THE EFFECT OF LYSINE LEVEL OR METHIONINE/COPPER/MANGANESE ON OSTEOCHONDROSIS LESIONS AND CARTILAGE PROPERTIES IN PIGS

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Summary

A total of 120 gilts (PIC 327 × 1050; 89.2 lb initial BW) were used in a 3 × 2 factorial, 84-d study to determine the effect of lysine (Lys) fed either below the calculated requirement (0.8% true ileal digestible (TID) Lys Phase I and 0.6% TID Lys Phase II), at requirement (1.0% TID Lys Phase I and 0.8% TID Lys Phase II), or above the requirement (1.3% TID Lys Phase I and 1.1% TID Lys Phase II) with standard concentrations or with high added methionine (Met, 1 %), copper sulfate (Cu, 250 ppm), and manganese sulfate (Mn, 220 ppm) on the occurrence and severity of osteochondrosis (OC) lesions, growth performance, soundness, carcass traits, and several cartilage criteria. Upon completion of the feeding period, pigs were harvested and the distal aspect of the left humerus and femur were evaluated by gross examination for OC lesions. The external surface was evaluated for abnormalities and received a severity score. For the external femur evaluation, increasing dietary Lys concentration tended (linear, *P*<0.08) to increase the number of abnormalities and there was a numerical trend for an increased severity score (*P*<0.13) with increasing dietary Lys. The addition of high Met/Cu/Mn to the diet reduced the number of abnormalities (*P*<0.02) and severity score (*P*<0.01) at the external femur compared to pigs fed diets with standard concentrations of Met/Cu/Mn. At the external humerus, increasing dietary Lys increased both the number of abnormalities (linear, *P*<0.01) and severity score (linear, *P*<0.01). The addition of high Met/Cu/Mn to the diet reduced the number of abnormalities (*P*<0.03) and severity score (*P*<0.03) for the external humerus. Increasing dietary Lys concentration or high-added Met/Cu/Mn had no effect (*P*>0.14) on the number of faces with lesions at the femoral growth plate or the severity score (*P*>0.19). The number of faces with lesions and severity score at the humerus articular cartilage was unaffected by increasing dietary Lys concentration (*P*>0.16) or the addition of high Met/Cu/Mn to the diet (*P*>0.37). The total faces with lesions were not impacted by increasing dietary Lys concentration (*P*>0.78) or additional high Met/Cu/Mn (*P*>0.86). The total abnormalities (external and number of faces) tended to increase with increasing dietary Lys (linear, *P*<0.12). The addition of high Met/Cu/Mn did not affect the total number of abnormalities (*P*>0.16). The total severity score for both external and OC evaluation increased with increasing dietary Lys concentration (linear, *P*<0.01). The addition of high Met/Cu/Mn decreased the total severity score (*P*<0.02) compared to pigs fed diets with standard concentrations of Met/Cu/Mn. Finally, increasing dietary Lys concentration

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increased the sum (linear, $P<0.05$) of abnormalities and total severity score. The addition of high Met/Cu/Mn tended ($P<0.09$) to reduce the overall severity score compared to pigs fed diets with standard concentrations of Met/Cu/Mn. In conclusion, feeding growing gilts dietary Lys to maximize growth performance may increase the severity of OC lesions, while a diet with additional Met/Cu/Mn may aid in the reduction of OC severity scores.

(Key words: amino acids, cartilage, health, osteochondrosis.)

Introduction

Previously, we have attempted to determine the role of nutritional ingredients related to cartilage and bone metabolism on the occurrence of osteochondrosis (OC). Osteochondrosis is the focalized disruption in the endochondral ossification of cartilage at the end of growing long bones leaving areas of retained cartilage in the subchondral bone. Our first experiment revealed several minerals and amino acids that showed positive results on reducing the severity of OC. In our second experiment, we attempted to further investigate the effect of minerals and amino acids but were unable to replicate the results of the first experiment. In our second experiment, pigs were fed in one phase, whereas the first experiment was conducted with a three-phase feeding program to allow optimum performance and to closely match amino acid requirements. In addition, the occurrence of OC was lower in the second experiment, and our ability to influence OC may have been compromised due to pigs starting at a heavier weight, being slaughtered at a lighter weight, the shorter duration of the trial, and the fact that pigs were fed slightly under their requirement for Lys early in the study and slightly more than their requirement for Lys late in the study. The occurrence of OC is thought to be bilateral or occur in multiple joints, thus we also collected the left humerus as well as the femur to improve our detection of OC. Previously, it has been thought that pigs with the fastest growth rate may have an increased occurrence of OC; however, the available data suggests conflicting results.

The objective of this experiment was to determine the effect of lysine level and the combination of additional methionine, copper sulfate, and manganese sulfate on growth performance, OC lesions, and several cartilage criteria in grow-finish pigs.

Materials and Methods

General. Procedures used in these experiments were approved by the Kansas State University Animal Care and Use Committee. The experiment was conducted at the Kansas State University Swine Research and Teaching Center. Each pen contained two pigs per pen and there were 10 replicates (pens) per treatment. The barn contains 80 pens with totally slatted concrete flooring (5 × 5 ft), providing approximately 12.5 sq ft/pig. Each pen was equipped with a one-hole dry self-feeder (Farmweld, Tuetopolis, IL) and nipple waterer to allow *ad libitum* access to feed and water.

