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## Lunch and Refreshments at Breaks

Mayberry Cafe, Danville, IN; Brad and Christine Born

## **Pre-Conference BBQ Dinner**

Shoup's Catering

## **Schedule of Presentations**

- 8:15 Registration
- 9:00 Welcome Dennis Liptrap, Ralco Nutrition
- 9:05 Identifying, prioritizing, and conducting research on nutritional challenges in a Midwest swine production system. *Omarh F. Mendoza, The Maschhoffs, LLC, Carlyle, IL*
- 9:50 Estimates of requirements of digestible Ca by growing pigs. Hans H. Stein, University of Illinois
- 10:20 Break
- 10:50 Mycotoxins from corn screenings, effects on diet preference and performance and mitigation strategies. *Merlin Lindemann, University of Kentucky*
- 11:25 Understanding the growth and health benefits of supplemental treated wheat straw in the weaned pig diet. *Dale W. Rozeboom, Michigan State University*
- 12:00 Lunch
- 1:10 Nutritional strategies for coping with high diet cost for grow-finish pigs. *Aaron Gaines, Ani-Tek Group, Shelbina MO*
- 1:55 Advantages of higher soybean meal diets for pigs. *Su A Lee, University of Illinois*
- 2:30 Break
- 2:50 Peroxidized lipids increase catabolism of amino acids and may induce additional protein damage. *Pedro Urriola, University of Minnesota*
- 3:25 Effects of fiber and plane of nutrition on the working boar. *Brian Richert, Purdue University*
- 4:00 Closing Remarks

### Midwest Swine Nutrition Conference

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# Identifying, Prioritizing, and Conducting Research on Nutritional Challenges in a Midwest Swine Production System

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### Summary

Pork production operations that integrate research and innovation within the organization will create a competitive advantage. However, sustainable innovation requires a significant commitment across multiple levels of the organization, with the application of principles of the scientific method at the center, while conducting research at scale that is pertinent to the challenges of the operation. The process of gathering and selection of ideas, design and execution of experiments, and interpretation of results, will require a multi-step process with clear ownership and objectives for information to be translatable to the business.

### Introduction

Any livestock production system such as an integrated (or semi-integrated) pork production operation is indeed a biological system with many different parts that are to work in concert to achieve the objectives of the operation. At its foundation, there is a necessity to understand and embrace the biological disciplines and economic relationships that undergird an operation's objectives, to remain competitive on a global scale (Peterson, 2012). With most of the production processes in a pork operation, there are still opportunities for progress, that is, to improve output, increase efficiency, reduce cost, etc. For that progress to be effective, it needs to be organized, sustainable, and continuous, and this is where the application of research and innovation processes need to be integrated within the framework of the operation, while centered on the application of the principles of the scientific method at scale.

### The Stage-Gate Innovation Process

Any organization will have a multitude of ideas for innovation, but there needs to be a process to sort out which ideas will lead to effective and continuous progress and those ideas that are not viable. Consequently, the ideas that are selected for research need to be filtered through a defined set of steps that are in alignment with the operation's objectives. A stage-gate process can be useful for managing projects and controlling when and how innovation impacts business operations. However, important to the success of the process is the commitment by a selected group of individuals that are to be the team or committee responsible for ownership of the process and its execution. This team needs to be formed by individuals across distinct functions in the organization, to achieve garnering perspectives across a range of biological disciplines (e.g., nutrition, health, genetics, reproduction, management, etc.) to ensure a multidisciplinary approach to innovation.

An example of the application of the stage-gate process by the team can be seen as illustrated in Figure 1. In this case, there are a total of five stages, i.e., idea prioritization, research activity, business case, implementation, and posthoc analysis with four gates The number of stages and gates can be adjusted to the size and scope of the research process for a given operation, however the essence of it, is to foster the team or committee to select the ideas that have viability and can make an impact to the business, in addition to make "go" or "no go" decisions on innovation projects (Shull, 2022).

### **Research Ideas Generation And Prioritization**

It is important that ideas for research are being captured across various levels of the organization; however, the use of guidelines or innovation themes that are in direct relationship with the operation's objectives will make this process more efficient. Some examples of these themes are feed cost improvement, enhancing early pig survival and improvement in sow reproductive performance, etc. While the in-

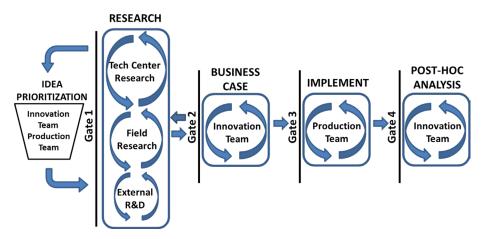


Figure 1. Example of stage-gate process for managing a swine innovation program.

novation themes may change over time, they provide clarity about the scope for the ideas to flow into the first stage of the process.

The process of capturing innovation ideas (idea funnel) and prioritizing them according to a set of criteria and timeline (i.e., short versus long term) may be the most crucial step in the innovation process. This will produce a balanced innovation portfolio that will match the operation's resources (human resources, budget, and infrastructure, etc.). In addition to the ideas that are generated from within the organization, key partnerships with academic institutions and private industry will complement the idea funnel. The more connected an innovation team is to the production team and external resources, the better an organization will be at discovering innovation opportunities.

Once ideas are collected, one approach that proves to be useful is to use a prioritization matrix with a defined set of criteria that allows the ranking of the different ideas in a quantitative manner. However, every organization will need to define the criteria, as it may be different according to their specific operation, objectives, and capabilities. The following section outlines a list of key factors that can be used as criteria with associated considerations.

### **Priority Matrix**

The anticipated value proposition of the idea will be the first step in this ranking process. For example, application of a technology may result in pig performance improvement, or in cost reduction at the same level of performance. It is recommended that each group ranking the ideas uses the same input and output values to avoid confounding due to market conditions. Arriving at an anticipated value proposition is much easier for innovation opportunities that have empirical data, whether internal or external; however, having a defined mode of action for ideas with limited data provides insight into the ability of an idea to affect a given outcome. The team will then use this information to assess the probability of success of the idea, and it is also important to consider the intangible value drivers as well.

Assessing the risk profile of a technology necessitates some considerations, for example technologies that are high cost are inherently higher risk compared to those that add minimal cost to an organization. Nevertheless. the implementation difficulty of an idea must not be overlooked; for example, changing processes that are managed by people requires training and introduces significant risk of achieving the full value proposition as a result of variation in execution,

but technologies that can be implemented in the background (e.g. feed additive) are not dependent on execution for its success. It is noteworthy to mention that the consistency and repeatability of a technology also determines its risk profile.

Another aspect to consider when prioritizing ideas is the time horizon. While it is natural to prioritize ideas that offer a quick turnaround, the impact of an innovation program may be limited if it is only focused on short-term projects. Oftentimes, a multi-year initiative may be needed to innovate a specific area or problem. Thus, it is important to find the right balance of short and long-term projects that meet the needs of an organization.

### **Commercial Scale Applied Nutritional Research**

Nursery pigs in commercial operations undergo a series of stressful events, such as weaning, commingling, transport, environmental, etc., which may cause nutritional fall out pigs and can result in increases in morbidity and mortality. It is now accepted that it is no longer sufficient to design a nursery nutrition program that focuses only on growth performance, but in addition, one that improves the probability of survival in the nursery (Mendoza and Shull, 2022). Nevertheless, there is a growing need for large scale practical research that evaluates nutritional technologies and strategies, and their effect on morbidity and mortality.

**Table 1.** Number of pigs per treatment required to detect a difference in mortality

Mortality difference between treatments	Number of pigs per treatment required
3.5% - 3.0% = 0.5% difference	9568
4.0% - 3.0% = 1.0% difference	2592
5.0% - 3.0% = 2.0% difference	735

 $^1Generated$  using a chi-square analysis and assumes a significance level of  $\alpha=0.05.$ 

**Table 2.** Least-squares means for the effect of a nutrient dense liquid (NDL) on growth performance and morbidity and mortality of pigs during the nursery period (Schmitt et al., 2018).

	Treatment		_	
	Control	NDL	SEM	P-value
Number of pens	64	64	-	-
Number of pigs	4613	4602 -		-
Growth performance, overall				
Average daily gain, lb	0.399	0.404	0.015	0.15
Average daily feed intake, lb	0.608	0.612	0.029	0.14
Gain:Feed	0.661	0.662	0.008	0.76
Morbidity and mortality, %	4.29a	3.35b	-	0.02

a,bMeans within a row with different superscripts are different ( $P \le 0.05$ ).

**Table 3**. Least-squares means for the effects of lysine level in the nursery period on weanto-finish pig growth performance (Tolosa et al., 2020).

	SID Lysine level <sup>1</sup>			
Item	Restricted	Control	Excess	SEM
Number of pens	33	33	33	-
Nursery				
Start weight, kg	5.8	5.8	5.8	0.05
End weight, kg	36.8 <sup>b</sup>	38.5 <sup>a</sup>	39.1 <sup>a</sup>	0.53
Average daily gain, kg	0.50 <sup>b</sup>	0.53 <sup>a</sup>	0.54 <sup>a</sup>	0.007
Average daily feed intake, kg	0.84	0.84	0.85	0.014
Gain:Feed	0.600 <sup>b</sup>	0.634a	0.630a	0.005
Wean-to-Finish				
Average daily gain, kg	0.78	0.78	0.78	0.006
Average daily feed intake, kg	1.75	1.74	1.76	0.018
Gain:Feed	0.445	0.443	0.445	0.003

a,bWithin a row, means without a common superscript differ at  $P \le 0.05$ .

<sup>1</sup> Control: SID Lys at estimated requirement in nursery diets; Restricted: 0.2 g/kg SID Lys below Control; Excess 0.1 g/kg SID Lys above Control.

The context of commercial practice needs to be considered for research results to be more applicable. Examples of this context include practical dietary programs that are akin to those fed to commercial pigs. For example, in nursery pig research, diets will need to include antibiotics, pharmacological levels of trace minerals, (e.g., Zinc and Copper) and other nutritional technologies when in use, to reflect practical diets.

Additional considerations for the research model need to include factors like having the appropriate statistical power to detect morbidity and mortality differences. For example, conducting an experiment with a nutritional intervention that may produce a one percentage unit change in mortality, e.g., from 4 to 3%, will require approximately 2,600 pigs per treatment; however, if the difference to detect is 0.50% units in mortality, that number of pigs per treatment approximates 10,000 pigs per treatment (Table 1). A study conducted by Schmitt et al. (2018) provides an example of a large-scale study that evaluated the effect of a nutritional intervention (nutrient dense liquid [NDL] consisting of an electrolyte-based solution). As shown in Table 2, overall growth performance (ADG, ADFI and G:F) was similar between treatments, however, pigs administered the NDL had reduced morbidity and mortality (0.94% unit change; P < 0.05) compared to pigs that were not supplemented.

For a pig production system that operates on a wean-to-market cycle, an important aspect in conducting research studies during the nursery period is to be able to evaluate the impact of a nutritional intervention over the entire growth period of the pigs up until the finishing stage. This is critical information that informs the net economic value of the idea or technology. For example, in a research study conducted by Tolosa et al. (2020; Table 3), the performance of pigs from wean-to-finish was evaluated, when a nutritional intervention (SID Lys level) was applied during the nursery period. In this study, there were growth performance differences during the nursery period (Table 3), however, when pigs were reared to finishing, the differences disappeared, and growth performance was similar between treatments. Therefore, when inter-

preting data that does not cover the entirety of the growth period, additional evidence needs to be considered before conclusions are made relative to overall biological and economical impact.

Similarly, as mentioned above, the question whether there will be enough observations to establish validity of the research hypothesis, as discussed by Aaron and Hays (2004) is one that is not to be taken lightly. The authors in this paper state that "although investigators often seem to choose replication arbitrarily on the basis of cost or availability of animals, housing considerations, convenience, or tradition, the question of how much replication is necessary, is a statistical one that has a statistical answer". It is therefore that power of test calculations are a requirement in the planning and design of any experiment; however, it is common to see experiments that are underpowered and do not have adequate replication. As an example, Table 4 shows the estimated number of replicates that are needed to detect differences (percent change relative to a control) in carcass gain:feed as calculated for a particular research farm. In this example, the sample size for the experiment to be able to detect a 1.6% difference in carcass gain:feed, will need to be an estimated 39 replicates per treatment (alpha level of 0.05, and Power of 0.80). The ability to execute research protocols with this statistical power requires significant investment in research infrastructure but is necessary for the research results to be translatable to a production system.

As an illustration, research conducted by Schmitt et al. (Table 5; Exp 2., 2017) conducted in the same research facility that was used to build Table 4 shows the effect of feeding a feed additive on the growth performance of finishing pigs. The results of this study show that pigs fed the nutritional technology had a 1.85% greater (P < 0.05) carcass gain:feed compared to pigs fed the control diet. Therefore, the design of this experiment was adequate to be able to detect differences in carcass gain:feed.

Other key factors when conducting applied research include repeating the research with distinct groups of pigs to understand the distribution of outcomes. For example, conducting experiments with the same nutritional technology with different groups of pigs, at different seasons of the year and with different health statuses will help inform the repeatability and variability in the response variable, allowing the researcher to quantify its value more accurately.

One of the areas that has the greatest opportunity for nutritional research is in sows. Notwithstanding, there are several important obstacles that need to be overcome for a successful sow research program, and this includes the appropriate infrastructure, labor, time, and commitment. For example, conventional sow farms are not typically equipped to feed more than one diet during the gestation or lactation periods and, therefore, conducting research by feeding multiple diets is not practical and, in addition, the timeline is longer compared to research with growing-finishing pigs. Regarding adequate number of observations for sow research (e.g., lactation), as an example, Table 6 shows the estimated sample size (sows per treatment group) that is required to detect differences in the feed intake of sows. For example, if the difference that is desired to be detected is 0.23 or 0.45 kg per day, the required number of sows is estimated to be 446 or 112, respectively. Most research institutions and organizations do not have the facilities to conduct experiments with these types of sample sizes, making it difficult to conduct research in this area.

Finally, the process of manufacturing the feed that will be used as dietary treatments for nutrition research projects at commercial scale is of similar importance. This includes the execution of current good manufacturing practices that encompass ingredient processing (i.e., grain grinding),

**Table 4.** Estimated number of replications per treatment to detect differences in carcass gain:feed

	3	
Carcass Gain:Feed,	α=0.05	α=0.10
% difference	(1-β)=0.80	(1-β)=0.80
0.50%	341	269
1.00%	86	68
1.60%	39	31
2.10%	23	18
2.60%	15	12

**Table 5.** Least squares-means for the effect of feeding Ambitine Feed Technology on growth performance and carcass characteristics of finishing pigs (Schmitt et al., Exp. 2, 2017).

	Treat	ment		
ltem	Control	Ambi- tine	SEM	P-value
Number of pens	68	68	-	-
Number of pigs	2,335	2,347	-	-
Growth performance				
Live weight, kg				
Start of test	89.4	89.27	0.277	0.44
End of test	121.84	122.02	0.508	0.63
Overall performance				
Carcass average daily gain, kg <sup>1</sup>	0.56	0.58	0.096	0.15
Average daily feed intake, kg	2.09	2.09	0.024	0.96
Carcass Gain:Feed <sup>2</sup>	0.271 <sup>b</sup>	0.276 <sup>a</sup>	0.002	0.03

a,b Means within a row without common superscripts are different ( $P \le 0.05$ ).

<sup>1</sup>Carcass average daily gain = Overall Average Daily Gain × Carcass yield.

<sup>2</sup>Carcass gain:feed = Carcass Average Daily Gain / Overall Average Daily Feed Intake.

<b>Table 6</b> . Sample size required to detect lactation feed intake dif-
ferences (α=0.05)

ADFI Difference Detected, kg (lb)	Number of Sows per Treatment Required
0.11 (0.25)	1782
0.23 (0.50)	446
0.34 (0.75)	199
0.45 (1.00)	112

Note: Power = 0.8; Lactation Daily Intake standard deviation = 1.36 kg; 1-sided test.

feed processing (i.e., conditioning, pelleting, cooling), and transport and delivery is crucial, but also include elements of quality control for feedstuffs selection, chemical characterization, and nutrient loading values. In addition, the addition of research diet formulas to a commercial feed mill, specifically if the feed mill is producing pelleted feed, may lead to considerable downtime due to formula changeovers, therefore being disruptive to the mill and potentially hindering future collaboration. Therefore, this is another factor that needs careful consideration when designing nutritional research studies.

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## Estimates of Requirements for Digestible Ca by Growing Pigs

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### Summary

The standardized total tract digestibility (STTD) of Ca in all major feed ingredients used in diets for pigs has been determined and the positive impact of microbial phytase on STTD of Ca has also been demonstrated. The requirement for STTD Ca by all groups of growing-finishing pigs has also been reported and it is, therefore, possible to formulate diets for growing-finishing pigs, either without or with microbial phytase, based on values for STTD Ca rather than total Ca. Because of the tight relationship between the needs for STTD Ca and STTD P, it is likely that the ratio between STTD Ca and STTD P is more important than the absolute values, but values for the ratio needed to maximize growth performance changes during the growing-finishing period. To maximize growth performance of 11 to 25 kg pigs, a ratio between STTD Ca and STTD P of less than 1.39:1 is required, whereas a ratio of less than 1.10:1 is required for pigs from 100 to 130 kg. However, to maximize bone ash in pigs, greater ratios are needed. In conclusion, results of research published over the last decade have provided information needed to formulate diets for growing-finishing pigs based on requirements for STTD Ca.

### Introduction

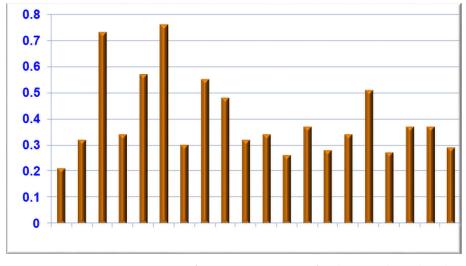
Current requirements for Ca in diets for growing pigs are based on requirements for total Ca whereas requirements for P are expressed as the requirement for standardized total tract digestibility (STTD) of P (NRC, 2012). The reason for this difference is that at the time the NRC (2012) requirements were established, no values for the STTD of Ca in feed ingredients used in diets for pigs were available. However, because of differences in digestibility of Ca among feed ingredients it was acknowledged that the accuracy of diet formulation would be improved if diets could be formulated based on STTD of Ca rather than total Ca (NRC, 2012). As a consequence, over the last decade, values for the STTD of Ca by growing pigs in most of the commonly used feed ingredients have been published and it is, therefore, now possible to formulate diets based on STTD Ca rather than total Ca. The widespread use of microbial phytase in diets for pigs has increased the need for formulation of diets based on STTD Ca rather than total Ca because phytase in addition to releasing P also releases Ca from phytate, and therefore, increases the STTD of Ca (Gonzalez-Vega et al., 2013; 2015a; 2015b; Lagos et al., 2022). Microbial phytase also reduces the endogenous losses of Ca from pigs by

preventing binding of endogenous Ca to phytate (Lee et al., 2019a), which further increases the STTD of Ca, and therefore, increases the need for formulating diets based on STTD Ca rather than total Ca.

Severe negative effects of excess Ca in diets for growing pigs have been demonstrated repeatedly (Gonzalez-Vega et al., 2016a; 2016b; Merriman et al., 2017; Lagos et al., 2019a; 2019b). It is likely that the negative effects of Ca are partly a result of binding of Ca to P in the digestive tract of pigs resulting in formation of un-digestible Ca-P complexes, which reduces digestibility of P. As a consequence, excess dietary Ca may create a P-deficiency even if the required concentration of digestible P is included in the diet (Stein et al., 2011; Lee et al., 2020). Unfortunately, commercial diets for pigs in the United States on average contain almost 0.20% more Ca than believed by formulating nutritionists (Lagos et al., 2023), which is likely due to a lack of knowledge about the concentration of Ca in all ingredients in the diets. An excess of 0.20% Ca will likely result in a reduction of average daily gain of at least 50 to 60 grams per day (Merriman et al., 2017). To avoid excess Ca in diets, it is necessary that the concentration of Ca in all feed ingredients, including all feed additives, in the diets is known when

<b>Table 1.</b> Standardized total tract digestibility (STTD) by growing
pigs of Ca in feed ingredients without and with microbial phytase.

Item, %	STTD	of Ca
Supplementation with phytase <sup>1</sup>	-	+
Mineral supplements		
Monocalcium phosphate <sup>2</sup>	86	86
Dicalcium phosphate <sup>2, 3, 4</sup>	80	80
Ca carbonate <sup>2, 3, 4, 5, 6, 7, 8, 9, 10</sup>	73	78
Plant feed ingredients		
Canola meal <sup>8, 9, 11</sup>	42	65
Soybean meal <sup>8, 9, 12</sup>	78	-
Sunflower meal <sup>9</sup>	26	-
Animal feed ingredients		
Meat and bone meal <sup>13</sup>	77	82
Meat meal <sup>13</sup>	77	86
Fish meal <sup>14</sup>	65	73
Poultry meal <sup>13</sup>	82	82
Poultry by product meal <sup>13</sup>	88	88
Skim milk powder <sup>8</sup>	97	-
Whey powder <sup>8</sup>	99	-
Whey permeate <sup>8</sup>	90	-
<ul> <li><sup>1</sup>Phytase level varied from 500 to 1,500 phyta</li> <li><sup>2</sup>González-Vega et al. (2015b).</li> <li><sup>3</sup>Zhang and Adeola (2017).</li> <li><sup>4</sup>Lee et al. (2019b).</li> <li><sup>5</sup>Blavi et al. (2017).</li> <li><sup>6</sup>Merriman and Stein (2016).</li> <li><sup>7</sup>Merriman et al. (2016a).</li> <li><sup>8</sup>Unpublished data from the University of Illir</li> <li><sup>9</sup>Zhang et al. (2016).</li> <li><sup>10</sup>Kwon and Kim (2017).</li> <li><sup>11</sup>González-Vega et al., 2013.</li> <li><sup>12</sup>Bohlke et al. (2005).</li> <li><sup>13</sup>Merriman et al. (2016b).</li> <li><sup>14</sup>González-Vega et al., 2015a.</li> </ul>	-	diet.



