufacturers and resulted in lower feed prices for the group. Some of these groups have also capitalized on their pig marketing options through economies of scale. Other opportunities for increasing run size in feed mills to increase mill efficiencies and reduce feed costs include utilizing blend feeding systems. This involves blending two or more feeds on farm to provide the phase feeding program and the use of water medicators to apply some medications and additives on farm.

The other major area where conflict can occur between the nutritional demands of the pig and the feed manufacturer is in the area of balancing least cost or optimal cost formulations with pelletability requirements. As pelletability effects can not be as easily incorporated into linear programming formulation programs as, for example, available amino acid ratios or DE, constraints such as maximum added fat and minimum wheat content are applied to raw materials. In most cases these constraints do not add significantly to the cost of the diet and the improved performance from better quality pellets can easily justify the constraint. However, in cases such as utilizing output



from AUSPIG, when raw materials are in particularly short supply or purchasing contracts dictate raw material usages, these constraints can become significant.

## **Conclusion**

From the above discussion it should be clear that there has been significant progress in recent years on the level of understanding of raw material composition and the subsequent effects on animal performance and feed manufacturing. Programs such as PGLP and those funded by the Pork CRC will continue to build on industry knowledge and will modify how raw materials are assessed and used in the near future. Any activity that will reduce the variability in composition of raw materials, or improve our knowledge of predicting this variability, has the potential to significantly improve the efficiency of feed manufacture and ultimately animal performance. The paper identified the different types of processing available to feed manufacturers and the effect these processes can have on feed and pellet quality. Unfortunately the capital cost of installing this equipment means that it is difficult for the industry to adapt quickly to developments in equipment technology. Accordingly, the opportunities for the feed industry largely lie in the short term to developments in enzyme technology and other additives into the feed. Near Infra-Red Spectroscopy will also play an increasing part in prediction and monitoring of responses to different treatments. Finally, the competing interests of the animal producer and the feed manufacturer were exposed. For lowest cost and efficient pork production it will be imperative that feed manufacturers, nutritionists, veterinarians and piggery managers maintain an open dialogue on their activities so that each part of the supply chain can contribute in a way that ensures profitability and sustainability.

## Feed milling symposium: conclusion

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In most pork producing countries and especially in Australia, it is well the most significant component of the costs of production is feed, accounting for about 65% of the costs (Australian Pork Limited, 2006). The livestock industries are facing increasing competition for raw materials from both the demand for human food and the demand for seed stock for biofuel production. There is also increasing recognition of the role the whole meat supply chain plays in food safety. Along with this recognition comes increased regulations and requirements on individual sectors of the supply chain. Recognition and addressing of these issues by the pork supply chain will ensure that the industry will be profitable and sustainable.

Behnke (2007) expanded on the practicalities of using flushing and sequencing procedures to reduce cross transference in feed manufacturing for both on-farm and commercial feed manufacturers. Application of these simple processes by pig feed manufacturers will greatly assist in maintaining pork as the safe and wholesome food that it is recognized to be. As prices of raw material used in pig feeds are likely to remain high, it is imperative that pig feeds are mixed effectively so that they are utilized as effectively as possible by the pigs. Again, there are significant benefits for the industry if the marker techniques for monitoring mixer efficiency described by Behnke (2007) are routinely applied in feed manufacturing operations, both on-farm and commercially.

Zilstra and Beltranena (2007) identified that use of co-products in the feed industry is likely to increase in the future and understanding the characteristics of these co-products in the feed milling environment and when they are digested by the pig will be crucial to successfully utilizing the co-products in pork production. Research outcomes from programs conducted within the PGLP and Pork CRC discussed by Gannon (2007), especially the role that NIRS can play will be vital in being able to assess the value of co-products in the Australasian pork sector. The insight into feed manufacturing processes and the drivers of efficiency, particularly those of pelleting operations, as described by Gannon (2007) provide a number of worthwhile opportunities that should be further explored by feed manufacturers, veterinarians additive manufacturers and pig producers alike to improve the overall profitability and sustainability of each sector.

The biennial APSA conferences have gained a well-earned reputation for addressing issues and opportunities for pork production and questioning and challenging common beliefs. This symposium continued this ideal and the ideas presented in this symposium will continue to challenge all those in the pig industry and provide a number of important areas warranting further research.

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