A total of 120 gilts (PIC line 327 × 1050; 89.2 lb initial BW) were blocked by weight in an 84-d growth assay. Dietary treatments were arranged in a 3 × 2 factorial consisting of three TID Lys levels fed below (0.8% Phase I, 0.6% Phase II), at the requirement (1.0% Phase I, 0.8% Phase II), or above their requirement (1.3% Phase I, 1.1% Phase II). The Met/Cu/Mn treatments were either at standard inclusions typical of swine diets (no added methionine, 9 ppm of Cu and 20 ppm of Mn) or high-added methionine (1% added DL-methionine), 250 ppm Cu (Copper sulfate), and 220 ppm Mn (Manganese sulfate). The addition of Met/Cu/Mn replaced sand to form the other dietary treatments. The dietary Lys requirement was determined from previous experiments conducted in this facility. Experimental diets were fed in meal form for 84 d in two 42 d phases. The values used in diet formulation and TID digestibilities were based on those published in the NRC (1998). Diet samples were analyzed for amino acid content and contained levels similar to calculated concentrations.

Growth Performance and Carcass Composition. Pigs and feeders were weighed every 14 d to determine ADG, ADFI, and F/G. At the end of the trial, pigs were weighed before transport to the Kansas State University Meats Laboratory, where the left front (elbow joint) and hind leg (knee joint) were collected for determination of OC lesions for one pig in each pen. Before transport, the heaviest pig from each pen was marked with a distinctive tattoo to allow data to be recorded for each pig. Pigs were loaded onto a trailer in small groups (15 pigs) and transported approximately 2 miles to the processing facility. For carcass data, 10^{th} rib backfat depth, longissimus muscle area (LMA), percentage lean, and hot carcass weight were evaluated. Fat depth was measured with a ruler at the 10^{th} rib, 2.4 inches off of the midline, while LMA was traced on translucent paper and calculated using a grid. Percentage lean was calculated using an equation from the National Pork Producers Council (NPPC).

Visual Soundness Scores. Prior to slaughter, the heaviest pig from each pen was scored by two evaluators for the front leg and rear leg (1-5 where 1=poor and 5=excellent) based on angle and conformation, and for locomotion (1-5 where 1=poor and 5=excellent) as an indication of mobility. The front and rear legs scores were added together to form the total score according to the National Swine Improvement Federation (NSIF) system (where 1-3=poor or unsuitable for breeding purposes, 4-7=average, and 8-10=excellent or desirable for breeding purposes).

Collection of Cartilage Data and OC Lesions Scores. The left front leg (elbow joint) and hind leg (knee joint) were collected and removed to visually determine the number of cartilage abnormalities and the occurrence of OC lesions by gross examination of the humerus and femoral condyles for one pig from each pen. The joints were cleaned of excess tissue and then stored in 10% formalin until evaluation. After external evaluation, the distal end of the humerus and femur were sliced into 3 mm thick sections by cutting perpendicular to the long axis of the bone using a bandsaw. Each joint was evaluated for the number of external abnormalities at femoral and humerus condyles, OC lesions at the articular cartilage and growth plate cartilage of the distal femur, humerus articular cartilage, and given a severity score (0-4) for all locations, where 0=normal, 1=mild, 2=moderate, 3=severe, and 4= OC dissecans based on the extent of tissue involvement. All pigs had OC lesions at one of the locations evaluated, so we were unable to analyze for differences in OC occurrence (number of animals with OC).

In addition, a cartilage sample was cut from the patella for cartilage property analysis. Cartilage samples were weighed, measured for thickness and length using a caliper, and then tested for the ability to absorb compression or to resist shearing using an Instron testing machine. Cartilage samples were placed between two flat surfaces of the Instron to perform texture profile analysis and compressed half of the thickness to measure the ability of the cartilage to resist compression force. A second measure was conducted in which the cartilage was cut using a Warner-Bratzler shear blade to determine the ability of the cartilage to withstand shearing force. Compression values and shear values were adjusted to a per gram of cartilage weight to equalize for differences in the actual cartilage sample weight.

Relationship between Growth Rate, Weight, Visual Soundness, and Overall Severity Score. Because there were differences in growth rate among our dietary treatments, a correlation between growth rate or weight and the overall severity score was conducted.

Visual evaluation of soundness or leg conformation was also evaluated for correlation with the overall severity score. Each prediction variable was plotted by the overall severity score and a linear regression line fitted to determine how much of the variation in overall severity score could be explained by the variables (r^2 value).

Statistical Analysis. Data were analyzed as a randomized complete block design using the PROC MIXED procedure of SAS with pig as the experimental unit to determine the main effect of treatment. The response criteria of growth performance, carcass composition, cartilage compression and shear energy, and number of abnormalities were tested. Although scored categorically, soundness and OC severity scores were analyzed via PROC MIXED because low numbers of observations at some of the severity scores prevented categorical analysis. Linear and quadratic effects of increasing dietary Lys were determined using single degree of freedom contrasts.