**Figure 1.** Variation in concentration of Ca among 20 sources of soybean meal (Sotak et al., 2016).

diets are formulated and control programs to confirm that final complete diets are produced without excess Ca need to be implemented. In addition, formulation of diets based on STTD Ca will further improve the accuracy of diet formulation because differences in Ca digestibility among ingredients can be taken into account and the impact of microbial phytase on STTD of Ca can be accounted for in the formulation. As a consequence, it may be beneficial to formulate diets based on values for STTD Ca rather than total Ca. The current contribution will attempt to review recent literature on STTD and requirements for STTD by growing-finishing pigs.

### **Digestibility of Ca**

Calcium is digested and absorbed in the stomach and small intestine of pigs and there appears to be no or very limited absorption of calcium in the large intestine (Gonzalez-Vega et al., 2014), but because it is easier to estimate total tract digestibility than ileal digestibility, digestibility of Ca is usually measured over the entire intestinal tract. However, due to the secretion of endogenous Ca into the intestinal tract (Gonzalez-Vega et al., 2013; Nelson et al., 2022), values for apparent total tract digestibility of Ca need to be corrected for endogenous losses of Ca, which results in calculation of values for STTD of Ca. Thus, measurements and calculation of STTD of Ca are identical to the procedures used to determine STTD of P (NRC, 2012). The STTD of Ca in most Ca-containing feed ingredients of animal or plant origin has been reported (Table 1; Gonzalez-Vega et al., 2013; 2015a; 2015b; Merriman et al., 2016b; Zhang et al., 2016). Likewise, values for the STTD of Ca in limestone, monocalcium phosphate, and dicalcium phosphate are available (Gonzalez-Vega et al., 2015a; Merriman and Stein, 2016; Kwon and Kim, 2017; Zhang et al., 2016; Zhang and Adeola, 2017; Lee et al., 2019b). There are, however, only few val-

> ues for the digestibility of Ca in cereal grains and cereal grain co-products (Bohlke et al., 2005), but due to the very low concentrations of Ca in these ingredients, this is of limited practical importance. In most practical diets, at least 50% of the Ca is from limestone/ calcium carbonate and in diets that contain microbial phytase, limestone may account for more than 75% of all the Ca in the diets. Limestone is, therefore, the most important ingredient when it comes to determining Ca digestibility. The STTD of Ca in calcium carbonate has been determined in several experiments and although small differences among sources of calcium carbonate have been report

ed (Lee et al., 2019b; Nelson, 2023), the STTD of Ca in calcium carbonate usually is between 65 and 75% (Gonzalez-Vega et al., 2015a; Merriman and Stein, 2016; McGhee and Stein, 2019; Lee et al., 2019b; Nelson, 2023). Some plant ingredients including soybean meal, bakery meal, and distillers dried grains with solubles have large variations in Ca concentrations (Fig. 1; Pedersen et al., 2007; Sotak-Peper et al., 2016; Liu et al., 2018), but the reason for these variations is that calcium carbonate is sometimes added to these ingredients as a flow agent. The actual concentration of calcium carbonate

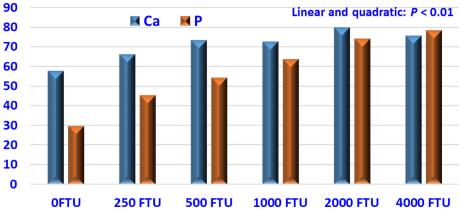


Figure 2. Impact of microbial phytase on digestibility of Ca and P by growing pigs (Lagos et al., 2022).

in diets for pigs, therefore, often is greater than indicated only by the inclusion of calcium carbonate or limestone in the formulation.

### Effect of Microbial Phytase on Ca Digestibility

Early work with microbial phytase demonstrated that phytase does not only increase the digestibility of P, but also the digestibility of Ca. The reason for this effect is that Ca and other divalent cations can bind to the negative charges on the P groups in phytate, but as microbial phytase hydrolyzes the ester bonds that connect P to the inositol ring in phytase, the cations attached to P will be released when P is released. Because microbial phytase is included in the majority of practical diets for pigs, effects of microbial phytase on the digestibility of Ca, therefore, needs to be taken into account in diet formulation. However, although it is acknowledged that microbial phytase increases the digestibility of Ca (Gonzalez-Vega et al., 2015a; 2015b; Lagos et al., 2022), quantitative effects of microbial phytase on the digestibility of Ca are not well understood, and the increase in STTD of Ca in a feed ingredient that is realized by increasing levels of dietary phytase has not been quantified for all phytases. The impact of phytase on Ca digestibility of plant feed ingredients is a combination of release of indigenous Ca from phytate, prevention of binding of Ca from

 Table 2. Theoretical release of Ca from phytate by

 microbial phytase1

 P ions released	Ca ions released	Ca:P ratio of re- leased Ca and P
1	2	2.00:1
2	1	1.50:1
3	1	1.33:1
4	1	1.25:1
5	0	1.00:1
 6	0	0.83:1

<sup>1</sup>Assuming there are a total of 5 Ca and 6 P bound to phytate.

limestone and animal proteins to phytate, and reduced endogenous losses of Ca (Gonzalez-Vega et al., 2013; 2015b; Lee et al., 2019a; Nelson et al., 2022). Because the number of Ca ions relative to P ions that are liberated from phytate changes as the dose of phytase increases, there is no easy way to predict the impact of phytase on Ca digestibility. The first P ion that is released from phytate will result in release of two Ca ions because Ca is a divalent ion, but for each subsequent release of P, only one Ca will be released and if phytate is completely hydrolyzed, no Ca ions will be released when the last one or two P ions are released. As a consequence, the ratio of released Ca to released P changes with the concentration and effectiveness of phytase being used (Table 2), which in turn results in different increases in digestibility of Ca and P as more phytase is added to the diets (Fig. 2; Lagos et al., 2022). In addition, because the number of Ca ions that are bound to P is affected by the presence of other divalent cations in the diet, including Zn and Mg, the release of Ca is not constant among different types of diets. As an example, if dietary Zn is increased, the impact of microbial phytase on Ca digestibility is reduced (Blavi et al., 2017) presumably because Zn occupies some of the binding sites in the phytate molecule, and therefore, prevents Ca from binding. However, currently, there is a lack of information about the release of different cations by phytase and the impact of increasing levels of phytase in the diets on Ca digestibility is not well understood.

### The Role of Vitamin D in Ca Digestibility

Absorption of Ca from the lumen of the small intestine occurs through Ca channels that are located on the luminal side of the enterocytes that line the villi in the small intestine. After absorption into the enterocytes, transport proteins aid in the transfer of Ca to the basolateral side of the enterocytes, where a sodium dependent active transporter will transfer the Ca ion to the interstitium before transport to the liver via the hepatic portal vein. At low concentrations of Ca in the diets, the efficiency of Ca absorption is

**Table 3.** Requirements for standardized total tract digestible (STTD) Ca and STTD P to maximize growth performance (average daily gain) or bone ash by growing-finishing pig<sup>1</sup>.

	······································						
Item	Weight, kg :	11-25	25-50	50-75	75-100	100-135	
STTD P <sup>2</sup> , % :		0.33	0.31	0.27	0.24	0.21	
Growth perfo	Growth performance <sup>3</sup>						
STTD Ca, %		0.47	0.41	0.34	0.29	0.23	
STTD Ca to S	TTD P ratio	1.39	1.31	1.26	1.19	1.10	
Bone ash <sup>4</sup>							
STTD Ca, %		0.55	0.56	0.54	0.52	0.49	
STTD Ca to S	TTD P ratio	1.67	1.81	2.00	2.15	2.33	

<sup>1</sup>Estimates were calculated from published data (González-Vega et al., 2016a; 2016b; Merriman et al., 2017; Lagos et al., 2019a; 2019b).

<sup>2</sup>Requirements for STTD P are from NRC (2012).

<sup>3</sup>There was a negative linear correlation between body weight (X) of growing-finishing pigs and STTD Ca:STTD P ratios (Y) needed to maximize average daily gain: Y = -0.0031X + 1.46. <sup>4</sup>There was a positive linear correlation between body weight (X) of growing-finishing pigs and STTD Ca:STTD P ratios (Y) needed to maximize bone ash: Y = 0.0063X + 1.58.

increased by binding of  $1,25-(OH)_2 D_3$ , which is the active form of vitamin D, to vitamin D receptors located on the enterocytes, resulting in increased expression of Ca channel proteins and Ca transport proteins (Gonzalez-Vega et al., 2016b). However, at greater concentrations of Ca in the diets, the tight junctions between enterocytes become selectively permeable allowing for Ca to be absorbed not only via the Ca channels in the enterocytes, but also via paracellular transport of Ca from the intestinal lumen to the interstitium (Lagos et al., 2019b). Because vitamin D is supplied to swine diets in the form of un-hydroxylated vitamin D<sub>3</sub>, pigs need to hydroxylate vitamin D at the 1 and at the 25 positions, which takes place in the kidneys and the liver, respectively, to generate the active form of the vitamin, which is 1,25- $(OH)_2 D_3$ . It is believed that pigs are efficient in this conversion, but recent data from experiments with gestating sows indicated that the STTD of both Ca and P is increased if metabolites of vitamin D<sub>3</sub>, in the form of either 1-OH-D<sub>3</sub> or 25-OH-D<sub>3</sub>, is supplied in the diets (Lee and Stein, 2022; Lee et al., 2022). In subsequent experiments, it was confirmed that the digestibility of Ca and P also is increased in growing pigs if one of the vitamin D3 metabolites is added to the diets (Univ. Illinois, unpublished). Because vitamin  $D_3$  and vitamin D<sub>3</sub> metabolites increase Ca absorption, whereas microbial phytase increases the number of Ca ions that are available for absorption, effects of vitamin D<sub>3</sub> metabolites and microbial phytase are additive. However, additional research is needed to fully understand the impacts of vitamin  $D_3$  metabolites on Ca absorption and to determine why un-hydroxylated vitamin D3 appears to be less efficient in aiding in absorption of Ca and P than a hydroxylated metabolite of vitamin D<sub>3</sub>.

### **Requirements for Digestible Ca**

The majority of Ca in the body of pigs is stored in bones and to synthesize bone tissue, both Ca and P are needed

along with a protein matrix. Although a number of other minerals are also needed in the synthesis of bone tissue, Ca is quantitatively the most important mineral and constitutes around 38% of bone ash whereas P is present at around 17% (Lagos et al., 2019a; 2019b). Because of these rather constant concentrations of Ca and P in bone tissue, the ratio between Ca and P is fixed and does not change regardless of provisions of Ca and P in the diets. As a consequence, if either Ca or P is deficient in the diet, pigs will synthesize less bone ash, meaning that the bones will become smaller, but still with the same Ca to P ratio (Gonzalez-Vega et al., 2016b; Mer-

riman et al., 2017; Lagos et al., 2019a; 2019b). It is, therefore, important that the ratio between Ca and P in diets for growing pigs meets the need for synthesis of bone tissue. However, whereas Ca is primarily used in bone tissue synthesis, P is also used in the synthesis of soft tissues and other compounds in the body, but as pigs grow older, the proportion of absorbed P used for bone tissue synthesis increases relative to the proportion used for soft tissue synthesis. In finishing pigs, the dietary ratio between Ca and P needed to maximize bone ash closely resembles the ratio between the two minerals in bone ash whereas in smaller pigs, a lower ratio between Ca and P is needed to maximize bone ash because a larger proportion of the absorbed P is used in soft tissue synthesis. To maximize bone ash in growing finishing pigs, the ratio between Ca and P, therefore, is linearly increased from 11 to 130 kg (Table 3). However, dietary concentrations of Ca and P needed to maximize bone ash are greater than the concentrations needed to maximize growth performance (NRC, 2012). Likewise, the ratios between STTD Ca and STTD P needed to maximize growth performance are different from those needed to maximize bone ash. Because of the strong negative effects of excess dietary Ca on growth performance, the ratios between STTD Ca and STTD P that are needed to maximize growth performance are maximum ratios that should not be exceeded. The ratio between STTD Ca and STTD P needed to maximize growth performance of pigs between 11 and 25 kg does not exceed 1.39:1, but this ratio gradually decreases as pigs become heavier, and for pigs between 100 and 130 kg, the ratio should not exceed 1.10:1 (Table 3). These ratios were developed in experiments where pigs were fed multiple levels of both Ca and P and the negative effects of exceeding the suggested ratios were clearly demonstrated (Gonzalez-Vega et al., 2016b; Merriman et al., 2017; Lagos et al., 2019a; 2019b). Later, these ratios were validated in an experiment with pigs fed the suggested ratios from 11 kg to market weight and it was confirmed that pig performance is maximized if these ratios are met, but not exceeded (Lagos et al., 2021). Results of the experiments conducted to determine requirements for STTD Ca also confirmed that the requirements for STTD P suggested by NRC (2012) are adequate to maximize growth performance of terminal pigs if Ca is not provided in excess of the suggested ratios.

Because of the different ratios between STTD Ca and STTD P needed to maximize growth performance and bone ash, it is suggested that the ratios needed to maximize growth performance is used for terminal pigs whereas the ratios needed to maximize bone ash are used in diets for developing gilts that are intended to be kept for reproduction. However, due to the negative effect of excess Ca on the STTD of P, it is also suggested that the provision of P in diets for developing gilts is at least 10% greater than in diets for terminal pigs, and therefore, 10% greater than NRC (2012) requirements. The advantage of formulating diets based on values for STTD Ca rather than values for total Ca is that differences in STTD among feed ingredients can be taken into account and effects of microbial phytase on digestibility of Ca is also accounted for.

### Conclusion

Diets for growing-finishing pigs can be formulated based on values for STTD Ca rather than total Ca because values for STTD of Ca in most Ca containing ingredients have been reported. It is also recognized that both microbial phytase and two metabolites of vitamin D increase STTD of Ca, and effects of phytase and vitamin D metabolites are additive. Excess Ca is negative for growth performance of pigs, and diets should, therefore, be formulated based on a ratio between STTD Ca and STTD P and the ratio between STTD Ca and STTD P is more important than the actual concentration of the two minerals in the diets. Requirements for STTD P suggested by NRC (2012) are accurate if Ca is not fed in excess of requirements. If STTD P is at the requirement, growth performance of growing finishing pigs between 11 kg and market weight is maximized if the ratio between STTD Ca and STTD P is gradually reduced from 1.39:1 to 1.10:1. However, for developing gilts, the provision of STTD P should exceed NRC (2012) requirements by at least 10% and the ratio between STTD Ca and STTD P should increase from 1.67:1 in 11 to 25 kg pigs to 2.33:1 in 100 to 130 kg pigs.

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# Evaluation of Increasing Levels of Mycotoxin-Containing Corn Fines on Diet Choice and Growth Performance of Nursery Pigs

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### Summary

Mycotoxin contamination of corn fed to pigs is known to negatively affect the pig's health and performance. Recent studies testing corn for mycotoxin contamination have reported 75-90% of corn to be contaminated; further, about 90% of this contaminated corn is reported to have multiple mycotoxins. Since mycotoxins result in a variety of adverse responses, the FDA reports guidance and advisory levels for certain mycotoxins, like fumonisins and deoxynivalenol, to hopefully avoid negatively affecting the health and performance of pigs. The physiological response for a single mycotoxin fed alone will be different than a diet with multiple mycotoxin contamination. Multiple mycotoxins can result in synergistic, additive, or antagonistic responses resulting in difficulty predicting a safe level to feed pigs. In an attempt to better understand the effects of feeding corn contaminated with fumonisins, deoxynivalenol, and zearalenone, recent studies were conducted that utilized corn screenings with high levels of contamination to investigate both diet choice/preference and growth performance of pigs in the nursery. It was found that at inclusion levels of 0, 10, and 20% corn fines, pigs showed the ability to discern, and discriminate against, the varying levels of mycotoxin contamination. Additionally, at inclusion level of 60% corn fines there was a detriment to pig growth performance but the addition of Biofix Plus with FUMzyme ameliorated the portion of the lost performance resulting from the fumonisin portion of the mycotoxin contamination.

### Introduction

Mycotoxins are secondary metabolites of filamentous fungi (molds) that, when ingested by animals, can cause a variety of adverse physiological responses. Mycotoxin contaminated feed may result in feed refusal, digestive problems, nervous system problems, reproductive problems, immune suppression, organ damage, and cancer (NRC, 2012). These responses on animal health and reproduction result in an estimated economic loss of \$1.4 billion annually in the United States (CAST, 2003) from three mycotoxins (aflatoxins, deoxynivalenol, and fumonisins). Cargill World Mycotoxin Report (2022) analyzed over 300,000 samples, 75% of the corn-based samples were above the detection limit for at least one of the mycotoxins assayed with 52% of all samples reported to contain 3 or more mycotoxins (Cargill, 2022). Multiple mycotoxin contamination interactions can result in synergistic, additive, or antagonistic effects. The producer of mycotoxins, fungi, have been detected

through the complete production cycle of flowering, harvest and storage of grain (García-Díaz et al., 2020).

It is known that mycotoxins fed to pigs can result in unsatisfactory performance and illnesses. Understanding the relationship between the mycotoxin interactions as well as the time-dose relationship may result in opportunities to improve pig health and production. Knowing the contaminants and contamination levels of the feed results in several intervention opportunities. One of the intervention opportunities is the screening of the corn for fines which often contain the highest concentrations of mycotoxins. Mycotoxin binders can be added to the diet to prevent absorption of the mycotoxin as well as enzymes can be added to diets to detoxify specific mycotoxins. A recent study by Taranu et al. (2011) reported that the addition of boron as calcium fructoborate ameliorated the reduction in performance and improved aspects of the cellular immune response caused by mycotoxins. The objective of the current experiment was

	Stu	dy 1		Study 2	
-			Weeks 1-6	Weeks 1-6	Weeks 6-8
Ingredient, %	Diet 1	Diet 3	Diet 1	Diet 4	Common Diet
Corn	59.55	39.55	60.00	0.00	69.80
Corn fines	0.00	20.00	0.00	60.00	0.00
Soybean meal	35.00	35.00	34.55	34.55	25.00
Choice white grease	2.00	2.00	2.00	2.00	2.00
L-Lysine	0.28	0.28	0.28	0.28	0.25
DL-Methionine	0.19	0.19	0.19	0.19	0.10
L-Threonine	0.11	0.11	0.11	0.11	0.08
Dicalcium Phosphate	1.20	1.20	1.20	1.20	1.10
Limestone	0.90	0.90	0.90	0.90	0.90
Salt	0.50	0.50	0.50	0.50	0.50
Copper sulfate	0.08	0.08	0.08	0.08	0.08
Trace-mineral premix <sup>1</sup>	0.10	0.10	0.10	0.10	0.10
Vitamin premix <sup>2</sup>	0.08	0.08	0.08	0.08	0.08
Santoquin <sup>3</sup>	0.02	0.02	0.02	0.02	0.02
Total:	100.00	100.00	100.00	100.00	100.00
Calculated nutrient composition	4				
Metabolizable energy, kcal/g	3360	3360	3361	3361	3371
Crude protein, %	22.07	22.07	21.89	21.89	18.04
SID Lysine, % <sup>5</sup>	1.25	1.25	1.24	1.24	0.98
Calcium, %	0.73	0.73	0.73	0.73	0.68
Total Phosphorus, %	0.63	0.63	0.62	0.62	0.56
STTD Phosphorus, % <sup>6</sup>	0.39	0.39	0.39	0.39	0.35

<sup>1</sup>Provided the following per kilogram of diet: Zn, 131 mg as ZnO; Fe, 131 mg as FeSO<sub>4</sub>·H<sub>2</sub>O; Mn, 45 mg as MnO; Cu, 13 mg as CuSO<sub>4</sub>·5H<sub>2</sub>O; I, 1.5 mg as Cal<sub>2</sub>O<sub>6</sub>; Co, 0.23 mg as CoCO<sub>3</sub>; Se, 0.28 mg as NaSeO<sub>3</sub>.

<sup>2</sup>Provided the following per kilogram of diet: vitamin A, 6,600 lU; vitamin D3, 880 lU; vitamin E, 44 lU; vitamin K (as menadione sodium bisulfite complex), 6.4 mg; thiamin, 4.0 mg; riboflavin, 8.8 mg; pyridoxine, 4.4 mg; vitamin B12, 33 μg; folic acid, 1.3 mg; niacin, 44 mg; pantothenic acid, 22 mg; D–biotin, 0.22 mg.

<sup>3</sup>Provided 130 mg ethoxyquin per kilogram of diet (Novus International, St. Louis, MO).

<sup>4</sup>Values for metabolizable energy, crude protein, SID lysine, calcium, total phosphorus, and STTD phosphorus were obtained from the feedstuff values listed in the NRC (2012).

<sup>5</sup>SID = standardized ileal digestible basis.

<sup>6</sup>STTD = standardized total tract digestible basis.

to investigate increasing levels of mycotoxin contaminated corn fines on performance and diet preference and the potential of two possible mitigants to ameliorate presumed performance detriments.

### **Experimental Procedures**

### Study 1

The 2020 corn harvest at the University of Kentucky Research Farm was screened to isolate corn fines from the corn using the Kwik-Kleen grain cleaner (Norwood Sales Inc., Horace, ND) with a screen slot of 6/64" by 3/4". The corn fines used in the initial study contained mycotoxin levels that were analyzed to be 23,038 ppb total fumonisin, 1,446 ppb zearalenone (ZEA), and 5,032 ppb total deoxynivalenol (DON) (Table 2). To evaluate the effects of feeding corn fines (screenings) containing mycotoxin levels at the FDA guidance (fumonisins) and advisory (deoxynivalenol) levels (FDA, 2001; FDA, 2010), dietary corn fines were added to a corn-soybean meal-based basal diet at concentrations of 0, 10, and 20 % of the diet (Diets 1-3 respectively) for a target level of about 5,000 ppb fumonisins, 1,000 ppb total deoxynivalenol, and 500 ppb zearalenone for Diet 3. To minimize differences in non-treatment components of the diet, two basal diets were mixed. The first basal diet served as the control diet (0 % fines) whereas the other basal diet served as Diet 3 (20 % fines). Diet 3 was made by replacing 20 % of the corn with corn fines. By blending appropriate amounts of Diets 1 and 3, an intermediate diet (Diet 2, 10 % fines) was created. Three additional treatments (Diets 4, 5, and 6) were used to evaluate a potential dietary mycotoxin mitigant. Diets 4, 5, and 6 were Diet 1-3, respectively, plus 40 mg B/kg diet (as sodium tetraborate decahydrate, 11.34% B). Experimental diets were formulated to meet or exceed the NRC (2012) requirement estimates relative to BW.

#### Table 2. Mycotoxin composition of the diets (Study 1).<sup>1,2</sup>

	Detection Level	Cleaned Corn	Corn Fines	Diet 1 (0%)	Diet 2 (10%)	Diet 3 (20%)
Mycotoxin	ppb	ppb	ppb	ppb	ppb	ppb
Fumonisin B1	200	953	16,597	642	1,787	3,062
Fumonisin B2	200	297	4,999	< 200	592	839
Fumonisin B3	200	< 200	1,442	< 200	203	286
Zearalenone	100	< 100	1,446	< 100	306	304
Deoxynivalenol	200	835	4,743	479	898	1,245
15-ADON	200	< 200	289	< 200	< 200	< 200

<sup>1</sup>Diets 1-3 were Diets 4-6, respectively, plus 40 ppm boron.

<sup>2</sup>Additional mycotoxins assayed which were below the detection level (DL) were: aflatoxin B<sub>1</sub> (DL-20 ppb), aflatoxin B<sub>2</sub> (DL-20 ppb), aflatoxin G<sub>1</sub> (DL-20 ppb), aflatoxin G<sub>2</sub> (DL-20 ppb), HT-2 toxin (DL-200 ppb), T2 toxin (DL-20 ppb), diacetoxyscirpenol (DL-200 ppb), Ochratoxin A (DL-20 ppb), Sterigmatocystin (DL-20 ppb), 3-acetyldeoxynivalenol (DL-200 ppb).

Table 3. Feed preference of pigs fed diets with different levels of mycotoxin containing corn fines (Study 1).1

Comparison 1					
I	Feed intake, kg/d	l		Diet 1 vs 0% vs 10	
Days	0%	10%	SEM	For the period, % consumed	Cumulative consumption
0 to 7	0.407	0.297	0.071	58.5 vs. 41.5	
7 to 14	0.572	0.386	0.103	58.9 vs. 41.1	58.8 vs. 41.2
14 to 21	0.735	0.431	0.113	58.3 vs. 41.7	59.3 vs. 40.7
				Barrows:	58.94 vs. 41.06
				Gilts:	59.74 vs. 40.26
Comparison 2					
I	Feed intake, kg/d		_	Diet 1 vs 0% vs 20	
Days	0%	20%	SEM	For the period, % consumed	Cumulative consumption
0 to 7	0.472	0.219	0.066	69.1 vs. 30.9*	
7 to 14	0.788	0.197	0.156	80.4 vs. 19.6**	75.9 vs. 24.1**
14 to 21	0.979	0.342	0.21	73.5 vs. 26.5**	74.7 vs. 25.3**
				Barrows:	84.66 vs 15.34**
				Gilts:	64.75 vs. 35.25*
Comparison 3					
I	Feed intake, kg/d			Diet 2 vs 10 vs 200	
Days	10%	20%	SEM	For the period, % consumed	Cumulative consumption
0 to 7	0.494	0.161	0.209	76.5 vs. 23.5**	
7 to 14	0.657	0.272	0.229	71.2 vs. 28.8**	73.3 vs 26.7**
14 to 21	0.822	0.347	0.216	70.8 vs. 29.2*	72.2 vs. 27.8**
				Barrows:	78.14 vs. 21.86**
				Gilts:	66.35 vs. 33.65

<sup>1</sup> Each mean represents 4 observations (2 barrow and 2 gilt replicates) per treatment comparison (\*P < 0.05; \*\*P < 0.01).

### Preference Trial

The preference study used a total of 48 crossbred pigs (Yorkshire x Landrace x Large White; 24 barrows and 24 gilts; mean initial BW:  $9.18 \pm 0.12$  kg; 1 week post weaning) that were blocked by sex and body weight (BW), and then randomly allotted within block to one of 3 treatment comparisons with 4 pigs/pen and 4 replicate pens. To avoid the possibility that pigs became accustomed to a feeder loca-

tion, dietary feeder locations were switched every Monday, Wednesday, and Friday throughout the duration of the experiment. Comparison 1 was of 0% (Diet 1) or 10% (Diet 2) fines, Comparison 2 was of 0% (Diet 1) or 20% (Diet 3) corn fines and Comparison 3 was of 10% (Diet 2) or 20% (Diet 3) corn fines.

Table 4. Growth performance of pigs fed increasing levels of mycotoxin containing corn fines (Study 1).1

Boron, ppm:	0				40				P-value		
Fines, %:	0	10	20	0	10	20	SEM	Boron	Fines	Boron*Fines	
Body weight, kg											
d 0	9.98	10.00	9.98	10.03	10.04	10.03	0.037	0.166	0.898	0.955	
d 21	24.98	24.19	24.42	24.71	24.80	24.38	0.367	0.676	0.272	0.246	
ADG, kg	0.71	0.67	0.69	0.70	0.70	0.68	0.017	0.777	0.231	0.195	
ADFI, kg	1.06 <sup>a</sup>	0.97 <sup>b</sup>	1.03 <sup>ab</sup>	1.02 <sup>ab</sup>	1.06 <sup>a</sup>	1.03 <sup>ab</sup>	0.031	0.353	0.631	0.040	
Gain:Feed ratio	0.68	0.69	0.68	0.69	0.67	0.66	0.018	0.392	0.583	0.431	

<sup>1</sup>Each mean represents the mean of 6 replicates (4 pigs/pen) for a 21-day trial.

<sup>a,b</sup>Means with different superscripts within rows differ (P < 0.05).

<sup>L</sup>Linear effect for dietary inclusion of fines (P < 0.05).

Table 5. Clinical chemistry	for growing pigs fee	d varying levels of myce	otoxin containing corn fi	nes (Study 1). <sup>1</sup>

		0	ppm Bord	on	40	) ppm Bor	on			P-vales	
Item	Merck Reference range	Diet 1 (0% Fines)	Diet 2 (10% Fines)	Diet 3 (20% Fines)	Diet 4 (0% Fines)	Diet 5 (10% Fines)	Diet 6 (20% Fines)	SEM <sup>3</sup>	Boron	Fines	Fines* Boron
Sodium, mmol/L	139-153	142.3	142.3	140.6	143.3	142.1	141.1	0.53	0.33	0.00 <sup>L</sup>	0.49
Potassium, mmol/L	4.4-6.5	6.20	6.31	5.78	6.27	6.08	6.04	0.17	0.83	0.13	0.35
Chloride, mmol/L	97-106	102.0	102.3	101.7	102.6	102.1	102.5	0.48	0.32	0.91	0.50
Calcium, mg/dL	9.3-11.5	11.74	11.78	11.68	11.78	11.36	11.28	0.16	0.04	0.21	0.27
Phosphorus, mg/dL	5.5-9.3	9.97	9.82	9.56	9.67	9.77	9.61	0.20	0.56	0.46	0.68
Magnesium, mg/dL	2.3-3.5	2.69	2.77	2.58	2.81	2.63	2.58	0.07	0.96	0.05 <sup>L</sup>	0.20
Total Protein, g/dL	5.8-8.3	5.36	5.33	5.08	5.25	5.31	5.28	0.08	0.75	0.21	0.18
Albumin, g/dL	2.3-4.0	4.06	3.98	3.98	4.03	4.03	3.88	0.09	0.72	0.46	0.70
Globulin, g/dL	3.9-6.0	1.30	1.36	1.10	1.22	1.28	1.41	0.13	0.64	0.85	0.24
A/G Ratio <sup>4</sup>		3.28	3.18	3.71	3.43	3.57	3.33	0.23	0.78	0.72	0.24
Glucose, mg/dL	66-116	126.9	134.7	132.2	121.2	127.8	128	2.76	0.02	0.03 <sup>L</sup>	0.88
Cholesterol mg/dL	81-134	85.0	82.8	74.5	86.5	84.5	80.6	2.32	0.11	0.00 L	0.54
BUN <sup>2</sup> , mg/dL	8.2-25	11.2	11.3	11.5	10.8	13	13	0.77	0.14	0.20	0.36
Creatinine, mg/dL	0.8-2.3	0.97	1.00	0.95	0.99	0.91	0.98	0.03	0.57	0.70	0.09
BUN/Creatinine Ratio, mg/d	IL	11.6	11.5	12.4	11.1	14.3	13.5	0.84	0.10	0.10	0.15
Alkaline Phosphatase, U/L	41-176	287.7	335.5	344.3	319.8	300.2	348.2	28.90	0.99	0.33	0.51
SGOT/AST <sup>3</sup> , U/L	15-55	39.3	45.3	37.3	42.6	37.6	45.7	4.98	0.75	0.99	0.27
Total Bilirubin, mg/dL	0-0.5	0.23	0.23	0.22	0.21	0.22	0.23	0.02	0.83	0.97	0.78

<sup>1</sup> Each main effect represents the mean of 6 replicates (2 pigs/pen) for blood collected on d 21 of the performance trial.

<sup>2</sup> BUN = blood urea nitrogen

<sup>3</sup> SGOT/AST = aspartate transaminase

<sup>4</sup> A/G Ratio = albumin/ globulin ratio

<sup>L</sup> Linear effect for dietary inclusion of fines (P < 0.05).

Table 6. Mycotoxin composition of the diets (Study 2).1

	DL	Clean Corn	Corn Fines	Diet 1 (0%)	Diet 2 (20%)	Diet 3 (40%)	Diet 4 (60%)	Diet 5 (60% + B)	Diet 6 (60% + FUMzyme)
Mycotoxin	ppb	ppb	ppb	ppb	ppb	ppb	ppb	ppb	ppb
Fumonisin B1	100	300	11400	600	3100	5600	8500	6600	5000
Fumonisin B2	100	100	3800	100	900	1700	2300	2100	1600
Fumonisin B3	100	<100	900	<100	200	400	600	500	400
Zearalenone	50	65	1626	63	281	758	774	631	708
Deoxynivalenol	100	770	2997	401	986	2328	2490	2275	2271

<sup>1</sup>Additional mycotoxins assayed which were below the detection level (DL) were: aflatoxin B1 (DL-20 ppb), aflatoxin B2 (DL-20 ppb), aflatoxin G1 (DL-20 ppb), aflatoxin G2 (DL-20 ppb), HT-2 toxin (DL-20 ppb), T2 toxin (DL-20 ppb), diacetoxyscirpenol (DL-200 ppb), Ochratoxin A (DL-20 ppb), Sterigmatocystin (DL-20 ppb), 15-acetyldeoxynivalenol (DL-200 ppb), 3-acetyldeoxynivalenol

### Performance Trial

The performance study used a total of 144 crossbred pigs (Yorkshire x Landrace x Large White; 72 barrows and 72 gilts; mean initial BW:  $10.20 \pm 0.23$  kg; 1 week post weaning) that were blocked by sex and body weight (BW), and then randomly allotted within block to one of six dietary treatments with 4 pigs/pen and 6 replicate pens. The six dietary treatments were a result of a 3 (corn fines inclusion level) x 2 (boron addition) factorial arrangement. In total, the experiment lasted for 3 weeks. To determine serum clinical chemistry profiles, a total of 2 pigs/pen were selected for blood sampling on day 21.

### Study 2

Corn harvested at the University of Kentucky Research Farm was screened by a grain cleaner to screen off corn fines that would contain the highest concentrations of mycotoxins. The corn fines used in this study contained mycotoxin levels that were analyzed to be 16,100 ppb total fumonisin, 1,626 ppb zearalenone, and 2,997 ppb total deoxynivalenol (Table 6). To evaluate the effects of feeding these corn fines (screenings) containing mycotoxin levels greater than the FDA cautionary levels, dietary corn fines were added to a corn-soybean meal-based basal diet at concentrations of 0, 20, 40, and 60 % of the diet (Diets 1-4, respectively, Table 6). To minimize differences in non-treatment components of the diet, Diets 1 and 4 were mixed with the first diet serving as the control diet (0 % fines) and the other diet serving as Diet 4 (60 % fines). Diets 5 and 6 were used to evaluate two potential dietary mycotoxin mitigants. Diets 5 and 6 consisted of 60 % corn fines plus 40 mg B/kg diet (as sodium tetraborate decahydrate, 11.34% B) and 0.25 % Biofix Plus with FUMzyme (DSM Nutritional Products, Parsippany, NJ). Experimental diets were formulated to meet or exceed the NRC (2012) requirement estimates regarding BW.

A total of 150 crossbred pigs (Yorkshire x Landrace x Large White; 90 barrows and 60 gilts; mean initial BW: 6.42  $\pm$  0.06 kg) were blocked by sex and body weight (BW), and then randomly allotted within block to one of six dietary treatments with 5 pigs/pen and 5 replicate pens. In total, the experiment lasted for 8 weeks; during Weeks 1 to 6 pigs were offered their respective experimental diet whereas during Weeks 6 to 8 all pigs were offered a common grower diet to assess potential compensatory growth effects. On week 6, 3 pigs/pen were selected for serum collection for a clinical chemistry panel.

Table 7. Growth performance of pigs	ed increasing levels of mycotoxin containing corn	fines and two potential mitigants (Study 2). <sup>1</sup>
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Diet:	1	2	3	4	5	6			P-values <sup>2</sup>	
Item	No Fines	20% Fines	40% Fines	60% Fines	60% Fines + B	60% Fines + FUMzyme	SEM	Fines	Diet 4 vs. Diet 5	Diet 4 vs. Diet 6
Body weig	ght, kg									
Wk 0	6.41	6.40	6.39	6.40	6.40	6.39	0.008	0.463	0.711	0.185
Wk 6	33.01 <sup>a</sup>	32.67 <sup>a</sup>	33.32 <sup>a</sup>	30.11 <sup>b</sup>	30.30 <sup>b</sup>	31.90 <sup>ab</sup>	0.739	0.027	0.860	0.107
Wk 8	45.07	46.04	45.87	43.15	44.48	45.53	1.168	0.536	0.434	0.171
Average c	laily gain, kg									
Wk 0-6	0.62 <sup>a</sup>	0.61 <sup>a</sup>	0.63 <sup>a</sup>	0.55 <sup>b</sup>	0.56 <sup>b</sup>	0.59 <sup>ab</sup>	0.017	0.026 <sup>L</sup>	0.838	0.099
Wk 6-8	1.00 <sup>c</sup>	1.07 <sup>abc</sup>	1.05 <sup>bc</sup>	1.09 <sup>abc</sup>	1.18 <sup>a</sup>	1.14 <sup>ab</sup>	0.042	0.098	0.130	0.425
Wk 0-8	0.70	0.72	0.72	0.67	0.69	0.71	0.021	0.529	0.423	0.164
Average o	laily feed inta	ake, kg								
Wk 0-6	1.03	0.99	1.02	1.12	1.14	1.11	0.077	0.675	0.902	0.899
Wk 6-8	1.91	2.03	2.06	2.11	2.13	2.07	0.057	0.164 <sup>L</sup>	0.812	0.678
Wk 0-8	1.22	1.19	1.24	1.31	1.35	1.32	0.067	0.515	0.623	0.885
Gain: feed	l ratio									
Wk 0-6	-	-	-	-	-	-				
Wk 6-8	0.52	0.53	0.51	0.52	0.56	0.55	0.014	0.178	0.071	0.127
Wk 0-8	-	-	-	-	-	-				

<sup>1</sup>Five pens of five pigs/pen per treatment mean. Week 1-6 pigs were fed their respective treatment diets; week 6-8 all pigs received a common corn-soybean meal diet.

a,b,cMeans with different superscripts within rows differ (P<0.05).

Linear effect for dietary inclusion of fines (Diets 1-4).

**Table 8.** Sphinganine and sphingosine levels in the serum, at week 6, of pigs fed varying levels of mycotoxin containing corn fines and a potential mitigant (Study 2),<sup>1,2,3</sup>

			Diet						
Item	1	2	3	4	6	SEM	L	Q	4 vs. 6
SA, pmol	7,062 <sup>b</sup>	59,204 <sup>b</sup>	81,757 <sup>ab</sup>	151,773 <sup>a</sup>	15,926 <sup>b</sup>	31,876	0.004	0.565	0.005
SO, pmol	217,185	254,498	300,960	331,312	291,516	87,330	0.259	0.945	0.702
SA:SO	0.042d	0.220bc	0.288 <sup>ab</sup>	0.425a	0.061 <sup>cd</sup>	0.067	0.001	0.524	0.001

<sup>1</sup>Serum was analyzed at week 6 for selected pigs.

<sup>2</sup>Means represent 4 pigs for Diet 1, 4 pigs for Diet 2, 3 pigs for Diet 3, 4 pigs for Diet 4, and 4 pigs for Diet 6. <sup>3</sup>SA=sphinganine, SO=sphingosine.

### **Results and Discussion**

Study 1

## Preference Trial

### Comparison 1

Table 3 shows the results of the three comparisons made during this experiment. During Comparison 1 with either 0% (Diet 1) or 10% (Diet 2) fines, the pigs showed an inclination choosing the diet containing 0% fines about 18% more than the diet with 10% fines within the first week and stayed consistent throughout the duration of the experiment. While there was a numerical difference, there was not a statistical difference.

### Comparison 2

The second comparison consisted of diets with either 0% (Diet 1) or 20% (Diet 3) corn fines. Pigs were able to significantly discern between diets within the first week choosing Diet 1 38% more compared to Diet 3. Weeks 2 and 3 resulted in the same preference of pigs choosing Diet 1 over Diet 2 at 80.4% and 19.6%, respectively (P < 0.01). Cumulative consumption resulted in a 74.7% preference for Diet 1. Barrows consumed the diet with no fines 84.66% of the time while gilts were below that at 64.75% (P < 0.01 and 0.05, respectively). The difference in the ability to discern between diets may be a result of the DON in the diet which males are more sensitive to (Kamle et al., 2022).

### Comparison 3

The third comparison was of 10% (Diet 2) and 20% (Diet 3) corn fines. Like Comparison 1 and 2, pigs preferred the diet with the lowest amount of mycotoxin-containing corn fines. The percent consumed difference for Weeks 1, 2, and 3 between Diets 2 and 3 were 53%, 42.4%, and 41.6%, respectively, all preferring Diet 2 (P < 0.01, 0.01, and 0.05, respectively). Overall, consumption of Diet 2 was 72.2% and Diet 3 was 27.8% (P < 0.01). This comparison also showed a greater preference by the barrows for Diet 2 compared to the gilts at 78.14% (P < 0.01) and 66.35%, respectively, for Diet 2.

### Performance Trial

Overall, ADG and G:F ratio showed no effect for fines or boron and their interaction (Table 4). Overall, performance measurements were not affected by the addition of fines. A study with similar mycotoxins conducted by Wilson et al. (2022) found that increasing the concentration of deoxynivalenol and zearalenone and decreasing fumonisins from 600, 400, and 250 ppb to 3570, 624, and 120 ppb, respectively, significantly decreased ADG, ADFI, and G:F ratio (P < 0.05). Frobose et al. (2015) observed the same negative effect on ADG, ADFI, and G:F when DON, fumonisin, and ZEA were naturally contaminated in the diet (4600, 2000, and 500 ppb, respectively). The level of fines in this study did not elicit a response in performance measures as was observed by Wilson et al. (2022) and Frobose et al. (2015). While these two studies contain the same mycotoxins as the current study, the concentrations differ which will affect how the pig physiologically reacts.