Results and Discussion

Growth. Overall (d 0 to 84), a Lys × Met/Cu/Mn interaction was detected for ADG (*P*<0.02, Table 2), thus the interactive means are presented. Increasing dietary Lys concentration improved ADG up to the requirement (quadratic, *P*<0.01). Addition of high Met/Cu/Mn to the diet reduced ADG (*P*<0.01) compared to diets with standard concentrations of Met/Cu/Mn, particularly in the diets with Lys fed below or at the requirement. Increasing dietary Lys tended (linear, *P*<0.09) to increase ADFI, while high-added Met/Cu/Mn reduced ADFI (*P*<0.01) compared to pigs fed diets with standard concentrations of Met/Cu/Mn. Increasing dietary Lys up to the requirement improved F/G (quadratic, *P*<0.01), but feeding diets containing high added Met/Cu/Mn had similar F/G (*P*>0.57) compared to pigs fed diets with standard concentrations of Met/Cu/Mn.

Carcass Data. No interaction between Lys × Met/Cu/Mn was detected for carcass traits (*P*>0.49, Table 3). There was a trend (linear, *P*<0.15) for increasing dietary Lys concentration to reduce backfat thickness, while high added Met/Cu/Mn tended (*P*<0.07) to reduce backfat thickness. Increasing dietary Lys up to the requirement increased loin eye area (quadratic, *P*<0.04), but the addition of high Met/Cu/Mn did not affect loin eye size (*P*>0.61). Increasing dietary Lys improved percentage lean (linear, *P*<0.01), while high-added Met/Cu/Mn had no affect on percentage lean (*P*> 0.14).

Leg Scoring. A Lys × Met/Cu/Mn interaction was not detected for leg scores (*P*>0.21, Table 4). Visual soundness scores were unaffected by dietary Lys level (*P*>0.26); however, the addition of high Met/Cu/Mn to the diet tended (*P*<0.07) to reduce front leg scores and locomotion (*P*<0.06). Previous research suggests that high levels of manganese may cause bones to become stiff and may decrease mobility.

Cartilage Properties. There tended to be an interaction for both cartilage weight and thickness between Lys × Met/Cu/Mn (*P*<0.06, Table 5); however, an interaction for instron measurements was not detected (*P*>0.17) and thus the main effects are presented. Both cartilage sample weight (quadratic, *P*<0.08) and length (quadratic, *P*<0.06) tended to increase with increasing dietary Lys level, while cartilage thickness was unaffected by dietary Lys level (*P*>0.27). Cartilage sample weight, thickness, and length were unaffected (*P*>0.11) by addition of high Met/Cu/Mn to the diet. Increasing dietary Lys decreased cartilage shear energy (quadratic, *P*<0.01); however, no other instron measurement was affected by Lys level (*P*>0.24). High-added Met/Cu/Mn had no effect on any cartilage instron measurement (*P*>0.23). We do not have a reason why pigs fed Lys at their requirement had decreased shear energy values compared with pigs fed below or above their requirement. This may be due to alterations in cartilage metabolism when amino acids are limiting or provided in excess.

Osteochondrosis Evaluation. No Lys × Met/Cu/Mn interactions were detected for OC measures (*P*>0.12, Table 6). All animals had gross OC at either the humerus or femur. For the external femur evaluation, increasing dietary Lys concentration tended (linear, *P*<0.08) to increase the number of abnormalities. There was a trend (*P*<0.13) for increasing dietary Lys to increase the severity score for the external femur. The addition of high Met/Cu/Mn to the diet reduced the number of abnormalities (*P*<0.02) and severity score (*P*<0.01) at the external femur compared to pigs fed diets with standard concentrations of Met/Cu/Mn. At the external humerus, increasing dietary Lys increased both the number of abnormalities (linear, *P*<0.01) and severity score (linear, *P*<0.01). The addition of high Met/Cu/Mn to the diet reduced the number of abnormalities (*P*<0.03) and severity score (*P*<0.03) for the external humerus. The increase in external abnormalities seen at both the femur and humerus with increasing dietary Lys concentration may be due to greater muscle mass providing additional stress on the joint. Faster growing pigs have been theorized to be more susceptible to mechanical stressors due to their increased weight relative to the maturity of the joint. Furthermore, the reduction in external abnormalities that were seen with the addition of high Met/Cu/Mn is similar to results with previous experiments and may be due to the positive influence of Met on cartilage metabolism and Cu/Mn in stabilizing the extracellular matrix through preventing excessive degradation. This combination of ingredients may allow articular cartilage a greater ability to repair damage caused by mechanical stressors.

At the femoral articular cartilage, increasing dietary Lys did not impact either the number of faces with lesions (*P*>0.35) or the severity score (*P*>0.36). The addition of high Met/Cu/Mn to the diet did not influence the number of faces with lesions (*P*>0.57) or the severity score (*P*>0.89) at the femoral articular cartilage compared to pigs fed diets without high Met/Cu/Mn. Increasing dietary Lys concentration had no affect (*P*>0.55) on the number of faces with lesions at the femoral growth plate or the severity score (*P*>0.52). The addition of high Met/Cu/Mn to the diet did not affect either the number of faces with lesions (*P*>0.14) at the femoral growth plate or the severity score (P > 0.19) compared to pigs fed diets without additional Met/Cu/Mn.

The number of faces with lesions and severity score at the humerus articular cartilage was unaffected by increasing dietary Lys concentration (*P*>0.16) or the addition of high Met/Cu/Mn to the diet (*P*>0.37).