Table 5 reports the effects that increasing levels of mycotoxin-containing corn fines with two levels of boron has on clinical chemistry. The addition of boron showed no significant effect among the mineral analytes except calcium (P = 0.04). The increasing level of fines resulted in a linear decrease in sodium concentration (P < 0.05). Deoxynivalenol has been reported to inhibit the sodium glucose-linked transporter 1 (SGLT1) in chickens (Awad et al., 2007). This inhibitory action agrees with Maresca et al. (2002) who reported that in human intestinal cells exposed to low concentrations of DON, SGLT1 appeared to be the most sensitive followed by the d-fructose transporter The data presented in this study for glucose in not in agreement with the affect DON has on the SGLT1 transporter. The data reported from this study indicates a significant linear increase in glucose as fines increase (P = 0.03). If the SGLT1 transporter is inhibited by DON as indicated by Maresca et al. (2002) and Awad et al. (2007) this would seemingly cause glucose concentrations to decrease. The addition of boron significantly increased glucose serum concentration (P = 0.02). There is evidence to suggest that boron may affect the levels of glucose by reducing the amount of insulin needed to maintain glucose concentrations (Bakken and Hunt, 2003). Cholesterol was also detected to have a significant

Table 9. Clinical chemistry for growing pigs fed varying levels of mycotoxin containing corn fines (Study 2).1

	Diet:	1	2	3	4	5	6		P-Va	lues
Item	Merck Reference range	0% Fines	20% Fines	40% Fines	60% Fines	60% Fines + B	60% Fines + FUMzyme	SEM <sup>3</sup>	Diet 4 vs. Diet 5	Diet 4 vs. Diet 6
Sodium, mmol/L	139-153	144.1	143.2	143.4	141.7	143.3	142.7	0.69 <sup>L</sup>	0.10	0.29
Potassium, mmol/L	4.4-6.5	6.31	6.30	5.87	6.01	5.95	5.93	0.19	0.82	0.74
Chloride, mmol/L	97-106	103.6	102.8	102.2	101.8	102.4	102.7	0.62 <sup>L</sup>	0.44	0.28
Calcium, mg/dL	9.3-11.5	11.66	11.58	11.55	11.43	11.34	11.38	0.13	0.53	0.79
Phosphorus, mg/dL	5.5-9.3	10.22	10.35	10.50	10.48	10.77	11.07	0.24	0.37	0.06
Magnesium, mg/dL	2.3-3.5	2.76	2.71	2.79	2.70	2.76	2.66	0.08	0.60	0.72
Total Protein, g/dL	5.8-8.3	5.45	5.36	5.42	5.26	5.26	5.25	0.08	0.99	0.90
Albumin, g/dL	2.3-4.0	4.22	4.14	4.16	3.95	4.22	4.09	0.08 <sup>L</sup>	0.02	0.20
Globulin, g/dL	3.9-6.0	1.23	1.22	1.25	1.31	1.05	1.16	0.07	0.01	0.13
A/G Ratio <sup>4</sup>		3.57	3.49	3.53	3.24	4.17	3.84	0.25	0.01	0.08
Glucose, mg/dL	66-116	112.8	115.3	118.0	113.1	111.2	114.8	2.19	0.51	0.57
Cholesterol <sup>5</sup> , mg/dL	81-134	84.86	74.96	80.26	76.67	74.14	85.42	3.07	0.54	0.03
BUN <sup>2,6</sup> , mg/dL	8.2-25	15.0	13.2	13.0	13.6	14.3	16.0	0.79	0.54	0.03
Creatinine, mg/dL	0.8-2.3	1.17	1.11	1.02	1.02	1.06	1.07	0.04 <sup>L</sup>	0.53	0.34
BUN/Creatinine Ratio, mg/c	۱L	12.9	12.0	12.8	13.9	13.9	14.9	0.81	0.99	0.36
Total Bilirubin, mg/dL	0-0.5	0.27	0.27	0.29	0.28	0.26	0.30	0.03	0.55	0.62
Alkaline Phosphatase, U/L	41-176	281.2	275.5	279.5	250.1	289.5	258.7	26.96	0.28	0.81
SGOT/AST <sup>3</sup> , U/L	15-55	25.3 <sup>b</sup>	31.5 <sup>ab</sup>	23.7 <sup>b</sup>	37.5 <sup>a</sup>	31.4 <sup>ab</sup>	30.6 <sup>ab</sup>	4.70	0.34	0.26

<sup>1</sup> Each mean analyte value represents the mean of 5 replicates (3 pigs/pen) for blood collected at week 6.

<sup>2</sup> BUN = blood urea nitrogen

<sup>3</sup> SGOT/AST = aspartate transaminase

<sup>4</sup> A/G Ratio = albumin/ globulin ratio

<sup>5</sup>Increasing levels of fines reduced serum cholesterol concentrations (P = 0.02)

<sup>6</sup>Increasing levels of fines reduced serum blood urea nitrogen concentrations (P = 0.048)

<sup>a,b</sup>Means with different superscripts within rows and columns under their respective analyte differ (P < 0.05).

<sup>L</sup> Linear effect for dietary inclusion of fines (P < 0.05).

linear decrease as fines increased in the diet for overall cholesterol. The increase in dietary fines results in an increase in total dietary fiber, a meta-analysis by Brown et al. (1999) analyzed 67 controlled clinical trials that indicated that high soluble fiber decreases total and low-density lipoprotein cholesterol. While these selected differences were observed in the clinical chemistry responses, it should be noted that all responses were within the Merck (2006) reference range with the exceptions that all treatments had low globulin results and high alkaline phosphatase results.

This data shows pigs can discern between diets with different concentrations of mycotoxin-containing corn fines. Taste allows pigs the ability of sensing nutrients and harmful substances, guiding the selection of nutritious feeds and avoiding toxins (Klasing and Humphrey, 2009). If given a choice, pigs will choose the diet with lower amounts of mycotoxins.

#### Study 2

Increasing the mycotoxin-containing corn fines resulted in a linear decrease (P = 0.003) in ADG for Diets 1-4 for Weeks 0-3 with values for Diets 1, 2, 3, and 4 of 0.44, 0.44,

0.43, and 0.39 kg/day, respectively. However, only the highest level of fines was detrimental. Weeks 3-6 also showed a tendency (P < 0.10) of the fines to reduce ADG resulting in a linear reduction in performance (P < 0.026) for Weeks 1-6 (Table 7; duration of experimental diet exposure). Throughout Week 1-6, pigs on the 0% fines diet had an ADG of 0.62 kg/day while the pigs on the 60% diet had a reduced ADG of 0.55 kg/day. This loss in ADG resulted in a linear decrease in BW due to the inclusion of fines. Between Diets 1 and 4, there was a discrepancy in weight by Week 3 of 1.18 kg and at Week 6 of 2.9 kg (8.8% reduction in BW). There is no way to know what percentage of the detriment was caused by fumonisins, deoxynivalenol, or zearalenone.

Average daily feed intake measurements were also taken but show an increase in feed intake as dietary fines increase. This increase in feed usage is not related to feed consumption, rather the pig care workers noted feed was being wasted into the pit. Because the values for feed disappearance do not represent actual ADFI, those values cannot be used to calculate an accurate G:F ratio. Boron was a possible mitigant added to 60% corn fines at 40 ppm (Diet 5), as sodium tetraborate decahydrate. The comparison of Diet 4 to Diet 5 indicates sodium tetraborate decahydrate does not alleviate the negative effects that corn fines had on BW and ADG in this study (Table 7). The other mitigant added to create Diet 6 was 60% fines and 0.25 % Biofix Plus with FUMzyme (DSM Nutritional Products, Parsippany, NJ).

The addition of FUMzyme resulted in a significant difference from Diet 4 at week 3 BW and Weeks 0-3 ADG (data not shown for Week 3; P = 0.023 and 0.021, respectively) with a tendency to be different for Weeks 1-6 (P < 0.10). While the BW and ADG did differe between Diets 1 and 4, those values no longer differed between Diets 1 and 6; for Weeks 1-6, Diet 6 was able to recover 57% of the lost ADG caused by the addition of mycotoxin-containing corn fines. To quantitively measure the effect that FUMzyme has on fumonisin toxicosis, sphinganine (SA) and sphingosine (SO) biomarkers were measured (Table 8). The overall SA values show a linear increase (P = 0.004) in concentration as fines increased from Diets 1-4. The SA concentration for pigs fed Diet 1 was measured at 7,062 pmol and pigs fed Diet 4 measured at 151,773 pmol, the addition of FUMzyme ameliorated 93% of the increased SA caused by fumonisins. The SO numbers increased at a lesser rate than the SA numbers and showed no significance. The high rate of increase for SA and the low rate of SO caused the SA:SO ratio to increase. This caused a linear increase (Diets 1-4; P = 0.001) in ratio to increased corn fines. Diet 1 analyzed for a ratio of 0.022 while Diet 4 analyzed for 0.425 and Diet 6 relieved 95% of the increased ratio. This SA: SO ratio increase was similarly seen (Rao et al., 2020) when pigs weighing 8.9 kg were fed a dietary treatment of 7,200 ppb fumonisin for 28 days (SA: SO = 0.55); furthermore, as the fumonisin concentration increased to 35,100 for 14 days, the ratio was observed to increase up to 1.58. This supports the idea that SA:SO as a biomarker is sensitive and reliable.

The serum clinical chemistry panel (Table 9) showed sodium, chloride, and albumin were linearly decreased for Diets 1-4. Cholesterol and blood urea nitrogen were also significantly decreased as the inclusion rate of fines increased (P = 0.02 and 0.049, respectively). The addition of FUMzyme was significant (P = 0.03) with recovering over 100% of the cholesterol change due to fines.

### Conclusion

In conclusion, when pigs are given a choice, they have the ability to discern between diets containing these mixed mycotoxins at different inclusion levels but when not given a choice of diets there was not necessarily a difference in performance. As mycotoxin-containing fines increased in the diet (60% of the diet), pigs eventually exhibited negative effects on ADG and BW but the enzyme Biofix Plus with FUMzyme ameliorated the portion of the lost performance resulting from the fumonisin portion of the mycotoxin contamination. Further, the improved performance from Weeks 6-8 for all pigs, on all diets that had fines, does show the importance of feeding clean corn to optimize ADG, ADFI and gain/feed ratio.

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## Understanding the Growth and Health Benefits of Supplemental Treated Wheat Straw in the Weaned Pig Diet

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### Summary

Three experiments were conducted at Michigan State University to understand the growth and health benefits of including wheat straw treated with acid, heat, and calcium carbonate, in weanling pig diets. The hypothesis of the first experiment was that weaned pigs fed increasing amounts of treated wheat straw would experience improved gut health, but too much dietary fiber may decrease performance. The hypothesis of the second experiment was that treatment of wheat straw provides a benefit to growth and health beyond that of untreated wheat straw. And the hypothesis of the third experiment was that the immune modulating properties of treated wheat straw would be observed in nursery pigs experiencing a vaccination-induced immune challenge. The results of these experiments confirmed that pig gains were maintained, feed efficiency improved, and immune function altered with treated wheat straw in nursery diets. Treatment of the wheat straw led to different effects as compared to untreated wheat straw. The effects of dietary treated wheat straw on immunity in vaccination-challenged nursery pigs in the third experiment were observed in the ascending colon. The immune challenge in the third experiment led to different circulatory and local immune responses to dietary treated wheat straw than were observed with unchallenged nursery pigs fed similarly in the first two experiments.

### Introduction

Dietary fiber is increasingly being thought important for improving the gastrointestinal development and function, health, and the performance of the growing pig (Jin et al., 1994; Pie et al., 2007; Weber et al., 2008). Parallel with this thought, are global discussions of human food security and the desire for animal proteins in people's diets, leading to increased interest in utilization of crop residues as animal feeds in locations where feedstuffs for livestock are undersupplied. The use of "processes that had (have) as their object the rendering of the cellulose in cereal straw more available to the animal" has long been considered a means to increase the supply of feedstuffs for animals, including pigs (Kellner, 1913; Woodman and Evans, 1947). Processes or treatments may be chemical, thermal, enzymatic, microbiological and combinations thereof.

In the 1970's it was Dr. H. Bergner and his students that studied the feeding of treated and untreated wheat straw for growing-finishing pigs from 30 to 100 kg live weight. They treated wheat straw with hydrochloric acid, steam, and calcium hydroxide, and called it "partly hydrolyzed straw meal." Bergner (1981) summarized several of their studies by stating that the feeding value of straw was doubled after the treatment, containing approximately 20% utilizable carbohydrates (Bergner and Betzin, 1979). More recently, it has been reported that broilers may be fed up to 25% treated rice straw throughout their lifetime without impacting bird growth (El-Husseiny et al., 2006; Michael, 2010; Michael, 2016).

### **Experiment Protocols**

The chemical analyses of wheat straw as baled or harvested (untreated, UWS) and treated wheat straw (TWS) are shown in Table 1. Treatment results in slightly less crude protein and minor changes in amino acid content. Treatment also results in greater crude fat. Because phosphoric acid and calcium carbonate are added during treatment, TWS contained 100 times more P and 16 times more Ca than UWS. Fiber components are also altered by the treatment process, resulting in less assayed crude fiber, NDF, and ADF. Percentages of hemicellulose, cellulose, and lignin decrease with treatment.

*Experiment One.* At  $27.1 \pm 1.3$  d of age, 108 pigs (PIC 327 x Yorkshire) were weaned and randomly allotted to three treatments: 1) Control, 2) 5% TWS, and 3) 10% TWS. There were six pens (six pigs per pen) per treatment. On d 28 one pig from each pen was randomly selected (equal number of males and females) to be sacrificed for sample collection.

*Experiment Two.* One hundred and ninety-two pigs (PIC 359 x Yorkshire) were weaned at  $26.8 \pm 1.1$  d old (BW =  $8.3 \pm 1.1$  kg) and randomly allotted to 24 pens with eight pigs per pen. Three dietary treatments were fed over a 28-d period: 1) Control, 2) 5% UWS, and 3) 5% TWS. One pig per pen (equal number of males and females) was randomly selected for sacrifice and sample collection on d 28.

*Experiment Three.* Twenty barrows (PIC 800 x Yorkshire) were weaned at  $27.7 \pm 0.8$  d (BW =  $8.6 \pm 1.2$  kg) and randomly assigned to four cohorts based on weight and parentage, with six pigs per cohort. Two cohorts, one heavy and one lightweight, were randomly assigned to two treatments:

1) Control and 2) 5% TWS. After being fed experimental diets for 21 d, pigs were challenged with two vaccinations, the injectable Ingelvac<sup>\*</sup> CircoFLEX<sup>\*\*</sup> Porcine Circovirus Type 2 vaccine and an orally administered Enterisol<sup>\*</sup> *Lawsonia Intracellularis* vaccine. Five pigs per cohort, 10 per treatment, were randomly selected and sacrificed on d 42.

In all experiments, dietary treatments were imposed over three nursery phases. All diets met or exceeded the nutrient requirements of NRC (2012) and were isocaloric within each phase. No antibiotic was fed, and neither were pharmacological amounts of copper or zinc. Growth performance data was recorded on a weekly basis. Blood was collected prior to euthanasia to assess plasma immunological markers immunoglobulin A (IgA), major histocompatibility complex (MHC) class II expression in peripheral blood circulating monocytes (PBMC) treated with lipopolysaccharide (LPS) and(or) intestinal fatty acid binding protein (iFABP). Upon sacrifice, segments of the distal ileum and ascending colon were removed to collect mucosal scrapings for the analysis of localized immunological markers interleukin 6 (IL-6), interleukin 10 (IL-10), interleukin 12 (IL-12), and (or) tumor necrosis factor-alpha (TNF- $\alpha$ ).

**Table 1.** Analyzed composition of three different wheat straws, both untreated and treated, harvested different years in separate locations in Michigan<sup>a</sup>.

	201	8b	202	20c	2023d		
Item	Untreated	Treated	Untreated	Treated	Untreated	Treated	
Moisture, %	5.37	5.80	4.47	5.68	8.32	6.97	
GE, kcal/100 g	200	150	189	163	175	166	
Crude protein, %	3.43	2.55	2.12	1.66	2.67	1.40	
Crude fat, %	1.14	2.83	1.03	4.05	0.74	4.53	
Ash, %	4.49	26.7	6.76	28.79	6.69	30.30	
Calcium, %	0.27	4.28	0.25	4.55	-	-	
Phosphorus, %	0.08	8.29	0.08	9.09	-	-	
Lys, %	0.09	0.06	0.10	0.07	-	-	
Met, %	0.05	0.03	0.04	0.03	-	-	
Thr, %	0.11	0.08	0.08	0.05	-	-	
Crude fiber, %	41.49	33.60	42.92	29.84	42.27	26.81	
NDF, %	73.99	37.39	78.66	42.41	75.31	53.65	
ADF, %	48.82	33.21	54.63	39.75	53.65	37.26	
Hemicellulose, %	25.17	4.18	24.03	2.66	21.66	2.62	
Cellulose, %	39.75	25.74	43.35	30.78	42.08	28.79	
Lignin, %	7.81	6.88	7.95	7.66	8.68	7.06	
Total dietary fiber, %	77.07	41.02	90.12	49.85	80.07	42.63	
Total soluble fiber, %	< 0.5	1.13	0.71	0.71	< 0.08	< 0.08	
Total insoluble fiber, %	76.57	39.99	88.59	48.35	78.96	40.91	

<sup>a</sup> Chemical analyses, enzymatic gravimetric analyses and subsequent use of predictive equations performed by the University of Missouri Agricultural Experimental Station Chemical Laboratory (Columbia, MO, USA).

<sup>b</sup> Wheat straw harvested as small square bales from private farm in Ingham County, Michigan.

<sup>c</sup> Wheat straw harvested as large square bales, stored, later cleaned using a vacuum system which removed smaller particulate (chaff, dust, leaves), leaving a greater proportion of stalks and stems, chopped, and bagged in plastic, from private farm and retailer of cleaned forages in St. Clair County, Michigan.

<sup>d</sup> Wheat straw harvested as small square bales from MSU South Campus Farms in Ingham County, Michigan.

### Effects of Dietary Treated Wheat Straw on Weanling Pigs

Growth Performance. In experiment one, our group reported that body weights and average daily gains are unchanged with the feeding of 5% and 10% treated wheat straw in the diet (Table 2; Lewton et al., 2019). Whitney and Shurson (2004) observed a similar outcome with the feeding of 10% dietary corn distiller's dried grains with solubles. The addition of 10% TWS decreased (P < 0.05), average daily feed intake during weeks 2, 3, 4, and overall, relative to those measures with Control pigs. Coincidentally then, feed efficiency was also greater for pigs fed 10% TWS in weeks 2, 4 and overall, relative to the efficiencies experienced by Control pigs (P < 0.05). A similar improvement was observed for pigs fed 5% TWS in week 2 and over the entire 4-week period. In our second study (Table 2; Polniak et al., 2023) we observed that treating wheat straw, as compared to untreated wheat straw, resulted in greater gain in week 3, and greater feed efficiencies in weeks 1, 3, and overall (P < 0.05). Pigs fed the 5% TWS treatment also tended to be more efficient than Control pigs (P = 0.07). Both studies showed that the treatment of wheat straw resulted in improved feed efficiency. We speculate that with an increased available lipid content from treating the wheat straw, a greater net energy may lead to more efficient pig growth. Our third experiment was intentionally designed not to measure growth performance.

Intestinal Markers. In Table 3, the expression of intestinal cytokines relative to the Control treatment within a study is symbolically presented. In the first experiment expression was measured in mucosa of the ascending colon, not the ileum. The feeding of 10% TWS down-regulated the expression of the pro-inflammatory cytokines TNF- $\alpha$ and IL-12 (P < 0.05). Expression of cytokines in colonic mucosa of pigs fed the Control and 5% TWS treatments were similar.

In our second experiment, proinflammatory cytokines IL-6 and IL-12 were decreased in the ileum with the feeding of TWS and UWS (P < 0.05). Ileal TNF- $\alpha$  tended to be decreased by feeding UWS (P = 0.08). Also, with the feeding of UWS, the anti-inflammatory cytokine IL-10 also tended to be expressed less (P = 0.09). In the second experiment, the expression of mucosal cytokines in the ascending colon with the feeding of 5% TWS were dissimilar to those observed in the first experiment. A down-regulation of IL-10 was the only difference when comparing the Control and 5% TWS treatments. Colonic mucosal TNF- $\alpha$  did not difference.

Table 2. Growth performance of nursery pigs fed 5 and 10% treated wheat straw (TWS) in study one and fed untreated wheat straw (UWS)
and 5% TWS in study two.

Item		Stud	y One		Study Two				
	Control	5% TWS	10% TWS	SEM	Control	5% UWS	5% TWS	SEM	
BW, kg									
Initial	8.47	8.57	8.49	0.36	8.30	8.30	8.31	0.21	
Final	17.73	18.08	17.76	0.36	19.83	19.26	19.81	0.21	
ADG, kg									
Week 1	0.20	0.22	0.18	0.02	0.17	0.17	0.18	0.01	
Week 2	0.27	0.28	0.27	0.02	0.34	0.32	0.32	0.01	
Week 3	0.36	0.34	0.33	0.02	0.50 <sup>a</sup>	0.45 <sup>b</sup>	0.51ª	0.02	
Week 4	0.50	0.51	0.54	0.02	0.64	0.63	0.63	0.02	
Overall	0.33	0.34	0.33	0.02	0.41	0.39	0.41	0.01	
ADFI, kg									
Week 1	0.27	0.26	0.23	0.03	0.27	0.26	0.26	0.01	
Week 2	0.51ª	0.46 <sup>ab</sup>	0.43 <sup>b</sup>	0.03	0.45	0.44	0.44	0.02	
Week 3	0.69 <sup>a</sup>	0.62 <sup>ab</sup>	0.60 <sup>b</sup>	0.03	0.74	0.69	0.70	0.02	
Week 4	0.91ª	0.88 <sup>ab</sup>	0.82 <sup>b</sup>	0.03	0.98	0.96	0.96	0.02	
Overall	0.59 <sup>a</sup>	0.56 <sup>ab</sup>	0.52 <sup>b</sup>	0.02	0.61	0.59	0.59	0.02	
G:F									
Week 1	0.76	0.81	0.78	0.03	0.62 <sup>a</sup>	0.63 <sup>a</sup>	0.71 <sup>b</sup>	0.02	
Week 2	0.52 <sup>a</sup>	0.61 <sup>b</sup>	0.64 <sup>b</sup>	0.03	0.76	0.72	0.73	0.01	
Week 3	0.52	0.55	0.55	0.03	0.69 <sup>a</sup>	0.66 <sup>a</sup>	0.72 <sup>b</sup>	0.01	
Week 4	0.55ª	0.59 <sup>a</sup>	0.66 <sup>b</sup>	0.03	0.65	0.66	0.65	0.01	
Overall	0.59 <sup>a</sup>	0.64 <sup>b</sup>	0.66 <sup>b</sup>	0.01	0.68 <sup>abx</sup>	0.67 <sup>b</sup>	0.70 <sup>ay</sup>	0.01	

<sup>ab</sup> Means within a row with different superscripts differ (P < 0.05).

xy Values in a common row lacking a common superscript tend to differ ( $P \ge 0.05$  to  $P \le 0.10$ ).

<b>Table 3.</b> Effect of 5 and 10% treated wheat straw (TWS) and (or) 5% untreated wheat straw (UWS) on the expression ( $\Delta\Delta$ CT relative to the
control treatment within study) of intestinal mucosa immunological markers of pigs determined by quantitative PCR.

		5	15		
	Stud	y One	Study	Study Three	
	5% TWS	10% TWS	5% UWS	5% TWS	5% TWS
Distal lleum					
TNF-α	NM	NM	$\downarrow$	$\leftrightarrow$	$\leftrightarrow$
IL-6	NM	NM	Ļ	Ļ	$\leftrightarrow$
IL-10	NM	NM	$\downarrow$	$\leftrightarrow$	$\leftrightarrow$
IL-12	NM	NM	Ļ	Ļ	$\leftrightarrow$
Ascending Colon					
TNF-α	$\leftrightarrow$	ţ	$\leftrightarrow$	$\leftrightarrow$	t
IL-6	$\leftrightarrow$	$\leftrightarrow$	Ļ	$\leftrightarrow$	t
IL-10	$\leftrightarrow$	$\leftrightarrow$	$\leftrightarrow$	Ļ	$\leftrightarrow$
IL-12	$\leftrightarrow$	Ļ	Ť	$\leftrightarrow$	$\leftrightarrow$

NM stands for "not measured."

**\downarrow** Change relative to control differed (*P* < 0.05).

↓ Change relative to control tended to be different ( $P \ge 0.05$  to  $P \le 0.10$ ).

 $\leftrightarrow$  Change relative to control was not different (P > 0.10).

in the second study, as it did in the first. A decrease in IL-6 (P < 0.05) and a tendency for IL-12 expression to increase (P = 0.05) in the ascending colon was observed for the 5% UWS, but not with the 5% TWS treatment. These results disagree with those of Pie et al. (2007), who observed that dietary fermentable fiber increased colonic IL-6 and with Weber et al. (2008) who reported no change in IL-6 cytokine expression in the colon. Our results suggest that treatment of the wheat straw changes potential health benefits of 'natural' wheat straw.

In experiment three, mucosal *Lawsonia Intracellularis* titers confirmed an active immune response to the vaccination challenge. Gene expression of mucosal markers did not differ in the ileum. Only two differences in colonic mucosal expression were identified, increases in the pro-inflammatory cytokines TNF- $\alpha$  and IL-6 (P < 0.05). An increase in the pro-inflammatory cytokine IL-6 in the colon of the weaned pig with the feeding of dietary fiber has been reported previously by Pie et al. (2007). But these findings disagree with the results of Lewton et al. (2019) who reported no change in TNF- $\alpha$  and a decrease in IL-6 expression. We are unsure of the mechanisms for why two pro-inflammatory cyto-

kines increased in the ascending colon with the *Lawsonia intracellularis* vaccine-challenge. Both may have increased as they participate in the acute phase response.

*Circulatory Markers.* Plasma Ig-A was measured in all three experiments (Table 4). In experiment one we observed that pigs fed 5% TWS had greater Ig-A concentration than pigs fed the Control treatment (P < 0.05). With a vaccination challenge in experiment three, plasma Ig-A concentrations were two-fold greater than those measured in the two previous experiments, but not affected by the feeding of TWS. Serum PCV2 titers confirmed an active immune response to the PCV2 vaccination. The greater Ig-A response caused by the vaccination could not be increased further by the feeding of dietary fiber.

In experiment one only, MHC class II expression in PBMC was measured (Table 4). When stimulated with lipopolysaccharide, 10% TWS increased (P < 0.05) MHC class II expression relative to the Control treatment measurement. In the immune challenge experiment only, plasma iFABP was measured and was not different for the Control and 5% TWS treatment groups (Table 4).

Table 4. Effect of 5 and 10% treated wheat straw (TWS) and(or) 5% untreated wheat straw (UWS) on amounts of immunological markers in plasma of pigs.

	Study One				Study Two				Study Three		
Item	Control	5% TWS	10% TWS	SEDMa	Control	5% UWS	5% TWS	SEM	Control	5% TWS	SEDMa
lg-A, mg/mL	0.47 <sup>ab</sup>	0.53 <sup>a</sup>	0.35 <sup>b</sup>	0.059	0.48 <sup>a</sup>	0.48 <sup>a</sup>	0.73 <sup>b</sup>	0.083	1.21	1.44	0.237
MHCII, LPS	138 <sup>a</sup>	155 <sup>a</sup>	215 <sup>b</sup>	35.1	NM	NM	NM	NM	NM	NM	NM
iFABP, pg/mL	NM	NM	NM	NM	NM	NM	NM	NM	355	361	26.5

<sup>a</sup> Standard error of the difference between means using an unpaired t-test.

a,bValues in a common row lacking a common superscript differ (P < 0.05). NM stands for "not measured."

### Conclusions

At this writing, we are exploring reasons for the effects of dietary wheat straw on intestinal and systemic markers observed in our experiments. Where in the intestine mRNA expression is being affected by dietary fiber varies among studies. Different findings among studies may be due to different fibrous feedstuffs fed and their differing characteristics such as proportions of soluble and insoluble fibers. Pie and others (2007) stated that their results demonstrated a strong link between fermentation end-products and the regulation of cytokines. Also differing could be age at weaning, the duration of fiber consumption prior to tissue sampling, and the current health status of pigs. The importance of intestinal microbiota is proposed, but what it should be and how it can be controlled with dietary fiber remains uncertain.

Use of an immune-challenge model adds further complexity to understanding alterations in local and systemic immune markers. The pig's response to a vaccination-challenge differs from that experienced in an active infection.

Weber and others (2008) concluded that variations in intestinal cytokine expression "are not necessarily associated with changes in growth performance or systemic inflammation." They also wrote that "contradictory roles for inflammatory cytokines within the intestine make it difficult to ascertain whether increased cytokine expression in response to dietary fiber is deleterious or beneficial." Based on the results of our experiments, we concur that dietary fiber can be fed without performance loss, even with improvements. Dietary fiber impacts the expression of immune markers locally in the gut and throughout the body of the young pig. The health benefits of dietary fiber diet depend on the health status of the pig, leaving practices of including fibrous feedstuffs in nursery rations with intent to prevent or to alleviate impacts of illness associated with weaning of the pig unestablished as recommendations across all farms.

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# Nutritional Strategies for Coping with High Diet Cost for Grow-Finish Pigs

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### Summary

To cope with high diet costs there are several nutritional strategies that can be utilized that include diet formulation, ingredients, feed processing, and feed additives. Diet formulation should focus on ensuring expensive nutrient levels such as energy, lysine, and phosphorus are set at economic optimum. Corn coproducts, alternative ingredients, and synthetic amino acids should be utilized given the opportunity to lower diet costs. However, it is important that sources of these ingredients are well understood so pig performance is predictable. From a feed processing standpoint reducing corn particle size can improve feed efficiency and lower feed costs. Like particle size there is a significant value proposition with pelleting when feed prices are high. Feeding pelleted diets results in improvements in growth performance and feed efficiency. Other economic advantages to pelleting include: the ability to reduce particle size of cereal grains and have it flow well through feeding systems, increased use of lower cost alternative ingredients that may not flow in meal form, and lower transportation costs. Lastly, nutritionists must consider the use of feed additives as these technologies can result in significant feed cost savings when biological responses are adequately understood and, consequently, they are appropriately applied within a production system.

### Introduction

The cost of feeding a pig from weaning to market has effectively doubled since the summer of 2020. Higher feed costs are attributed to commodity ingredient inflation for all the major feedstocks with corn, soybean meal, and dried distiller's grains with solubles (DDGS) effectively doubling in price. Interestingly, the price of fat has tripled due to the increased demand from the biofuels industry. Given future capacity needs for renewable diesel production the price of fat will likely remain high, and its use will be limited in swine diets. To cope with high diet costs there are several nutritional strategies that can be utilized to include: least cost diet formulation, feed ingredients, feed processing, and feed additives.

### **Diet Formulation**

A key decision in diet formulations with current energy costs is where to set dietary energy levels. The economics of where to set energy levels will be specific for a given production system and will depend on whether pigs are marketed on a fixed time versus fixed weight basis. Furthermore, an understanding of the pig's response to dietary energy is required. Economic models to determine the optimum energy level for a production system are often based on margin over feed or cost per unit of weight gain calculations. Variables to consider for these calculations include costs for ingredients, manufacturing and delivery; carcass payment, and biological response to energy. These models are either stand alone or have been incorporated directly into the formulation system. The use of these models for diet formulations is imperative considering the costs of dietary energy as energy levels that result in optimum feed efficiency may not always result in the most economical or safest feeding program. Economic models can be applied to other expensive nutrients like amino acids and phosphorus as well. The Department of Animal Sciences and Industry at Kansas State University has developed multiple economic models that are readily available to nutritionists and can be found at https://www.asi.k-state.edu/extension/swine/calculators.html.

### Feed Ingredients

With least cost diet formulation programs, a nutritionist can easily and quickly determine which of innumerable feedstock options will be the most economical. Given the increased availability and cost savings opportunity there has

Table 1. Key nutrients contained in DDGS sources with varying oil content

DDGS Oil Content, %							
> 10%	> 6 and < 9%	< 4%					
89.31	89.35	89.25					
27.33	27.36	27.86					
10.43	8.90	3.57					
4.11	4.04	4.64					
32.50	30.46	33.75					
11.75	12.02	16.91					
6.73	9.63	10.00					
2384	2343	2009					
	89.31 27.33 10.43 4.11 32.50 11.75 6.73	> 10%       > 6 and < 9%         89.31       89.35         27.33       27.36         10.43       8.90         4.11       4.04         32.50       30.46         11.75       12.02         6.73       9.63					

Source: NRC (2012).

 Table 2. Maximum Synthetic L-Lysine by Phase of Growth to Prevent Performance Loss<sup>1</sup>

Grow-Finish Diet	Start Wt. (kg)	End Wt. (kg)	Max L-Lys HCl, %
Diet 1	24	36	0.60
Diet 2	36	46	0.55
Diet 3	46	55	0.50
Diet 4	55	72	0.40
Diet 5	72	87	0.35
Diet 6	87	100	0.30
Diet 7	100	Market	0.25

<sup>1</sup>Source: Borg et al. (2023).

been an increase in the use of biofuel coproducts such as DDGS. The inclusion of DDGS is limited by anti-nutritional factors (i.e., mycotoxins), quality (i.e. nutrient profile and heat damage), fiber concentration, and for finishing pigs, also by the concentration of unsaturated fatty acids that impact pork fat quality, specifically pork bellies for bacon production. However, most groups of swine can be fed at least 15-20% DDGS and, sometimes, greater concentrations can be used. With the financial incentive for ethanol plants to further extract oil from DDGS there is considerable variation in nutrient content across different ethanol plant sources. This point is illustrated in Table 1 indicating considerable differences in nutrient content for DDGS sources with varying levels of oil removal. Thus, when utilizing higher concentrations of DDGS it is imperative that diets are carefully formulated with appropriate nutrient loading values by plant source.

Though it is not a new option there has also been increased usage of alternative ingredients such as bakery meal in diet formulations with the higher feed cost environment. Bakery meal is a mixture of bread, cookies, cake, crackers, and dough. This product is similar to corn in protein and amino acid composition but higher in fat. The salt content of bakery meal may be high, and the standard salt supplementation can be reduced. A typical inclusion level in finishing pigs is 10-20% of the diet as higher levels can create feed flowability issues particularly when diets are fed in meal form. Like DDGS there can be considerable variation in bakery sources and even within the same source.

With the economic competitiveness of synthetic amino acids over the last decade, significantly higher levels have been used in swine diets. With feed invoice price being a driving force to increase synthetic amino acid levels it is imperative that nutritionists understand the consequence of exceeding synthetic L-Lysine limits on growth and feed conversion. The consequence of exceeding these limits seems to be greater in late finishing diets. Thus, in practice, nutritionists feed higher levels of synthetic L-Lysine in the earlier dietary phases and then decrease the level as the pig advances to market weight. Example guidelines for maximum synthetic lysine by phase of growth to prevent performance losses are shown in Table 2.

### **Feed Processing**

### Particle Size

The impact of corn particle size on pig performance is well established. Wondra et al. (1995) evaluated the impact of corn particle size on the growth performance of growfinish pigs and found that for each 100-micron change in particle size feed conversion changed by approximately 1.2 %. Improvements in feed conversion due to corn particle size reduction can be attributed to improvements in energy digestibility because of an increase in surface area allowing better access for enzyme digestion. This improvement in feed conversion can deliver substantial returns. For example, assuming feed cost of \$0.059/kg of feed and a feed conversion increase of 1.2% for every 100-micron change in particle size, the opportunity cost is \$0.99 per pig (Table 3). Due to the impact of particle size on the energy value of corn this should be accounted for when formulating diets to maintain the target lysine to energy ratios. Similar consideration should be given to the other major ingredients where particle size may be reduced further. With a reduction in particle size, it's important to consider the impact on feed flowability at the farm level. As a general guideline, a corn particle size of 500-to-550-micron size is best for diets fed in meal form. This particle size allows feed to flow through feeding systems while allowing pigs to digest efficiently. If pellet diets are being fed, then further reductions in particle size can be utilized.

### Pelleting

Historically, pelleting of diets has been a common practice in geographical locations where feed costs are higher such as the Southeast United States. Pelleting of diets has been shown to improve average daily gain (Range = 1.04 to 4.43%; Average = 2.96%) and feed efficiency (Range = 5.35 to 12.29%; Average = 7.61%) in finishing pigs (Wondra et al. 1995, PIC Tech Memo 309, 2004; Nemechek et al. 2015; Dejong et al. 2016). The improved feed efficiency of pelleted diets is independent of corn particle size with the magnitude of response influenced by factors such as pellet quality and the percentage of fines in the feed. Diet type can have a profound impact on pellet quality so it's important that adjustments are made to the pelleting process and (or) equipment to ensure a high-quality pellet. Since 2007 with expansion of the ethanol industry and higher feed costs there has

been renewed interest in pelleting by Midwest swine producers as well. Where adoption of pelleted diets has been limited is in cases where performance improvements are not great enough to compensate for the added cost of pelleting and (or) an increase in pig mortality. From a health perspective, pelleted diets have been shown to increase the incidence of ulcers in finishing pigs (Wondra et al. 1995), which can ultimately lead to increases in mortality (Friendship, 2004). Like particle size there is a significant value proposition with pelleting when feed prices are high. For example, assuming feed cost of \$0.059/kg of feed and a feed conversion improvement of 5.35% with pelleting costs included the net margin is \$2.47 per pig (Table 4). Other economic advantages to pelleting include: the ability to reduce particle size of cereal grains and have it flow well through feeding systems, increased use of lower cost alternative ingredients that may not flow in meal form, and lower transportation costs.

## Feed Additives

With today's high feed cost nutritionists must consider the use of feed additives. Feed additives have been used extensively in swine diets because of their known biological impacts. Commonly used feed additives used in swine diets include antimicrobials, ionophores, enzymes, acidifiers, essential oils, direct fed microbials, select vitamins and miner-

#### Table 3. Impact of particle size on swine feed efficiency

		Corn Particle Size	
	500	600	700
\$0.059/kg feed cost			
Initial weight, kg	22.68	22.68	22.68
Selling weight, kg	131.55	131.55	131.55
Growth rate, kg/d	0.839	0.839	0.839
Feed:Gain	2.65	2.68	2.71
Feed cost, \$/pig	\$82.68	83.67	84.66
Difference, \$/pig	\$0.00	\$0.99	\$1.98

Assumptions: Feed: Gain increases 1.2% for every 100-micron change in particle size (Wondra et al. 1995).

	Meal	Pellet	Difference
\$0.059/kg feed cost			
Initial weight, kg	22.68	22.68	
Selling weight, kg	131.55	131.55	
Growth rate, kg/d	0.839	0.839	
Feed:Gain, lb/lb	2.65	2.51	0.14
Feed cost, \$/pig	\$82.68	\$80.21	\$2.47

particle size (Dejong et al. 2016) and pellet manufacturing cost of \$7.17 per metric ton.

als, and yeasts. In a recent review by Rao et al. 2023 the impact of feed additives on growth performance and carcass characteristics were summarized. The categories of feed additives in this review included: acidifiers, betaine, chromium, conjugated linoleic acid, copper, direct fed microbials, carbohydrases, proteases, phytases, multi-enzymes, essential oils, L-carnitine, yeasts, and zinc. These additive categories all provide different mechanisms of action that can potentially improve growth performance and (or) feed efficiency. For growth performance, direct fed microbials, copper, L-carnitine, and multi-enzymes showed relatively large (>2.1% improvement) and positive effects. For feed efficiency, acidifiers, betaine, conjugated linoleic acid, multienzymes, direct-fed microbials, L-carnitine, and yeasts showed relatively large (>2.5% improvement) and positive effects. It is important to note that this review did not include antimicrobials or ionophores categories, which have also been shown to improve growth performance and feed efficiency as well. When considering the use of feed additives there are 3 key things to consider: 1) evaluate additives that have the ability to improve feed efficiency based on their mechanism of action with appropriately designed experiments; 2) given variability in response utilize a metaanalysis approach in evaluating the overall value proposition of a feed additive technology; and 3) ensure the feed additive works on top of your current diet program.

### Conclusions

Diet formulation should focus on ensuring expensive nutrient levels such as energy, lysine, and phosphorus are set at economic optimum. Thus, the use of economic models can help nutritionists determine the optimum level for these nutrients. Given the increased availability of corn coproducts, alternative ingredients, and synthetic amino acids should be utilized given the opportunity to lower diet costs. However, it is important that sources of these ingredients are well understood so pig performance is predictable. From a feed flow standpoint reducing corn particle size to 500-to-550-micron size is best for diets fed in meal form. This particle size allows feed to flow through feeding systems while allowing pigs to digest efficiently. If pellet diets are being fed, then further reductions in particle size can be utilized. Feeding pelleted diets results in improvements in growth performance and feed efficiency. If pelleting capacity is available this practice should be considered to help lower feed costs. Lastly, nutritionists must consider the use of feed additives as these technologies can result in significant feed cost savings.

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# Advantages of Higher Soybean Meal Diets for Pigs

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### **Summary**

Crystalline amino acids (AA) are widely used in diets for pigs to either lower the concentration of soybean meal (SBM) in the diets or to balance the AA profile of other protein sources including high protein corn protein. However, results of recent experiments demonstrated that even when diets are balanced for all indispensable AA, the excess levels of Leu in corn protein results in reduced growth performance of pigs. This is likely due to interactions between Leu and Val, Ile, and Trp and it is, therefore, possible to partly restore the lost performance obtained for diets with corn protein if extra Val, Ile, and Trp, is included. However, it appears that pigs fed diets based on corn and SBM have better growth performance than those fed corn protein and crystalline AA. It has also not been possible to demonstrate that low crude protein diets that contain crystalline AA instead of some of the SBM have greater net energy than diets based only on corn and soybean meal. It therefore appears that SBM may provide benefits to pigs in addition to the AA that are provided from SBM.