Overall, the total faces with lesions were not impacted by increasing dietary Lys concentration (*P*>0.78) or additional high Met/Cu/Mn (*P*>0.86). The total abnormalities (external abnormalities and the number of faces with lesions) tended (linear, *P*<0.12) to increase with increasing dietary Lys. The addition of high Met/Cu/Mn did not affect the total number of abnormalities (*P*>0.16). The total severity score (external and OC evaluation) increased with increasing dietary Lys concentration (linear, *P*<0.01). The addition of high Met/Cu/Mn decreased the total severity score (*P*<0.02) compared to pigs fed diets without Met/Cu/Mn. Finally, increasing dietary Lys concentration did not affect the overall severity score (*P*>0.64). The addition of high Met/Cu/Mn tended (*P*<0.09) to reduce the overall severity score compared with pigs fed diets without high Met/Cu/Mn.

Correlation of Growth Rate and Visual Evaluation with Overall Severity Score. Because of the difference in growth rates between dietary treatments and increasing severity found in pigs fed increasing dietary Lys concentrations, we plotted ADG, weight, total leg score, or locomotion score versus the overall severity score for each pig. Fitting a linear regression line to the data resulted in almost no correlation with ADG (R^2 0.0316, Chart 1), weight (R^2 0.0262, Chart 2), Total leg score (R^2 0.0153, Chart 3), or Locomotion score (R^2 0.0197, Chart 4). This suggests that even though increasing dietary Lys increased severity scores, this increase can not only be attributed to increasing growth rate. As well, visually evaluating structural correctness as a function of front and rear leg scores or locomotion scores also did not correlate with the overall severity score.

In summary, increasing dietary Lys concentration up to the requirement improved ADG and F/G, confirming our estimation of the requirement. Increasing dietary Lys resulted in a decrease in backfat, an increase in loin eye area, and thus pigs with greater percentage lean. The addition of high Met/Cu/Mn to the diet reduced ADG, ADFI, and backfat depth. These results were expected as high dietary Met is know to dramatically reduce feed intake and limit growth performance. The addition of high Met/Cu/Mn also tended to have adverse affects on visual front leg and mobility scores. This may have been mainly due to the high level of Mn fed in these diets as excessive Mn is known to result in stiffer bones and reductions in mobility. Increasing dietary Lys had negative effects on external abnormalities and the total severity score; however, the additional growth achieved in pigs fed increasing dietary lysine did not correlate with the overall severity score. In a previous study, we reported negative effects of high dietary arginine and glycine on OC. Arginine is the precursor of nitric oxide, one of the mediators of the inflammatory response and may have negative implications on joint health. This suggests that it is not simply growth rate that influences OC severity but rather may be the result of increased dietary protein supplying additional non-essential amino acids that may have negative effects on cartilage metabolism. The addition of Met/Cu/Mn at high levels resulted in a reduction in the number of external abnormalities and overall severity score. This is similar to our first study and is probably a result of the positive influence Met has on increased cartilage metabolism, the role of Cu in crosslinking collagen molecules, and Mn in proteoglycans within the extracellular matrix. These dietary ingredients potentially could enhance the ability of the cartilage to repair damaged tissue or prevent excessive degradation of the extracellular matrix as the loss of proteoglycan content and decreased collagen type II content is seen in cartilage with OC.

In conclusion, increasing dietary Lys concentration resulted in an increase in both external abnormalities and total severity score but could not be attributed to ADG, while the addition of high Met/Cu/Mn reduced external abnormalities at the femur and humerus and overall severity score. This study suggests that the addition of high Met/Cu/Mn to the diets of growing gilts may aid in the reduction of defects in the cartilage surface and thus provide a mechanism to limit reductions in sow herd longevity due to OC.

Table 1. Diet Composition (as-fed)

	Phase I[a]			Phase II[b]		
Item	Below	Requirement	Above	Below	Requirement	Above
Ingredient						
Corn	76.20	69.05	58.25	83.90	76.75	65.95
Soybean meal (46.5% CP)	16.05	23.25	34.15	8.75	16.00	26.85
Soy oil	3.00	3.00	3.00	3.00	3.00	3.00
Monocalcium P (21% P)	1.85	1.8	1.75	1.50	1.45	1.40
Limestone	1.03	0.98	0.9	1.03	0.98	0.90
Salt	0.35	0.35	0.35	0.35	0.35	0.35
Vitamin premix	0.15	0.15	0.15	0.13	0.13	0.13
Trace mineral premix	0.08	0.08	0.08	0.08	0.08	0.08
L-Lysine HCl	0.15	0.15	0.15	0.15	0.15	0.15
DL-Methionine	-	0.02	0.09	-	-	0.04
L-Threonine	0.02	0.04	0.07	-	0.03	0.05
Manganese sulfate	-	-	-	-	-	-
Copper sulfate	-	-	-	-	-	-
Sand[c]	1.15	1.15	1.15	1.15	1.15	1.15
Calculated analysis						
Total lysine, %	0.80	1.00	1.30	0.60	0.80	1.10
True ileal digestible amino acids						
Lysine, %	0.71	0.89	1.16	0.53	0.71	0.98
Isoleucine:lysine ratio, %	70	70	69	71	70	69
Leucine:lysine ratio, %	168	153	140	192	168	149
Methionine:lysine ratio, %	30	30	32	34	30	30
Met & Cys:lysine ratio, %	63	60	60	71	63	60
Threonine:lysine ratio, %	67	67	67	68	68	67
Tryptophan:lysine ratio, %	19	19	20	18	19	19
Valine:lysine ratio, %	68	66	64	88	83	79
ME, kcal/lb	1546	1546	1546	1553	1553	1553
CP, %	13.9	16.7	20.8	11.2	14.0	18.1
Ca, %	0.81	0.80	0.80	0.71	0.71	0.70
P, %	0.71	0.73	0.77	0.61	0.63	0.66
Available P equiv.	0.52	0.52	0.52	0.43	0.43	0.43
Lysine:calorie ratio, g/mcal	2.3	2.9	3.8	1.7	2.3	3.2

[a]Phase I fed in meal form from d 0 to 42.

[b]Phase II fed in meal form from d 42 to 84.

[c]Methionine added at 1%, manganese sulfate at 0.05%, and copper sulfate at 0.1% replaced sand to form the other dietary treatments.

Table 2. Effect of Dietary Lysine and Methionine/Copper/Manganese on Growth Performance[abc]

	TID Lysine[c]			Met/Cu/Mn[d] TID Lysine[c]				Probability, *P* <				
									Lysine			
Item	Below	Requirement	Above	Below	Requirement	Above	SED	Trt	Linear	Quadratic	Met/ Cu/Mn	Lys × Met/Cu/Mn
D 0 to 84												
Initial weight, lb	89.0	89.3	89.5	88.8	89.2	89.3	1.10	0.80	0.18	0.76	0.55	0.99
ADG, lb	1.97[f]	2.22[g]	2.19[g]	1.68[e]	2.04[f]	2.17[g]	0.060	0.01	0.01	0.01	0.01	0.02
ADFI, lb	5.67[g]	5.65[g]	5.65[g]	5.06[e]	5.19[ef]	5.50[fg]	0.169	0.01	0.09	0.64	0.01	0.12
F/G	2.89[e]	2.55[f]	2.58[f]	3.02[e]	2.54[f]	2.54[f]	0.077	0.01	0.01	0.01	0.57	0.68
Final weight, lb	263.1[f]	285.8[g]	283.0[g]	237.4[e]	269.9[f]	283.8[g]	5.57	0.01	0.01	0.01	0.01	0.06

[a]Each value is the mean of 10 replications with two pigs per pen initially 89.2 lb and an average final weight of 280 lb.
[b]Pigs were fed meal diets in two 42 d phases.
[c]Diets contained 0.71, 0.89, and 1.16 % TID lysine during Phase I and 0.53, 0.71, and 0.98 TID lysine during Phase II, respectively.
[d]Diet contained additional methionine (1%), copper (250 ppm), and manganese (220 ppm).
[e,f,g]Means with different superscripts differ by P < 0.05.

Table 3. Main Effect of Lysine Level and Additional Methionine/Copper/Manganese on Carcass Characteristics[a]

	TID Lysine[b]			Met/Cu/Mn[c]			Probability, *P*<				
							Lysine				
Item	Low	Requirement	High	Standard	Added	SED	Lys	Linear	Quadratic	Met/ Cu/Mn	Lys × Met/Cu/Mn
Final wt, lb[d]	261.6	286.9	291.9	286.4	273.9	4.64	0.03	0.01	0.02	0.10	0.81
HCW, lb[e]	178.4	199.6	201.3	197.8	188.4	---	---	---	---	---	---
Backfat, in	0.61	0.57	0.54	0.61	0.54	0.053	0.33	0.15	0.99	0.07	0.80
Loin eye area, in^2	7.48	9.59	10.13	8.98	9.16	0.494	0.01	0.01	0.04	0.61	0.49
% lean	56	59	61	58	59	0.11	0.01	0.01	0.14	0.14	0.92

[a]Each mean represents 10 replications with one pig from each pen initially 89.2 lb and an average final wt of 280 lb.
[b]Diets contained 0.71, 0.89, and 1.16 % TID lysine during Phase I and 0.53, 0.71, and 0.98 TID lysine during Phase II, respectively.
[c]Diet contained 1% added methionine, copper (250 ppm), and manganese (220 ppm).
[d]Average final weight of the heaviest pig from each pen used to determine lesions and carcass data.
[e]Hot carcass weight used as the covariate in analysis.

Table 4. Main Effect of Lysine Level and Additional Methionine/Copper/Manganese on Visual Soundness Scores[ab]

	TID Lysine[c]			Met/Cu/Mn[d]			Probability, $P <$				
							Lysine				
Item	Below	Requirement	Above	Standard	Added	SED	Lys	Linear	Quadratic	Met/ Cu/Mn	Lys × Met/Cu/Mn
Front leg	2.7	2.4	2.5	2.7	2.4	0.13	0.49	0.45	0.36	0.07	0.37
Rear leg	2.8	2.4	2.6	2.6	2.6	0.12	0.28	0.39	0.18	0.69	0.28
Total[e]	5.4	4.8	5.1	5.3	4.9	0.21	0.26	0.34	0.18	0.17	0.21
Locomotion[f]	2.9	2.6	2.8	3.0	2.6	0.14	0.51	0.68	0.28	0.06	0.34

[a]Each mean represents 20 replications for Lysine treatments and 30 for Met/Cu/Mn with one pig per pen initially 89.2 lb and an average final wt of 280 lb.
[b]Front, Rear, and Locomotion scores are the mean of two evaluators for each animal.
[c]Diets contained 0.71, 0.89, and 1.16 % TID lysine during Phase I and 0.53, 0.71, and 0.98 TID lysine during Phase II, respectively.
[d]Diet contained 1% added methionine, copper (250 ppm), and manganese (220 ppm).
[e]Sum of front and rear scores according to NSIF system (7-10, excellent, 4-6, average, 1-3, poor)
[f]An independent measure of mobility scored from 1-5 (1=poor and 5=excellent) according to NSIF system.