## Introduction

Utilization of feed-grade crystalline amino acids (CAA) and the increased availability of grain co-products and alternative feed ingredients have led to replacement of soybean meal (SBM) in diets for pigs. One reason for the success of using CAA at the expense of SBM is that diets containing CAA have a lower concentration of crude protein (**CP**) compared with SBM-based diets. There is a common conception in the industry that diets with lower concentration of CP have greater net energy (NE) compared with diets with greater CP. As a consequence, it is believed that diets based on CAA contain more NE than diets based on SBM. However, this perception is based on theoretical calculations and there is no scientific evidence to support this. As a matter of fact, in a few experiments where diets based on CAA were compared with diets without CAA and with more SBM, it was not possible to demonstrate a difference in NE (Noblet et al., 2001; Munoz, 2020). However, these experiments were not specifically designed to determine the effects of replacing SBM by CAA and the perception that diets based on CAA contain more NE is therefore still widespread.

In recent years, high protein corn co-products (**HPCP**) that contain between 40 and 50% CP have been developed. It is therefore possible to formulate diets containing corn

co-products as a protein source, but such diets will contain more than twice as much Leu as recommended. Leucine is an indispensable AA that stimulates the catabolism of branched-chain amino acids (**AA**) in the liver (Harper et al., 1984). However, if pigs are fed diets with excessive Leu, degradation of not only Leu, but also Val and Ile, may increase because of the stimulating effects of Leu or its metabolite on branched-chain AA degrading enzymes (Wiltafsky et al., 2010). Therefore, excess dietary Leu may result in an imbalanced supply of branched-chain AA by increased degradation of Val and Ile and thus reduce pig feed intake, protein synthesis, and growth performance (Gatnau et al., 1995; Wiltafsky et al., 2010; Kwon et al., 2019).

Therefore, the objective of this contribution was to investigate the effects of using SBM in pig diets on the growth performance of pigs and concentration of NE in diets.

## Effects of using SBM in diets fed to growing pigs

# *Effects of excess Leu from HPCP on growth performance of pigs*

An experiment was conducted to test the hypothesis that excess Leu from corn protein will result in negative effects on the growth performance of growing pigs and that crystalline  $_{L}$ -Val,  $_{L}$ -Ile, or  $_{L}$ -Trp will alleviate the negative effects of the second second

Table 1. Growth performance and plasma urea nitrogen (PUN) of growing pigs fed experimental diets<sup>1,2</sup>

ltem <sup>3</sup>			High-protein corn product diets								
	SBM diet	Basal (no CAA)	+ lle	+ Val	+ Trp	+ Ile, Val	+ Ile, Trp	+ Val, Trp	+ Ile, Val, Trp	SEM	P-value
Body weight, kg											
d 1	28.7	28.5	28.7	28.7	28.4	28.5	28.7	28.5	28.5	0.83	0.969
d 28	55.3 <sup>a</sup>	50.7 <sup>b</sup>	50.7 <sup>b</sup>	51.0 <sup>b</sup>	50.7 <sup>b</sup>	50.2 <sup>b</sup>	50.0 <sup>b</sup>	52.6 <sup>ab</sup>	51.3 <sup>b</sup>	1.35	< 0.001
ADG, g/d	950 <sup>a</sup>	793 <sup>b</sup>	785 <sup>b</sup>	797 <sup>b</sup>	797 <sup>b</sup>	776 <sup>b</sup>	760 <sup>b</sup>	862 <sup>ab</sup>	813 <sup>b</sup>	28.8	< 0.001
ADFI, g/d	1,816 <sup>a</sup>	1,621 <sup>b</sup>	1,623 <sup>b</sup>	1,650 <sup>ab</sup>	1,704 <sup>ab</sup>	1,617 <sup>b</sup>	1,683 <sup>ab</sup>	1,758 <sup>ab</sup>	1,695 <sup>ab</sup>	57.7	0.034
G:F	0.53	0.49	0.48	0.48	0.47	0.48	0.45	0.49	0.48	0.015	0.079
PUN, mg/dL											
d 1	9.0	7.6	8.3	8.3	8.0	7.4	8.0	8.3	8.1	0.45	0.486
d 14	7.9	8.0	7.6	8.9	7.5	8.5	7.8	8.8	7.4	0.71	0.773
d 28	12.0 <sup>a</sup>	8.1 <sup>b</sup>	8.6 <sup>b</sup>	8.1 <sup>b</sup>	8.8 <sup>ab</sup>	7.6 <sup>b</sup>	8.3 <sup>b</sup>	8.3 <sup>b</sup>	7.8 <sup>b</sup>	0.76	0.004
HS, μg/mL	0.193	0.149	0.183	0.178	0.191	0.180	0.183	0.201	0.188	0.013	0.070
Trp, μmol/L	60.5 <sup>ab</sup>	28.4 <sup>c</sup>	43.9 <sup>bc</sup>	40.2 <sup>bc</sup>	61.6 <sup>ab</sup>	45.9 <sup>bc</sup>	62.3 <sup>ab</sup>	79.9 <sup>a</sup>	59.4 <sup>ab</sup>	7.04	< 0.001

a-bMeans within a row without a common superscript letter differ (P < 0.05).

<sup>1</sup>Each least squares mean represents 8 observations.

<sup>2</sup>Data are from Kwon et al. (2023).

<sup>3</sup>ADG = average daily gain; ADFI = average daily feed intake; CAA = crystalline amino acids; G:F = gain to feed ratio; HS = hypothalamic serotonin; SBM = soybean meal.

fects of excess dietary Leu (Kwon et al., 2023). A basal diet based on corn and a HPCP was formulated. Two levels of crystalline L-Ile (0 or 0.10%), 2 levels of crystalline L-Val (0 or 0.10%), and 2 levels of crystalline L-Trp (0 or 0.05%) were added to the basal diet for a total of 8 diets. A SBM diet based on corn and SBM was also used in addition to the 8 diets. A total of 288 growing pigs [initial body weight (**BW**): 28.6 kg; SD = 2.5] were randomly assigned to 9 dietary treatments in a 28-day growth performance experiment. Pigs were housed in pens with partly slatted concrete floors and average daily gain (**ADG**), average daily feed intake (**ADFI**), and gain to feed ratio (**G:F**) were calculated. Blood samples were collected and analyzed for plasma urea nitrogen (**PUN**). After blood collection, all pigs were euthanized and the hypothalamus was collected for serotonin analysis.

Results (Table 1) indicated that final BW and ADG of pigs fed the SBM diet were greater (P < 0.05) than for pigs fed all other diets, except pigs fed the diet with the addition of both L-Val and L-Trp. Average daily feed intake of pigs fed the SBM diet was greater (P < 0.05) compared with the diet containing HPCP and no CAA and the diet containing HPCP and Ile. Gain to feed ratio of pigs and hypothalamic serotonin tended (P < 0.10) to be different among dietary treatments, but pairwise comparisons were not significant. There was no difference among dietary treatments for PUN on d 1 or d 14, but on d 28, PUN of pigs fed the SBM diet was greater (P < 0.05) than that of pigs fed all other diets except the diet supplemented with L-Trp. Pigs fed the basal diet had lower (P < 0.05) free Trp in plasma compared with pigs fed all other diets except for pigs fed the diets with added Val, Ile, or Val and Ile.

It was concluded that pigs fed the SBM diet had better growth performance than pigs fed the diets containing HPCP. The combination of Val and Trp supplementation may be beneficial for preventing detrimental effects of excess Leu on growth performance of pigs.

## *Effects of excess Leu from HPCP and use of CAA on growth performance and intestinal tract function of weanling pigs*

A follow-up experiment was conducted to test the hypothesis that the use of HPCP without or with CAA affects the growth performance and intestinal tract function of weanling pigs (Mallea et al., 2023). A two-phase feeding program was used. A total of 320 weanling pigs (initial BW: 6.1 kg; SD = 0.7) were randomly assigned to 10 dietary treatments. A SBM diet based on corn and SBM was formulated and a basal diet was formulated based on corn and 10% HPCP. Another basal diet was formulated based on corn and 20% HPCP. Two levels of crystalline L-Ile (0 or 0.10%), 2 levels of crystalline L-Val (0 or 0.10%), and 2 levels of crystalline L Trp (0 or 0.05%) were added to the basal diet with 20% HPCP for a total of 8 diets. Pigs were housed in pens with fully slatted plastic floors and ADG, ADFI, and G:F were calculated. Fecal scores were assessed visually. Blood samples were collected from one pig in each pen and analyzed for PUN, total protein, albumin, peptide YY (PYY), and immunoglobulin G (IgG). Ileal tissue samples were also collected and villus height, crypt depth, and lamina propria thickness were measured.

Results (Table 2) indicated that the inclusion of 10 or 20% HPCP in diets reduced (P < 0.05) final BW on d 28 and

Table 2. Growth performance of weanling pigs fed the experimental diets<sup>1,2</sup>

ltem <sup>2</sup>	CDM	100/				20%	HPCP					
	SBM diet	10% HPCP	-	+ lle	+ Val	+ Trp	+ Ile, Val	+ Ile, Trp	+ <mark>Val</mark> , Trp	+ Ile, Val, Trp	SEM	P-value
BW, kg												
d 1	6.33	6.29	6.34	6.33	6.30	6.32	6.30	6.32	6.31	6.29	-	-
d 14	7.43	7.27	7.35	7.21	7.35	7.2	7.35	6.97	7.40	7.43	0.35	0.979
d 28	14.23 <sup>a</sup>	12.58 <sup>bcd</sup>	12.07 <sup>cd</sup>	11.89 <sup>cd</sup>	13.03 <sup>abc</sup>	11.93 <sup>cd</sup>	12.60 <sup>bcd</sup>	11.44 <sup>d</sup>	13.00 <sup>abc</sup>	13.78 <sup>ab</sup>	0.55	0.009
ADG, g/d												
Phase 1	78	70	72	63	75	63	75	47	78	81	9	0.280
Phase 2	486 <sup>a</sup>	380cde	337de	334de	405 <sup>bc</sup>	338de	375cde	319 <sup>e</sup>	400bcd	453 <sup>ab</sup>	24	< 0.001
Overall	282a	225cd	205cde	199de	240bc	200cde	225cd	183e	239bc	267 <sup>ab</sup>	14	< 0.001
ADFI, g/d												
Phase 1	142	137	136	138	141	136	149	119	146	158	10	0.245
Phase 2	662 <sup>a</sup>	550 <sup>cd</sup>	503cde	488de	572 <sup>bcd</sup>	500cde	550 <sup>cd</sup>	442 <sup>e</sup>	578 <sup>abc</sup>	647 <sup>ab</sup>	34	< 0.001
Overall	402 <sup>a</sup>	343 <sup>b</sup>	319 <sup>bc</sup>	313 <sup>bc</sup>	357 <sup>ab</sup>	318 <sup>bc</sup>	350 <sup>ab</sup>	280 <sup>c</sup>	362 <sup>ab</sup>	402 <sup>a</sup>	19	< 0.001
G:F												
Phase 1	0.55	0.49	0.53	0.45	0.52	0.46	0.49	0.36	0.52	0.52	0.06	0.180
Phase 2	0.73	0.69	0.67	0.69	0.71	0.67	0.68	0.73	0.70	0.70	0.03	0.347
Overall	0.70	0.65	0.64	0.64	0.67	0.62	0.64	0.66	0.66	0.66	0.02	0.154

<sup>1</sup>Data are least square means of 8 observations per treatment.

<sup>2</sup>Data were from Mallea et al. (2023).

<sup>3</sup>ADFI = average daily feed intake; ADG = average daily gain; BW = body weight; G:F = gain to feed ratio; HPCP = high protein corn coproducts.

ADG and ADFI in phase 2 and for the entire experimental period compared with the SBM diet. Final BW of pigs fed the 20% HPCP diet with the 3 CAA was greater (P < 0.05) compared with the 20% HPCP diet containing no CAA, the HPCP diets with Ile, Trp, or Ile and Trp. Average daily gain and ADFI in phase 1 were not affected by dietary treatment. However, the ADG of pigs fed the 20% HPCP diets with the 3 CAA was greater (P < 0.05) in phase 2 and for the overall experiment compared with the 10% and 20% HPCP diets with no CAA and the 20% HPCP diets with Ile, Trp, Ile and Val, or Ile and Trp. Pigs fed the 20% HPCP diet with the 3 CAA had greater (P < 0.05) ADFI in phase 2 and for the overall experiment than pigs fed diets with 10 or 20% HPCP containing no CAA or the HPCP diets with Ile, Trp, or Ile and Trp. Gain:feed was not affected by dietary treatment. In phase 2 and overall, fecal scores (Table 3) were reduced (P < 0.05) if HPCP was used instead of SBM. No differences among experimental diets were observed for PUN, total protein, albumin, peptide YY, or IgG on d 14 (data not shown) and for total protein, peptide YY, and immunoglobulin G on d 28. Plasma urea N of pigs fed the SBM diet on d 28 was greater (P < 0.05) compared with the HPCP diets supplemented with Ile, Val, Val and Trp, or Ile, Val, and Trp. Albumin in plasma was greater (P < 0.05) in pigs fed the SBM diet than in pigs fed the HPCP diets supplemented with Ile, Trp, Ile and Val, Ile and Trp, or Val and Trp, but was not different from pigs fed the two HPCP diets with no CAA and the HPCP diets supplemented with Val or Ile,

Val, and Trp. Ileal villus height, crypt depth, villus height to crypt depth ratio, and lamina propria thickness, as well as microbial protein, concentrations of VFA and ammonium in feces were not affected by dietary treatments (data not shown).

It was concluded that using HPCP instead of SBM in diets for weanling pigs has a negative effect on growth performance, but ileal morphology, microbial protein, and fecal VFA and ammonium were not affected. The detrimental effect on growth performance of pigs fed HPCP-based diets was partially ameliorated with the supplementation of Ile, Val, and Trp.

### Determination of net energy in low protein diets containing CAA and normal protein diets fed to growing pigs

This experiment was conducted to test the hypothesis that NE in diets is not increased by increasing CAA in diets fed to group-housed pigs (Lee et al., 2023). First, a diet based on corn and SBM, minerals, and vitamins was formulated; this diet contained no CAA. Then, a normal protein diet was formulated based on corn and SBM, minerals, and vitamins and crystalline Lys, Met, and Thr; this diet was similar to a typical diet used in the industry. Four additional diets were formulated by reducing CP by 1, 2, 3, or 4% units compared with the normal-CP diet containing three CAA. Therefore, a total of six diets were used. A total of 24 growing pigs (initial BW: 29.9 kg; SD = 2.4) were randomly allo-

Table 3. Fecal score and	plasma characteristics of pigs <sup>1,2</sup>
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Item <sup>3</sup>	CDM	100/				20%	НРСР					
	SBM diet	10% HPCP	-	+ lle	+ Val	+ Trp	+ lle, Val	+ lle, Trp	+ Val, Trp	+ Ile, Val, Trp	SEM	P-value
Fecal score <sup>4</sup>												
Phase 1	2.42	2.25	2.12	2.03	2.17	2.03	2.51	2.08	2.21	2.11	0.21	0.295
Phase 2	1.67ª	1.44b	1.23c	1.23c	1.21c	1.37 <sup>bc</sup>	1.26 <sup>bc</sup>	1.26 <sup>bc</sup>	1.21c	1.33 <sup>bc</sup>	0.13	0.001
Overall	2.05 <sup>a</sup>	1.84 <sup>abc</sup>	1.67 <sup>bc</sup>	1.63 <sup>c</sup>	1.75 <sup>bc</sup>	1.70 <sup>bc</sup>	1.89 <sup>ab</sup>	1.67 <sup>bc</sup>	1.71 <sup>bc</sup>	1.72 <sup>bc</sup>	0.14	0.030
d 28												
PUN, mg/dL	6.25 <sup>a</sup>	4.62 <sup>abc</sup>	4.50 <sup>abc</sup>	4.00 <sup>bcd</sup>	3.62 <sup>cd</sup>	5.00 <sup>abc</sup>	4.62 <sup>abc</sup>	5.62 <sup>ab</sup>	2.59 <sup>d</sup>	2.37 <sup>cd</sup>	0.66	0.006
Total protein, mg/dL	4.82	4.86	4.68	4.71	4.83	4.75	4.60	4.66	4.62	4.80	0.10	0.390
Albumin, mg/dL	3.18 <sup>a</sup>	3.12 <sup>ab</sup>	2.97abcde	2.85cde	3.03abcd	2.83de	2.72e	2.86bcde	2.82 <sup>de</sup>	3.11abc	0.09	0.009
PYY, ng/mL	2.69	2.72	2.79	2.75	2.96	2.72	2.96	3.16	2.95	2.94	1.03	0.971
lg G, mg/mL	6.59	4.59	4.61	5.64	4.59	4.93	4.68	4.70	4.71	3.91	0.91	0.679

<sup>1</sup>Data are least square means of 7 to 8 observations per treatment.

<sup>2</sup>Data were from Mallea et al. (2023).

<sup>3</sup>PUN = plasma urea nitrogen; immunoglobulin G = IgG; peptide YY = PYY.

<sup>4</sup>Fecal scores were visually assessed every other day by 2 independent observers for 28 days. Fecal score: 1, normal feces; 2, moist feces; 3, mild diarrhea; 4, severe diarrhea; and 5, watery diarrhea.

cated to six calorimeter chambers in the Swine Calorimeter Unit at the University of Illinois. There were four pigs per chamber. The six chambers were allotted to six diets using a  $6 \times 6$  Latin square design with six periods. Therefore, there were six replicate chambers per diet. The O<sub>2</sub> consumption and CO<sub>2</sub> and CH<sub>4</sub> production were measured and fecal and urine samples were collected for 6 days. Fasting heat production (**FHP**) was also determined. Diets and ground fecal samples and lyophilized urine samples were analyzed for gross energy (**GE**) and urine samples were analyzed for N.

Results (Table 4) indicated that feed intake, fecal and urine GE output, the ATTD of dry matter and GE, total heat production (THP), FHP, and retained energy were not different between pigs fed the two normal-CP diets. Respiratory quotient (**RQ**) of pigs fed the normal-CP diet without CAA was less (P = 0.045) compared with the normal-CP diet with CAA. Concentrations of digestible energy (DE) and metabolizable energy (ME) in the normal-CP diet without CAA were greater (P < 0.05) than in the normal-CP diet with CAA, but there was no difference in concentration of NE between the two normal-CP diets. Feed intake, GE intake, and fecal GE output of pigs were linearly (P =0.044) increased by reducing CP in diets, but reducing CP in diets did not affect the ATTD of dry matter and GE or urine GE excretion by pigs. Total heat increment, FHP, retained energy, and concentrations of DE, ME, and NE were not affected by reducing CP in diets containing CAA.

### Discussion

The growth performance of both growing pigs and weanling pigs was negatively affected by the use of HPCP compared with diets containing SBM. One of the primary limitations of using corn co-products is its relatively high fiber concentration, which leads to reduced nutrient utilization in pigs (Woyengo et al., 2014). Excessive dietary Leu that is supplied by including corn protein is also cause for concern, as it stimulates the breakdown of branched-chain AA in skeletal muscle and liver (Harper et al., 1984). The increased Leu results in an increased production of the Leu metabolite,  $\alpha$ -keto isocaproate, which also increases the secretion of branched-chain  $\alpha$ -keto acid dehydrogenase enzyme complex that catalyzes the branched-chain AA (Wiltafsky et al., 2010). It is likely that it is the excess Leu that resulted in reduced growth performance of pigs fed diets containing HPCP (Kwon et al., 2019).

Levels of PUN are mostly influenced by the quantities and balance of AA that are absorbed by pigs (Nyachoti et al., 2006). The reason for the increased PUN in growing and weanling pigs fed the SBM diets compared with pigs fed the HPCP diets is likely a result of greater ADFI for pigs fed the SBM diet. In addition, PUN is often used as a response criterion in AA requirement studies because it responds rapidly to changes in dietary AA concentration and to changes in AA utilization efficiency in pigs (Coma et al., 1995). The reduced PUN that was observed as CAA was supplemented to the HPCP diets is likely a result of increased availability of Ile and Val, which ameliorated the imbalance among AA for protein synthesis that was caused by excessive Leu.

Ileal morphology, microbial protein, and fecal VFA and ammonium productions may vary depending on the composition of diets and it is possible that they are changed in pigs fed diets containing SBM or HPCP. Diets containing higher CP may increase the size and thickness of the intestinal villi of pigs because more AA need to be digested

Table 4. Effects of reducing crude protein (CP) on apparent total tract digestibility (ATTD) of dry matter (DM) and gross energy (GE) and
concentrations of digestible energy (DE), metabolizable energy (ME), and net energy (NE) in diets and total heat production (THP) and fast-
ing heat production (FHP) from group-housed pigs <sup>1,2</sup>

ltem <sup>3</sup>	Dietary CP	Normal		Low,	Low, CP reduction <sup>4</sup> (% unit)			Co	ntrast P-v	alue <sup>5</sup>	
		No CAA <sup>3</sup>	3 CAA	-1	-2	-3	-4	SEM	Normal	Linear	Quadratic
Feed intake, kg/	d	2.79	2.70	2.73	2.88	2.95	2.89	0.19	0.520	0.044	0.442
Fecal GE output	, kcal/d	1,267	1,247	1,275	1,289	1,352	1,313	78	0.712	0.078	0.547
ATTD of DM, %		89.65	89.73	89.72	90.18	89.88	90.14	0.26	0.801	0.141	0.783
ATTD of GE, %		88.00	87.79	87.75	88.14	87.82	87.93	0.31	0.565	0.664	0.662
Urine GE output	t, kcal/d	213	182	181	187	174	173	29	0.154	0.575	0.714
THP, kcal/BW <sup>0.6</sup> /	′d	384	375	378	376	390	377	20	0.544	0.621	0.704
FHP, kcal/ BW <sup>0.6</sup>	/d	223	220	235	218	238	221	17	0.834	0.878	0.481
Retained energy	, kcal/BW <sup>0.6</sup> /d	411	399	406	432	478	433	43	0.755	0.100	0.405
Respiratory quo	tient, fasted	0.66	0.64	0.63	0.64	0.64	0.67	0.03	0.136	0.112	0.217
Respiratory quo	tient, fed	0.99	1.03	1.01	1.05	1.05	1.04	0.04	0.045	0.199	0.444
Energy in diets,	kcal/kg										
GE		3,846	3,802	3,800	3,788	3,785	3,787	-	-	-	-
DE		3,384	3,337	3,335	3,339	3,324	3,330	12	0.004	0.381	0.932
ME		3,310	3,272	3,269	3,273	3,266	3,271	10	0.012	0.891	0.909
NE		2,646	2,605	2,663	2,631	2,665	2,634	47	0.413	0.559	0.392

<sup>1</sup>Each least squares mean represents 6 observations except normal CP diet and diet containing -2% CP (n = 5). <sup>2</sup>Data are from Lee et al. (2023).

 $^{3}BW = body weight; CAA = crystalline amino acids; CP = crude protein.$ 

<sup>4</sup>Dietary CP was reduced from the concentration of CP in the normal diet containing 3 CAA.

<sup>5</sup>Normal = normal CP diet without CAA vs. normal CP with CAA; linear = linear effects of reducing dietary protein; quadratic = quadratic effects of reducing dietary protein.

and absorbed by increasing surface area. However, diets containing high fiber may negatively impact ileal morphology by causing inflammation, villus atrophy, and reduced nutrient absorption. Productions of microbial protein, fecal VFA, and ammonium are a result of the fermentation of dietary fiber in the large intestine by gut microorganisms. Results from the study by Mallea et al. (2023) indicated that ileal morphology, microbial protein, and fecal VFA and ammonium productions were not changed by diets containing SBM or HPCP, which demonstrated that under the condition of this experiment, there were no major differences in fermentability or intestinal tract function by using SBM or HPCP in diets for pigs.

The greater RQ observed for pigs fed the normal-CP diet supplemented with CAA than for pigs fed the normal-CP diet without CAA is likely the result of the RQ for protein being lower than for carbohydrates. Because the normal-CP diet without CAA contained more SBM and thus a greater concentration of CP, the RQ in this diet was less compared with the normal-CP diet with CAA. Diets with a greater CP may lead to increased excretion of urine energy because of increased deamination of AA and thus increased urea production, which is the primary energy component in urine (Noblet et al., 2001).

Pigs fed diets containing AA close to the AA requirements would expend less energy on metabolizing AA to carbon skeletons and amino groups (Le Bellego et al., 2001; Brown-Brandl et al., 2004) than pigs fed diets with excess protein. Therefore, it was believed that if pigs were fed diets containing less SBM and greater CAA, the efficiency of ME would be increased. However, contrary to common belief in the industry that low-CP diets provide greater NE compared with high-CP diets, the results from the study by Lee et al. (2023) did not demonstrate an increased NE or an increased NE to ME ratio when dietary CP was reduced.

Concentrations of DE and ME in the normal-CP diet supplemented with three CAA were less than in the normal-CP diet without CAA. The differences in energy are attributed to the more corn and less SBM used in the normal-CP diet with three CAA, as corn contains less GE and DE than SBM. This difference resulted in the difference in ME in diets, but this was not observed for NE. Results from the study by Lee et al. (2023) also contradict the notion that the NE in corn is much greater than the NE in SBM because the inclusion of corn was increased and the inclusion of SBM was reduced as CP was reduced in diets. However, it has been reported that ME and NE in SBM are greater than published values (Sotak-Peper et al., 2015; Lee et al., 2021). The results of this study also indicated that the NE of SBM may be close to that in corn.

# Conclusions

In conclusion, pigs fed the SBM diet had better growth performance than pigs fed the diets containing HPCP. The inclusion of CAA in the HPCP diets partially improved the growth performance of pigs, but did not fully replicate the results observed with SBM-based diets. Plasma urea N of pigs fed the SBM was greater than that of pigs fed the HPCP diets. Ileal morphology, microbial protein, and fecal VFA and ammonium productions were not changed by diets containing SBM or HPCP. Diets containing low protein (i.e., low SBM and high crystalline AA) did not increase the concentration of NE by group-housed pigs. Therefore, the common conception in the industry that diets with low protein have greater NE compared with diets with greater protein needs to be corrected.

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# Peroxidized Lipids Increase Catabolism of Amino Acids and May Induce Additional Protein Damage

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### Summary

High cost of lipid sources and high temperature thermal treatment of feed ingredients increase the necessity to understand the impact that quality of lipids have on pig performance and health. An increasing set of evidence suggests that amino acids are catabolized towards production of metabolites needed to cope with oxidative stress when pigs are fed low quality peroxidized corn or soybean oil. In the pigs fed low quality peroxidized oils, there is increased catabolism of tryptophan in the kynurenine pathway, concomitantly threonine is spared from catabolism. Likewise, peroxidized lipids in high protein feed ingredients such as soybean products can contribute to protein damage and increase in protein carbonyl concentrations. It is important to revise current parameters of quality of lipid sources to integrate potential interventions in amino acid digestibility and requirements.

### Introduction

There is an increasing demand from consumers, nongovernmental organizations, and the federal administration to decrease the environmental footprint of livestock production. In response to these demands, organizations such as the National Pork Board have pledged to decrease the carbon footprint of pork production by 40% by 2030. The need to decrease the carbon footprint of pork production must also happen with concomitant improvements in other aspects of sustainability of pork production, such as improvements in animal health and the increased circularity of nutrients. There are tradeoffs and synergies among sustainability goals, pork production constraints, environment, consumer demands, antimicrobial resistance, animal health, and the demand for animal-based proteins. A systems approach is necessary to organize interventions of major areas for action in a coherent manner and to communicate with consumers, producers, and other areas of society.

Precision nutrition, decreasing the burden of diseases, increased waste recovery, and prevention of foreign animal diseases are proposed ways to improve sustainability via swine nutrition (Urriola, 2022). In addition to the challenges of decreasing the carbon footprint of production, we need to adapt swine nutrition programs to increase supply and availability of new feed ingredients (e.g., cover crop coproducts) and decrease the supply of traditionally well characterized ingredients (e.g., soybean oil diverted towards biofuels). This dynamic ingredient availability challenges our nutrition systems by demanding new ways to determine the nutritional value of ingredients (Figure 1). Currently, the price of traditional sources of lipids in the diets for pigs such as corn oil, soybean oil, and canola oil are at record levels due to the increased demand of these plant oils to produce renewable biodiesel. This increased diversion of oils away from animal feeding sets greater pressure on finding alternative sources for lipids, while the high price of lipid sources increases the need for precision nutrition to understand the quality of the sources of said lipids (Figure 1).

In addition to the soaring prices of feed ingredients, there is a necessity to recover nutrients in the food system from sources such as animal based-protein meals and food waste. All these sources of nutrients may contain infectious disease agents (e.g., African swine fever virus, porcine epidemic diarrhea virus) and pose a risk to the transmission of infectious diseases. Decreasing the transmission of infectious diseases may require the thermal treatment of feed ingredients to inactivate the pathogens at greater temperature than previously described (Shurson et al., 2022). Specifically, the thermal resistance of African Swine Fever virus may be greater than previously reported in the literature.

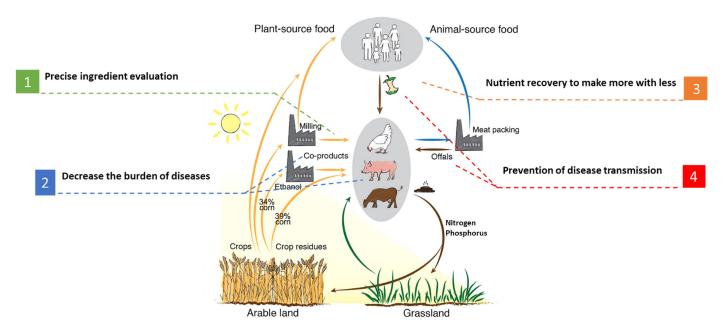


Figure 1. Four major nutritional interventions to increase the sustainability of pork production, modified from (Van Zanten et al., 2018).

Likewise, prolonged thermal treatment may be necessary to inactivate pathogens in plant-based feed ingredients such as soybean meal, in which viruses appear to have greater thermal resistance than in corn and corn coproducts. This thermal treatment of feed ingredients may increase lipid peroxidation and it is possible that thermally processed feed ingredients may contain a greater concentration of peroxidized lipids than previously measured.

There is a diverse set of chemical reactions that occur due to the heating of feed ingredients, and these reactions include the Maillard reaction (sugar-amino acid), crosslinking (protein-protein, lipid-protein), amino acid racemization, and amino acid degradation. These reactions and their products may induce beneficial properties to proteins such as solubilization, degradation of trypsin inhibitors, and the increase of amino acid digestibility. Although, other reactions may have deleterious effects on the nutritional value of the heated ingredient. Dietary oxidized lipids have the potential to directly interact with free amino acids and with the residual of base amino acids in protein feed ingredients, such as lysine. This causes the formation of adducts via Schiff bases and Amadori rearrangement with the consequence of decreased digestibility of amino acids (Meade et al., 2005).

Heat treated feed ingredients may have greater concentration of peroxidized lipids than non-heat-treated feed ingredients. The increased intake of peroxidized lipids poses a metabolic burden on animals by increasing oxidative stress (Hanson et al., 2016; Hung et al., 2019). The most prominent studied mechanism that swine nutritionists have available to assist pigs cope with oxidative stress is the provision of vitamin E and selenium. However, oxidative stress is diverse and the role of amino acids in ameliorating the impact of oxidative stress is less studied than selenium and vitamin E. Therefore, the first objective of this proceedings paper is to describe recent research describing the impact of peroxidized lipid consumption on metabolism of amino acids using a metabolomic based approach. In addition, peroxidized lipids interact with amino acids and the side residue to form carbonyl products. There are no systematic surveys of the prevalence of carbonyl products in soybean coproducts available to the swine industry in North America. Therefore, the second objective of this manuscript is to describe results from a recent survey of the concentration of carbonyl products in soybean products and their association with other dietary components.

# Research on amino acid metabolism during the peroxidized lipid oxidative challenge

# Peroxidized lipids decrease growth performance and the health of pigs

Feeding pigs diets containing ingredients with a high concentration of peroxidized lipids linearly decreases the growth performance of nursery pigs (Hung et al., 2017). The observed decrease in average daily gain and average daily feed intake is in contrast with studies undertaken under commercial conditions. Under commercial feeding conditions, nursery pigs may be raised under greater stocking density, more disease pressure, and more challenging diets than pigs in university research facilities. Under these commercial conditions, the consumption of peroxidized lipids did not decrease the average daily gain or the feed efficiency of the average pigs in the pen. However, feeding peroxidized oil increases pig removal rate, increases weaned pig mortality, and decreases the proportion of full-value pigs (Chang et al., 2019). All these metrics are typically not estimated in conventional experimental settings but are of foremost importance to the economic profit of pork producers. Therefore, while the magnitude of impact of peroxidized lipids on average daily gain is variable, the impact on pig health is substantial and requires investigation on the metabolic effects of peroxidized lipid consumption.

## *Peroxidized lipids increase catabolism of tryptophan in mice models*

Mice (n = 16; C57BL/6) were separated into two dietary treatments and fed a control diet of fresh refined sov-

bean oil and a diet containing heat treated peroxidized soybean oil. Oil was oxidized by heating it, in which the temperature was increased from 22 °C to 185 °C gradually, and then held constant at 185 °C for 6 h. Oxygen was injected by bubbling air (50 ml/min). Comparative chemometric analysis of the control feed and the oxidized corn oil feed indicates that oxidized oil contained a lower concentration of triglycerides containing polyunsaturated fatty acids compared with saturated fatty acids (Wang et al., 2018). Secondary break-down products of the peroxidized lipids were greater in the oxidized oil than in the control oil as indicated by a greater concentration of TBARS and increased concentrations of 2,4 decadienal and 4-hydroxynonenal.

Specific to the metabolic response, mice fed oxidized corn oil had greater expression of the PPARα pathway and also an increase of the fatty acid  $\beta$ -oxidation, as well as an inhibition on the purine metabolism pathway. Despite these adaptations, secondary lipid oxidation products, especially aldehydes in the oxidized oil, caused an increase in oxidative stress. An increase in the activity of the aldehyde dehydrogenase in association to the need for detoxification of aldehydes was observed. It is possible that the activation of the two metabolic adaptations affect tryptophan-NAD pathway for NAD<sup>+</sup> production. First, in certain conditions, NAD+ itself can serve as antioxidant, and it could be used to deal with oxidative stress. On the other hand, both aldehyde dehydrogenase and fatty acid  $\beta$ -oxidation use NAD<sup>+</sup> as a cofactor. While the inhibition of purine metabolism could contribute to the NAD+ production needed as cofactor.

### *Peroxidized lipids increase catabolism of tryptophan and threonine in nursery pigs*

In a subsequent study, the observations in nutrient metabolism of mice was evaluated in 128 nursery pigs (Guo et al., 2023). The nursery pigs were fed peroxidized corn oil at incremental levels from 3 to 9% in replacement for

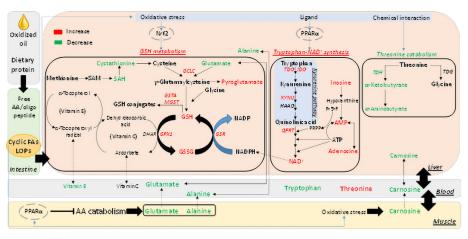


Figure 2. Prominent metabolic response elicited by feeding peroxidized lipids to nursery pigs.

non-oxidized oil. The concentration of serum free alanine, glutamine, and tryptophan after feeding oxidized corn oil to nursery pigs decreased, while the concentration of free serum threonine increased in a dose dependent manner. In addition, hepatic metabolomic analysis indicated the separation in metabolite composition in the liver at a dose dependent manner. Among the metabolite differences were NAD<sup>+</sup>, AMP, glutathione, oxidized glutathione, and pyroglutamic acid. The difference in the concentration of these metabolites was confirmed using internal standards. The concentration of NAD<sup>+</sup> increased in a dose dependent manner with the increased consumption of peroxidized oil. The concentration of NAD<sup>+</sup> was concomitant to the concentration of metabolites in the kynurenine pathway of tryptophan metabolism.

This increase in metabolite concentrations agreed with the increased expression levels of tryptophan dioxygenase (TDO2), indoleamine 2,3-dioxygenase (IDO2), kynureninase (KYNU), and quinolinate phosphoribosyltransferase (QPRT). While serum concentration of tryptophan decreased, the concentration of threonine increased. Threonine is either catabolized by threonine dehydratase (TDH) or by the enzyme threonine dehydrogenase (TDG). Therefore, threonine catabolism was explored by analyzing reactions catalyzed by threonine dehydrogenase (TDG) and threonine dehydratase (TDH). The activity of TDG was not affected given that the concentration of downstream metabolites was also not affected, while the activity of the TDH pathway appears to be inhibited because the concentration of its downstream metabolites was decreased. This was also demonstrated by the co-incubation of liver extracts with hydrophilic extracts of oxidized corn oil supplemented with threonine. The concentration of  $\alpha$ -ketobutyrate in the liver was decreased as the concentration of oxidized oil hydrophilic compounds increased, indicating potential enzyme inhibition. Taken together, this data indicates that nursery pigs consuming diets with adequate levels of vitamin E and Se also engage in transcriptional regulation of amino acid metabolism, increasing the catabolism of tryptophan while sparing threonine (Figure 1).

## *Lipid aldehydes and phenolic antioxidants on protein oxidation in soybean products*

Oxidized lipids are not limited to just heat-treated soybean oils or corn oils, it also occurs in any feed ingredients that have lipids and that are heat treated. These ingredients include soybean products such as solvent extracted soybean meal (SE-SBM), in which residual ether extract concentration is less than 0.5%, and mechanically extracted soybean meal (ME-SBM), in which residual ether extract concentration is 5-8%.

Lysine, glutamate, aspartate, serine, tyrosine, tryptophan, proline, histidine and arginine may crosslink with diverse components of the feed ingredients and form complex structures which may have lower accessibility to digestive enzymes (Meade et al., 2005). It is well described that oxidized proteins decrease the average daily gain and average daily feed intake of 42-day old pigs when fed for about 19 days (Frame et al., 2020). The decrease in growth performance appears to be due to a decrease in amino acid digestibility rather than an increase in oxidative stress. However, there are no studies that have measured lipid peroxidation and protein carbonyl concentration in soybean meal products. Therefore, 54 sources of SE-SBM and 8 sources of ME-SBM were examined using a chemometrics approach to determine the extent of protein oxidation (Zhang et al., 2023). Primarily, protein carbonyl concentration was analyzed by a colorimetric and precipitation test using 2,4 dinitrophenylhydrazone (DNPH). In addition, lipid oxidation was measured by traditional methods such as p-Anisidine, aldehyde concentration characterization and Trolox Equivalent Antioxidant Capacity. The concentration of components in soybean products with potential antioxidant capacity such as tocopherols, total phenolics, and isoflavones, were measured using diverse assays.

Mechanically extracted soybean meal had on average a greater concentration of protein carbonyl than sources of solvent extracted soybean meal. In addition, ME-SBM had a greater concentration of p-Anisidine value and greater concentration of aldehydes (2-heptanal) than sources of SE-SBM. Consequently, Total Antioxidant Capacity as measured by Trolox-equivalence was less in ME-SBM than SE-SBM.

### Conclusions

Increasing sustainability of pork production requires greater precision in evaluation of quality of sources of lipids and heat treatment of feed ingredients to mitigate the risk of pathogen transmission. Thermally treated feed ingredients may contain oxidized lipids which increase catabolism of tryptophan and spare threonine but the impact of the metabolic adaptations on dietary amino acid requirements is unknown. There is variation in oxidized protein carbonyl concentration among protein meals, so we need to establish the impact of oxidation on amino acid digestibility, animal health, and performance.

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# Effects of Fiber and Plane of Nutrition on the Working Boar

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### **Summary**

Boars fed a high soyhull fiber diet had increased body weight, backfat depth and body condition score (BCS) indicating an underestimation of the net energy value of soyhulls in limit fed boars. Boars fed a high soyhull fiber diet had decreased salivary cortisol levels but no differences between treatments were observed in blood glucose 1-2 hours post feeding. Overall, feeding boars a high soyhull fiber diet had no effect on semen characteristics. Restricting feed (1.8 kg/d vs 2.7 kg/d) as soon as a boar is selected at 5-6 months of age has minimal impacts on the trainability of the boar to mount a dummy and successfully collect semen in a stud but may extend their longevity in a stud due to their slower growth. Underfeeding young boars below maintenance has a greater impact on boar body mass and semen production, but older mature boars could have their weight slowly reduced with minimal impacts on semen production over a 9-week period. The NRC (2012) equation used to calculate the maintenance energy requirement of the boar appears to underestimate the modern boar's energy needs and further research is needed.

## Introduction

The swine industry often focuses research on gestation and lactation or nursery and grow-finish production phases, However, the research efforts on the nutrition and management of boars is few and far between for a number of reasons (stud facilities and boar availability, duration of studies, handling boars, etc.). The boar's spermatogenic cycle is roughly 6 weeks so to truly see long term semen impacts experiments should be longer than 6 weeks.

Most boars in the industry are limit-fed with the daily allowance allocated based on bodyweight or body condition (Knox et al., 2008). Boar diets in the US, similarly to other swine diets, are predominantly corn-soybean meal based with other ingredients added to meet nutrient requirements (NRC, 2012). Fiber added to diets of gestating sows and gilts has yielded beneficial results in reproductive performance (Grieshop et al., 2001) and behavior (Robert et al., 2002). Experiments on energy concentrations in the boar diet (Kemp et al., 1989; Boyd et al., 1996) concluded that differing energy levels can impact semen production. This left the question - as the boar continues to genetically change has the nutrition and management of the boar changed as well or are the traditional nutritional targets still meeting the boar's needs? Due to the lack of recent research on the boar on how different feedstuffs and feeding strategies can impact semen production and growth, this impelled us to initiate research with the modern stud boar. The objectives of these current studies were to determine how fiber and different feeding strategies impact semen production and characteristics as well as the growth and development of the artificial insemination (AI) boar.

# **Materials and Methods**

All three experiments reported herein were conducted at Purdue University Swine Research and Educational Center in West Lafayette, Indiana.