Table 5. Main Effects of Lysine Level and Additional Methionine/Copper/Manganese on Cartilage Characteristics[a]

	TID Lysine[b]			Met/Cu/Mn[c]			Probability, $P <$				
							Lysine				
	Below	Requirement	Above	Standard	Added	SED	Lys	Linear	Quadratic	Met/ Cu/Mn	Lys × Met/Cu/Mn
Cartilage measurements											
weight, g	1.13	1.21	1.04	1.07	1.18	0.078	0.11	0.26	0.08	0.11	0.05
thickness, mm	3.47	3.28	3.18	3.34	3.28	0.252	0.52	0.27	0.83	0.74	0.06
length, mm	32.68	33.75	32.35	32.83	33.03	0.718	0.14	0.65	0.06	0.75	0.46
Instron measurements[d]											
Compression energy, newtons/g[e]	13.2	9.5	19.0	12.7	15.1	6.74	0.37	0.40	0.27	0.67	0.35
Shear energy, newtons/g[f]	558.1	432.2	591.1	538.1	516.2	59.06	0.03	0.58	0.01	0.66	0.34
Total Energy, newtons/g[g]	572.9	611.4	716.2	696.2	570.6	297.11	0.50	0.30	0.75	0.23	0.61
Ratio CF/SF[h]	0.122	0.178	0.183	0.151	0.172	0.076	0.68	0.43	0.71	0.74	0.66
RatioCE/SE[h]	0.025	0.025	0.033	0.024	0.031	0.012	0.73	0.49	0.72	0.52	0.51

[a]Each mean represents 20 replications for Lysine treatments and 30 for Met/Cu/Mn with one pig per pen initially 89.2 lb and an average final wt of 280 lb.
[b]Diets contained 0.71, 0.89, and 1.16 % TID lysine during Phase I and 0.53, 0.71, and 0.98 TID lysine during Phase II, respectively.
[c]Diet contained methionine (1%), copper (250 ppm), and manganese (220 ppm).
[d]Instron measurements were conducted on model 4201 Instron.
[e]Amount of force required in newtons per gram of cartilage to compress the cartilage half its thickness.
[f]Amount of force required to shear the cartilage into two pieces in newtons per gram of cartilage.
[g]Total amount of energy required to shear cartilage into two pieces in newtons per gram of cartilage.
[h]Ratio of compression force or compression energy to shear force or shear energy, respectively.

Table 6. Main Effect of Lysine Level and Additional Methionine/Copper/Manganese on Osteochondrosis Evaluation[ab]

	TID Lysine[c]			Met/Cu/Mn[d]			Probability, $P <$				
							Lysine				
	Below	Requirement	Above	Standard	Added	SED	Lys	Linear	Quadratic	Met/ Cu/Mn	Lys × Met/Cu/Mn
External femur											
No. abnormalities	0.7	1.4	1.3	1.5	0.8	0.33	0.11	0.08	0.23	0.02	0.73
Severity score	0.8	1.4	1.3	1.5	0.8	0.32	0.14	0.13	0.21	0.01	0.26
External humerus											
No. abnormalities	0.8	1.4	1.9	1.6	1.1	0.30	0.01	0.01	0.99	0.03	0.52
Severity score	0.7	1.3	1.7	1.4	1.0	0.26	0.01	0.01	0.74	0.03	0.62
Femur articular cartilage											
No. of faces	4.4	4.4	3.6	4.3	3.9	0.84	0.56	0.35	0.59	0.57	0.78
Severity score	1.4	1.4	1.2	1.3	1.3	0.28	0.57	0.36	0.60	0.89	0.86
Femur growth plate											
No. of faces	0.5	0.4	0.6	0.3	0.7	0.32	0.83	0.88	0.55	0.14	0.13
Severity score	0.2	0.3	0.3	0.2	0.3	0.16	0.81	0.52	0.96	0.19	0.12
Humerus articular cartilage											
No. of faces	0.9	1.6	1.9	1.5	1.4	0.70	0.36	0.16	0.81	0.86	0.61
Severity score	0.5	0.6	0.8	0.7	0.5	0.28	0.55	0.28	0.99	0.37	0.82
Overall											
Total faces	5.8	6.2	6.1	6.1	5.9	1.13	0.94	0.83	0.78	0.86	0.45
Total abnormalities[g]	7.3	8.9	9.3	9.2	7.8	1.23	0.25	0.12	0.56	0.16	0.66
Total severity[h]	3.6	4.8	5.2	5.2	3.9	0.59	0.03	0.01	0.44	0.01	0.32
Overall severity[i]	15.2	16.5	16.9	18.7	13.7	3.56	0.89	0.64	0.88	0.09	0.43

[a]Each mean represents 20 replications for Lys treatments and 30 replications for the Met/Cu/Mn treatment with pigs initially 89.2 lb and an average final wt of 280 lb.
[b]Joints were scored on a scale of 0-4 (0=normal,1=mild, 2=moderate, 3= severe, and 4=OC dissecans) for each location.
[c]Diets contained 0.71, 0.89, and 1.16 % TID lysine during Phase I and 0.53, 0.71, and 0.98 TID lysine during Phase II, respectively.
[d]Diet contained additional methionine (1%), copper (250 ppm), and manganese (220 ppm).
[e]Analysis of the number of animals with osteochondrosis determined by Cochran-Mantzel-Haenzel statistic of Proc Freq.
[f]Combined severity of osteochondrosis at the femoral articular cartilage, femoral growth plate, and humerus articular cartilage.
[g]Total number of external abnormalities and faces with lesions.
[h]Total severity of external severity scores and OC severity scores at all locations.
[i]Calculated as abnormalities multiplied by severity for each location and then summed for all locations.

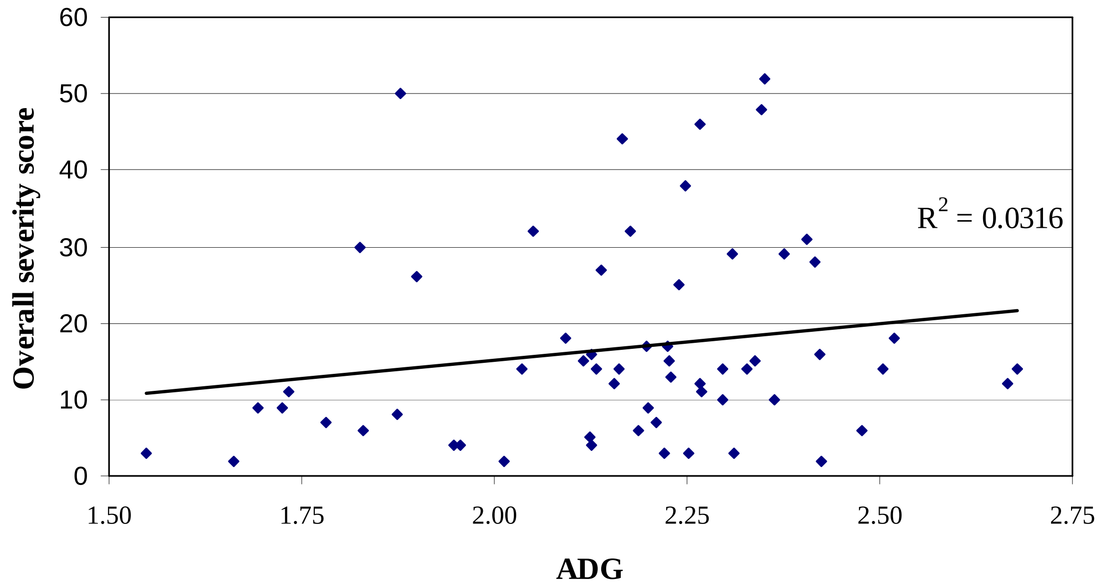

Chart 1. Average Daily Gain (ADG) versus Overall Severity Score of Osteochondrosis Using 60 Gilts.

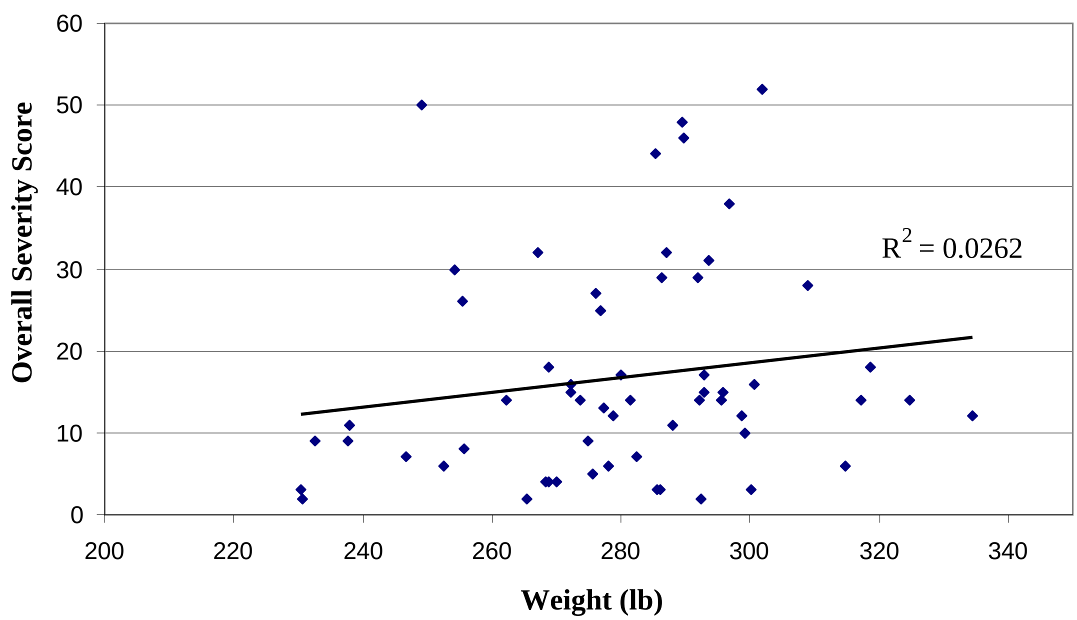

Chart 2. Weight versus Overall Severity Score of Osteochondrosis Using 60 Gilts.