## Fiber study

All procedures for this study were approved by the Purdue Animal Care and Use Committee (PACUC #2201002241). The objective of this study was to determine if the inclusion of fiber (8.48% vs 14.85% NDF) in the diet of boars influenced bodyweight, backfat depth, loin depth, Knauer sow caliper measurement, flank to flank distance, visual body condition score, fecal consistency score and volatile fatty acids (VFA),

<b>Table 1.</b> Diet composition evaluating the effect of increasing fiber	
on boar productivity and for the training and feeding level studies	

	Control	Fiber
Corn	75.635	57.825
SBM,47.5% CP	19.370	18.350
Soy hulls	0.000	14.300
Swine grease	1.000	5.650
Limestone	1.320	1.120
MonoCal. Phos.	1.280	1.360
Vitamin and TM premix <sup>1</sup>	0.150	0.150
Choline Chloride, 60%	0.100	0.100
Phytase <sup>2</sup>	0.100	0.100
Salt	0.500	0.500
CarniChrome 50 <sup>3</sup>	0.010	0.010
Availa Sow <sup>4</sup>	0.075	0.075
Clarify <sup>5</sup>	0.210	0.210
Defusion Plus <sup>6</sup>	0.250	0.250
Total	100.000	100.000
Calculated nutrients		
ME, kcal/kg	3285.4	3285.2
NE, kcal/kg	2497.0	2476.3
NDF, %	8.48	14.84
CP, %	15.48	15.18
SID Lys, %	0.65	0.65
Ca, %	0.80	0.80
Avail. Phos., %	0.40	0.40

<sup>1</sup> Provided per kg of diet: vitamin A, 9,010 IU; vitamin D<sub>3</sub>, 2,254 IU; vitamin E, 60 IU; vitamin K, 2.2 mg; riboflavin, 7.1 mg; pantothenic acid, 20 mg; niacin, 40 mg; B<sub>12</sub>, 0.037 mg. biotin, 0.24 mg; folic acid, 1.74 mg; pyridoxine, 4.00 mg; iron, 100 mg; zinc, 120 mg; manganese, 50 mg; copper, 20 mg; iodine, 0.70 mg, and selenium 0.3 ppm.

<sup>2</sup> Phyzyme<sup>®</sup> (Danisco Animal Nutrition, Marlborough, UK) providing 600 phytase units (FTU)/kg.

<sup>3</sup> Carnichrome 50 (Lonza Inc, Fair Lawn, NJ) provided per kg of diet: chromium, 0.20 mg; carnitine 49.6 mg

<sup>4</sup> Availa Sow (Zinpro Corporation, Eden Prairie, MN) is an organic zinc amino acid complex that provides 50.0 ppm Zn, 20 ppm Mn, 10 ppm Cu.

<sup>5</sup> Clarify<sup>®</sup> Larvicide (Central Life Sciences, Schaumburg, IL). <sup>6</sup>Defusion Plus preservatives (Provimi, Lewisburg, OH).

semen characteristics (sperm concentration, motility, and morphology), blood glucose and non-esterified

fatty acids (NEFA), salivary cortisol, and behavior.

Twenty-seven boars (Acuity genetics) from two age groups (7 months and 18 months) and two genetic lines (maternal and terminal) were utilized in this study over a twelve-week period with one week prior to the study serving as baseline for statistical analysis. Boars were blocked by age and breed and randomly allotted to receive 2.72 kg/d of corn-soybean meal diet (CONTROL, n = 13; Table 1) formulated to meet NRC (2012) requirements or an isocaloric (calculated ME = 3285 Kcal/kg) corn-soybean meal diet with 14.3% soyhulls and supplemental choice white grease (4.65%) inclusion (FIBER, n = 14). Boars were fed once daily at 0700 h.

Semen was collected once per week per boar using the gloved hand technique. While at the farm 3 mL of semen was mixed with 27 mL of extender (Androhep Plus, Minitube, Verona, WI) to make a 1:10 dilution. The semen was then assayed for sperm concentration (Nucleocounter SP-100, Chemometec A/S, Denmark), motility (Computer Assisted Sperm Analysis, CASA, CEROS II, IMV technologies, Brooklyn Park, MN), and morphology.

Timepoints for blood, saliva, feces, ultrasound, bodyweight, flank to flank tape measures, Knauer sow caliper measurement, and visual body condition score were weeks -1, 3, 7, and 11. Blood (10 cc) was collected from the lateral or medial auricular ear vein while the boars were mounted for semen collection using a butterfly needle (BD vacutainer) and syringe and placed on ice. Immediately after blood collection, blood glucose levels were analyzed using the Aimstrip Plus glucose reader (Germaine Laboratories, Inc., San Antonio, TX). The blood samples were centrifuged (21,130 ×g / 25 minutes) and serum was stored for future NEFA analysis. Saliva was collected using the Sarstedt Salivette kit (Sarstedt Ag & Co. KG, Germany) to measure salivary cortisol (Salimetrics, State College, PA) approximately 1 hr after feeding. Feces were collected for fecal scoring using the Bristol stool chart (University of Bristol) with a score of 1 being the firmest and 7 being the loosest. Fecal samples were also stored for future analysis of fecal lactoferrin, myeloperoxidase (MPO), and VFA.

On weeks -1,1, 3, 5, 7, 9, and 11 video was collected for a twenty-four-hour period to analyze behavior of the boars. Cameras (KT&C 2.9-12mm Varifocal 750TVL Outdoor IR Day/Night Bullet Security Camera, KT&C, Fairfield, NJ) were hung above and in front of the boars. Videos were analyzed for posture, feed/ water consumption, exploratory, and stereotypic behaviors using continuous sampling.

Data sets were analyzed using PROC MIXED in SAS 9.4 (Cary, North Carolina) using week and phase as repeated analysis. PROC GLM was used to analyze non-repeated measures. Boar served as the experimental unit. Significance was determined at P<0.05 and a trend was observed at  $0.05 < P \le 0.10$ .

### Training Study

All procedures were approved by the Purdue Animal Care and Use Committee (PACUC #082200230). The objective of this study was to determine how different feeding strategies (1.81 kg/d vs 2.72 kg/d; Control diet Table 1) influenced new boars to the stud bodyweight, backfat depth, loin depth, Knauer sow caliper measurement, flank to flank distance, visual body condition score, libido, and duration of semen collection training.

Twenty-five eight-month-old terminal line Duroc boars (Topig Norvsin) were utilized during this study. Boars were randomly allotted upon selection and arrival (two months prior to training, 5-6 months old) to

one of the two feeding levels (2.72 kg/d, n = 13; 1.81 kg/d, n = 12).

After feeding these diet levels for approximately two months, training to a mounting dummy and semen collection was initiated and conducted Monday-Friday for a twoweek period. Time was recorded with a stopwatch (AC-CUSPLIT, Pleasanton, California) for each training period. Boars were given a maximum amount of ten minutes to successfully mount the dummy and begin collection. Boars were scored each training period on a scale from 1 to 5, with 1 being no interest in the dummy to 5 being a successful collection. Boars were not considered fully trained until three consecutive collections occurred. Time points for ultrasound, bodyweight, flank to flank tape measures, Knauer sow caliper measurement, and visual body condition score were collected upon arrival, at the start of the training period, and at the completion of the training period.

### Feeding Strategy Study

All procedures were approved by the Purdue Animal Care and Use Committee (PACUC #082200230). The objective of this study was to determine how different feeding strategies (80%, 100%, 150%) of maintenance requirements of boars influenced bodyweight, backfat depth, loin depth, visual body condition score, Knauer sow caliper measurement, semen characteristics (concentration, motility, and morphology), blood urea nitrogen and creatine.

Twenty-eight boars (Acuity and Topig Norvsin) from two age groups (9 months and 15 months) and two genetic lines (maternal and terminal) were utilized during this study over two nine-week periods with one week prior to each nine-week period serving as a baseline for statistical analysis. Boars were blocked by breed and age and randomly allotted to receive a control corn-soybean meal based diet

<b>Table 2.</b> Effect of supplemental dietary fiber on boar growth
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					P-Value	
	Control	Fiber	SE	Diet	Age	Breed
Body Weight	n=13	n=14				
Wk -1, kg	226.8	230.3	3.80	0.521	<.0001	0.852
Wk 3, kg	228.5	236.2	4.04	0.181	<.0001	0.760
Wk 7, kg	235.4	242.6	4.54	0.268	<.0001	0.787
Wk 11, kg	242.9	257.1	4.13	0.023	<.0001	0.740
Overall Chg BW, kg	16.04	24.45	2.47	0.022	<.0001	0.342
10th Rib Backfat						
Wk -1, mm	12.6	13.7	0.78	0.313	0.862	0.784
Wk 3, mm	12.5	14.3	0.72	0.095	0.557	0.359
Wk 7, mm	14.4	16.6	1.13	0.173	0.400	0.494
Wk 11, mm	14.1	18.2	0.95	0.006	0.209	0.099
Overall Chg BF, mm	1.5	4.4	0.66	0.005	0.132	0.012

(Table 1) to meet NRC (2012) requirements at: 1) HIGH (150% of maintenance, n = 9); 2) MAIN (100% of maintenance, n = 10); or 3) LOW (80% of maintenance, n = 9) feed intakes (FI) for the first repetition of this study. The NRC (2012) maintenance equation used in this study was ME for maintenance =  $(100*kg BW ^0.75) + 15$  kcal for sperm production). After this first period the remaining 24 boars were utilized for a second nine-week period with one-week prior serving as baseline for statistical analysis. Boars from the first repetition that were fed the HIGH or LOW FI were crossed over to the opposite treatment, while the boars on maintenance continued on maintenance for the second 9-week period (LOW-HIGH, 150% maintenance, n = 7), (MAIN-MAIN, 100% maintenance, n = 8), (HIGH-LOW, 80% maintenance, n = 9).

Semen was collected once per week per boar using similar procedures as in the Fiber study above. The semen samples were analyzed for concentration (Nucleocounter SP-100, Chemometec A/S, Denmark), motility (Computer Assisted Sperm Analysis, CASA, CEROS II, IMV technologies, Brooklyn Park, MN), and sperm morphology. Blood samples (from an ear vein) were collected in the first period on week 9 and in the second period on weeks -1, 5 and 9 for NEFA, BUN, and testosterone assays. Saliva was only collected during the second period on weeks -1, 3, 6, and 9 for analysis of cortisol. Timepoints for ultrasound, bodyweight, flank to flank tape measures, Knauer sow caliper measurement, and visual body condition score were collected on weeks -1, 3, 6, and 9 for both periods.

## Results

### Fiber Study

FIBER boars gained 8.4 kg more than CON boars during the 12-week study (P=0.022; Table 2). On weeks 3 and 11,

Table 3. Effect of supplemental dietary fiber on semen characteristics

					P-Value	
	Control	Fiber	SE	Diet	Age	Breed
	n=13	n=14				
Total sperm, x 10 <sup>9</sup>	80.98	79.17	5.28	0.804	0.190	0.202
Total normal sperm x 10 <sup>9</sup>	63.79	58.73	3.82	0.347	0.835	0.715
Volume, mL	174.4	157.1	14.1	0.384	0.556	0.050
Motility, %	85.04	82.83	1.18	0.198	0.688	0.514
Progressive motility, %	74.42	72.81	1.74	0.517	0.797	0.801
Normal, %	79.3	75.4	1.8	0.123	0.014	0.152
Normal acrosome, %	88.15	87.49	0.54	0.392	0.111	0.109
Head/Tail <sup>1</sup> , %	2.0	2.2	0.2	0.625	0.025	0.080
DMR <sup>2</sup> , %	3.8	4.2	0.6	0.581	0.461	0.095
Distal droplets, %	8.1	8.7	0.9	0.604	0.051	0.148
Proximal droplets, %	7.6	9.2	1.0	0.220	0.184	0.032

<sup>1</sup>Head/Tail = Head abnormalities or tail defects

<sup>2</sup>DMR = Distal Midpiece Reflex

**Table 4.** Effect of feeding below (LOW = 80%), at (MAIN = 100%), or above Maintenance (HIGH = 150%) for 9 weeks (Period 1) on semen characteristics

Age of Boars:		Old			Young				P-Value	
Feed Intake:	HIGH n=2	MAIN n=3	LOW n=2	HIGH n=7	MAIN n=7	LOW n=7	SE	FI	AGE	FIxAGE
Total Sperm, x10 <sup>9, a</sup>	103.4	80.99	84.92	78.25	81.26	63.64	17.57	0.170	0.237	0.422
Total Normal Sperm, x10 <sup>9, b</sup>	73.30	65.15	64.26	68.78	68.31	57.02	16.37	0.447	0.811	0.873
Volume, mL <sup>c</sup>	133	153	166	135	131	113	13.1	0.629	0.022	0.015
Motility %	72.2	73.4	69.1	78.1	71.1	66.0	7.35	0.444	0.973	0.683
Progressive Motility %	57.4	58.0	56.4	68.8	59.0	52.9	6.78	0.338	0.478	0.362
Normal %	80.7	79.4	87.8	86.7	80.8	82.1	3.99	0.254	0.829	0.198
Normal Acrosome %	90.6	90.3	91.0	90.2	90.5	85.3	4.48	0.759	0.482	0.637
Proximal Droplet %	4.5	5.6	3.4	3.3	4.7	3.3	1.60	0.330	0.469	0.903
Distal Droplet %	8.5	8.4	5.8	6.7	6.6	6.5	2.44	0.705	0.539	0.765
Distal Midpiece Reflex %	3.4	3.2	1.3	2.5	3.5	3.4	1.08	0.460	0.456	0.217
Head Tail Abnormality %	3.4	2.4	2.4	3.0	3.7	6.3	3.68	0.456	0.488	0.767

<sup>a</sup> FI x Wk x Age P=0.0003

<sup>b</sup> Fl x Wk x Age P=0.0005

<sup>c</sup> Fl x Wk x Age P=0.0068

**Table 5.** Effect of feeding below (L=80%), at (M=100%), or above Maintenance (H=150%) for second 9 weeks (Period 2) during a crossover of L and H treatments for a subsequent 9 week period

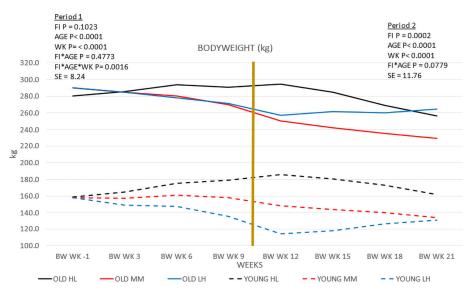
Age of Boars:		Old			Young				P-Value	
Feed Intake:	LOW- HIGH n=2	MAIN- MAIN n=3	HIGH- LOW n=2	LOW- HIGH n=5	MAIN- MAIN n=5	HIGH- LOW n=7	SE	FI	AGE	FI*AGE
Total Sperm, x10 <sup>9</sup>	82.57	58.15	65.98	50.43	51.45	55.10	10.09	0.273	0.017	0.313
Total Normal Sperm, x10 <sup>9</sup>	71.26	46.19	49.8	43.64	42.52	47.46	9.82	0.182	0.074	0.301
Volume, mL <sup>a</sup>	109	91	73	91	79	74	11.4	0.061	0.276	0.400
Motility %	87.7	88.4	86.3	91.5	86.0	88.2	3.17	0.574	0.588	0.405
Progressive Motility %	79.1	77.4	75.3	82.7	77.4	79.2	2.94	0.262	0.188	0.548
Normal % <sup>b</sup>	82.5	83.3	76.7	88.7	80.5	85.3	2.60	0.096	0.020	0.011
Normal Acrosome %	99.6	91.8	94.3	90.5	91.8	92.6	2.51	0.306	0.021	0.057
Proximal Droplet %	5.8	4.2	6.3	5.0	6.2	5.1	2.51	0.954	0.987	0.656
Distal Droplet %	6.3	6.5	7.6	4.2	5.4	5.2	2.32	0.834	0.219	0.919
Distal Midpiece Reflex %	3.3	4.4	7.7	0.1	5.6	3.3	1.82	0.044	0.079	0.086
Head Tail Abnormality %	2.1	2.1	1.0	3.3	2.2	1.4	0.83	0.097	0.290	0.684

<sup>a</sup>FI x Wk P=0.0001

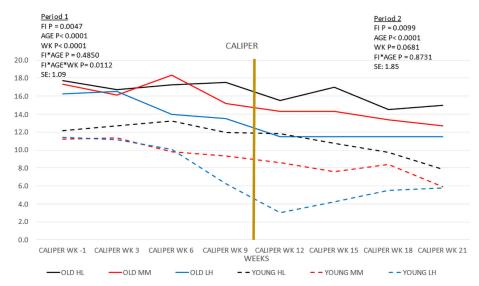
<sup>b</sup>Fl x Wk x Age P=0.0349



Figure 1. Effect of feed intake at time of selection and stud entry on boar body weight.



**Figure 2.** Effect of feeding below (L=80%), at (M=100%), or above Maintenance (H=150%) for 9 weeks (Period 1) followed by a crossover of L and H treatments for a subsequent second 9 weeks (Period 2) on boar body weight.



**Figure 3.** Effect of feeding below (L=80%), at (M=100%), or above Maintenance (H=150%) for 9 weeks (Period 1) followed by a crossover of L and H treatments for a subsequent second 9 weeks (Period 2) on boar body Knauer caliper measurement.

FIBER boars tended to have increased backfat depth compared to CON boars (P<0.095), resulting in FIBER boars gaining 2.9 mm more backfat compared to CON (P=0.005) over the 12-week study. No difference in loin muscle depth was observed between treatments. On weeks 7 (3.7 vs 3.4, P=0.092) and 11 (3.9 vs 3.5, P=0.061) FIBER boars tended to have higher BCS score compared to CON boars. FIBER boars tended to have larger caliper scores on week 3 (18.7 vs 17.5 P= 0.077) and week 7 (18.6 vs 17.6 P= 0.056) than CON boars. FIBER boars on week 11 did have larger caliper scores than CON boars (19.4 vs 17.6, P=0.003), but no difference in overall change was observed for flank-toflank tape or caliper measurements. FIBER boars tended to have softer stools during week 3 of the trial compared to CON boars (P=0.053), but the fecal scores were not different at any other week. FIBER boars had lower salivary cortisol levels (P=0.003) than CON boars. However, there was no difference in blood glucose concentration between treatments.

Fiber in the diet did not affect any semen characteristics throughout the study (Table 3). However, the percent morphologically normal sperm was higher in young boars compared to the older boars (80.6% vs 74.1%, P=0.014) resulting from fewer head/tail abnormalities (1.8% vs 2.4%, P=0.025) and distal droplets (7.2% vs 9.6%, P=0.051). Maternal boars had fewer proximal droplets (6.9% vs 9.9%, P= 0.032) and tended to have fewer head/tail abnormalities (1.8% vs 2.3%, P=0.080). Terminal boars tended to have fewer distal midpiece reflex abnormalities than maternal boars (3.3% vs 4.8%, P=0.095). Maternal boars tended to have higher volume (186 mL vs 146 mL, P=0.050). However, the overall sperm (84.87\*109 vs 75.28\*109) and normal sperm (62.23\*109 vs 60.29\*109) count was not significant (P > 0.10) between maternal and terminal boars.

### Training Study

Feeding boars 33% less feed from the time of selection at 5-6 months of age until training in the boar stud decreased the boar BW (Figure 1) by 6.1 kg at week 9 (P=0.034) and 13.8 kg at week 12 (P=0.0001). Training to collect from a dummy during week 10-12, the low FI treatment numerically reduced the percent of boars collecting early in the training period (day 2 of training, 50% vs 77%; P = 0.16). However, by the end of the 2-week training period both feeding levels had approximately 92% of the boars trained to collect from a dummy.

## Feeding Strategy Study

During the first 9 week period there was a 3-way interaction between the FI level, boar age, and week on boar BW (Figure 2; P=0.0016). The young boars on the LOW FI lost more weight and young boars on the HIGH FI gained more weight than the older boars fed similar treatments. Similar shifts in body caliper measurements (Figure 3) were observed in period 1 (FI\*AGE\*WK; P=0.0112) due to the young boars having a greater change in caliper measures when fed the LOW diet, especially during the last couple weeks of period 1. These changes in BW also resulted in 3-way interactions for semen volume, total sperm, and total normal sperm (Table 4) during period 1, presented as young boars fed LOW FI having semen reductions sooner and at greater amounts than old boars fed the LOW FI (data not shown). During Period 2, boars that switched to the LOW FI treatment from the HIGH FI lost weight (Figure 2) while those boars now fed the HIGH FI treatment gained weight (P=0.0002) and body caliper measurements responded in a similar fashion (Figure 3; P=0.0099). During Period 2, semen volume tended to be reduced by Low FI (Table 5; P=0.061) with increased distal midpiece reflex abnormalities (P=0.044) but tended to have less head or tail abnormalities (P=0.097). There was an interaction with diet and boar age for percent normal sperm (P=0.011), with LOW FI in older boars decreasing the percent normal sperm while in the young boars those fed MAIN had the lowest percent normal sperm.

Future research is needed with varying levels and types of fiber inclusion in the diet along with differing ages and breeds of boars to understand how to correctly implement fiber into the limit-fed boar diet. In addition, future research is needed to refine the net energy values of fibrous feedstuffs like soyhulls for limit-fed boars to better understand how fiber impacts semen characteristics and production, growth, and development. Starting to restrict feed as soon as selection is made for a boar only marginally impacts the trainability of the boar to mount a dummy and semen collect in a stud but may help to reduce the boars size and extend their longevity in a stud. Under-feeding young boars below maintenance has a greater impact on boar body mass and semen production, but older mature boars could have weight slowly reduced with minimal impacts on semen production. The NRC (2012) equation used to calculate the maintenance energy requirement of the boar appears to underestimate the modern boars energy needs and further research is needed to correctly maintain a modern boar's body weight.

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