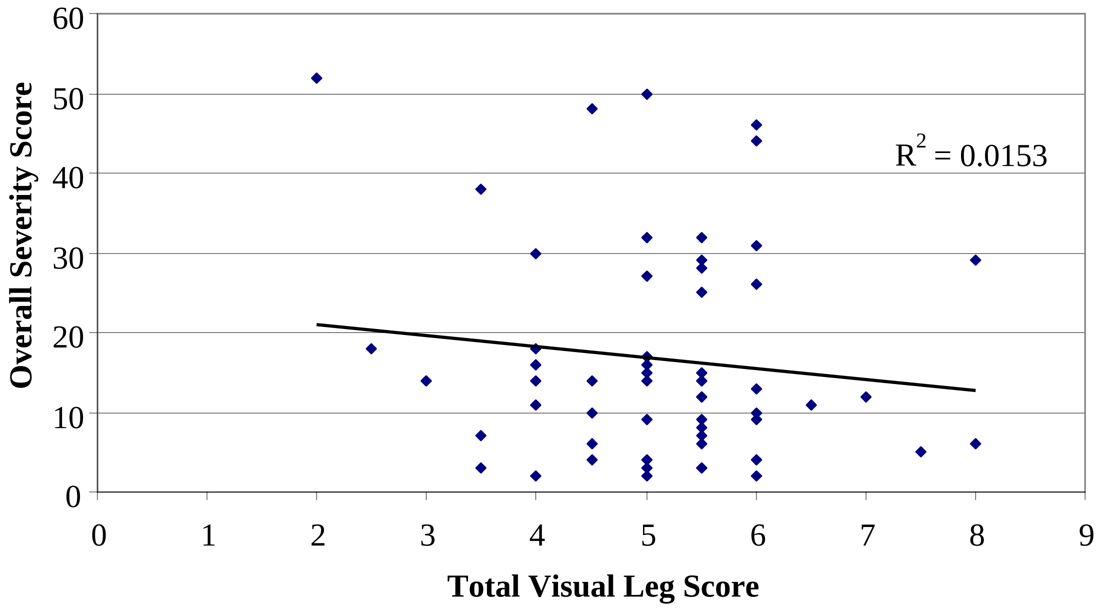

Chart 3. Total Visual Leg Score versus Overall Severity Score of Osteochondrosis. Total leg score is the sum of the front and rear leg scores, which were scored from 1-5 where 1=poor and 5=excellent from two evaluators and then summed to form the total score (2-10) according to the NSIF system on 60 gilts.

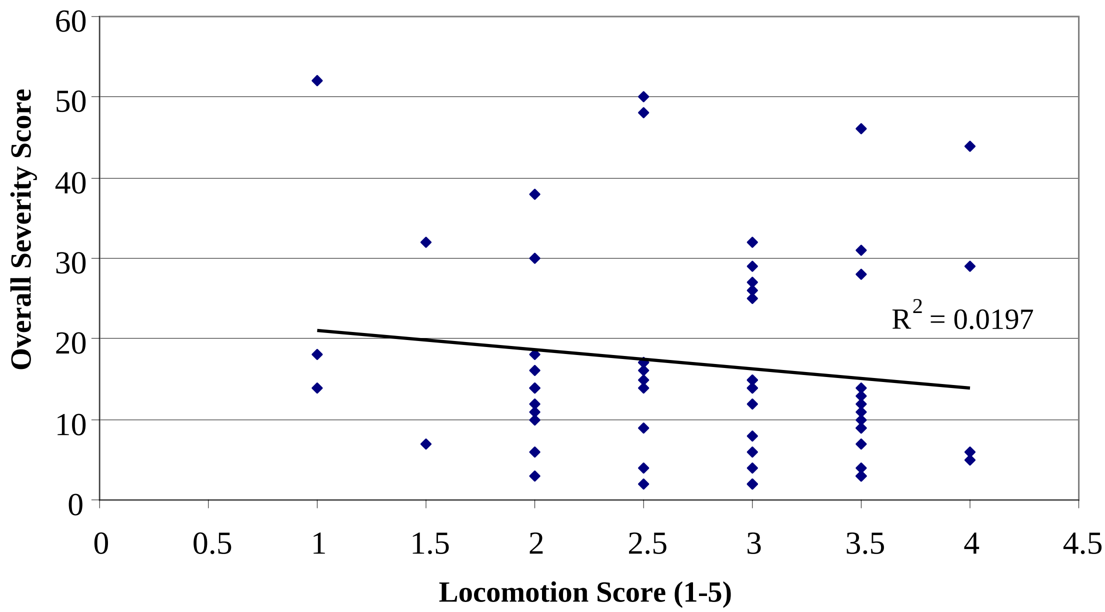

Chart 4. Locomotion Score versus Overall Severity Score of Osteochondrosis. Locomotion (measure of mobility) was scored from 1-5 where 1=poor and 5=excellent according to the NSIF system and is the average of two evaluators using 60 gilts.