

Nonruminant Nutrition: Enzymes

T203 Influences of four kinds of exogenous enzymes on performance, jejunal digesta viscosity and litter moisture of broilers fed wheat-based diet. H. Shirzadi*, H. Moravej, and M. Shivazad, *University of Tehran, Karaj, Tehran, Iran.*

The intention of this work was to determine the effects of 4 enzyme preparations containing xylanase and β -glucanase activities on performance of broiler chicks fed wheat-based diet compare with those that fed corn-based diet without enzyme. Enzyme A provided per kilogram of diet (endo-1,4- β -glucanase activity: min 800 units; endo-1,4- β -glucanase activity: min 1800 units; endo-1,4- β -xylanase activity: min 2600 units), enzyme B (endo-1,3(4)- β -glucanase: 100 AGL; endo-1,4- β -xylanase: 1100 visco units), enzyme C (endo-1,4- β -glucanase: 1500 BGU; endo-1,4- β -xylanase: 3600 FXU), enzyme D (1420 units; xylanases: 660 units). Two hundred thirty-four male day-old broiler chicks (Ross 308) were randomly allocated to 6 treatment groups, with 3 replicates and 13 birds per replicate in floor pen. All data was analyzed through the General Linear Model procedure of SAS for a randomized complete block design. The 6 dietary treatments consisted of a 60% corn-based ration without enzyme and 5 other rations containing 60% wheat supplemented with and without enzyme (A, B, C, and D). All parameters were measured at 42d, except viscosity (at 28d). Body weight was not significantly ($P > 0.05$) affected by enzyme addition. Furthermore, feed intake and feed conversion ratio were also not affected by enzyme supplementation ($P > 0.05$), and there were no significant differences between all treatment. In addition, litter moisture was not affected by enzyme, and these results were similar to results of corn-based diet ($P > 0.05$). In addition, the results demonstrated that the viscosity of jejunal contents was not significantly ($P > 0.05$) reduced by enzyme addition, however, this parameter had decreasing trend. The results of current study led to the conclusion that xylanase and β -glucanase activities of these enzymes did not seem to positively affect the xylans and β -glucans content of wheat used in this trial.

Key Words: enzyme, performance, viscosity

T204 Cloning, expression and characterization of a thermostable beta-propeller phytase from *Bacillus licheniformis*. S. J. Fu^{1,3}, J. Y. Sun¹, X. Y. Weng², L. C. Qian¹, and Z. Q. Shen⁴, ¹*Microbiology Division, Institute of Feed Science, College of Animal Science, Zhejiang University, Hangzhou Zhejiang, People's Republic of China,* ²*College of Life Science, Zhejiang University, Hangzhou Zhejiang, People's Republic of China,* ³*Key Laboratory of Preventive Veterinary Medicine and Animal Biotechnology, Binzhou Animal Husbandry and Veterinary Research Institute, Binzhou Shandong, People's Republic of China,* ⁴*Shandong Lydu Biological Technology Co., Ltd, Binzhou Shandong, People's Republic of China.*

A novel sabler phytase gene (phyC) was cloned from *Bacillus licheniformis*. It was 1146 bp in size and encoded a polypeptide of 381 amino acids. The mature peptide of phyC was successfully expressed in *Pichia pastoris* under the control of AOX1 promoter. The recombinant PhyCm (rePhyCm) was secreted into culture medium. After 96 h of 0.5% methanol induction, the activity of the rePhyCm in the culture supernatant reached the peak, 8.64 U/mg, which was 5.1 times as high as that of the native PhyC (1.7 U/mg). Studies on enzymatic properties showed that the optimum temperature and pH of the rePhyCm were 70°C and 7.5, respectively. The rePhyCm was very stable in a wide pH range of

5.0–9.0 and showed relatively good thermal stability. After incubation at the pH 5.0–9.0, 37°C for 1 h, all the residual activities of the rePhyCm were over 80%. After being exposed to 80°C for 2min in the presence of 1mM CaCl₂, the rePhyCm retained 80% of the initial enzyme activity. The rePhyCm exhibited a molecular mass of approximately 42 kDa on SDS-PAGE, indicating that the rePhyC was expressed and efficiently secreted into the growth medium.

Key Words: *Bacillus licheniformis*, phytase, *Pichia pastoris*

T205 Body weight and feed conversion responses in broilers after feeding a lysophospholipid bio-surfactant and β -mannanase based feed enzyme. G. Mathis¹, B. Lumpkins¹, H. Stomp², A. Lamptey², and A. G. Yersin^{*2}, ¹*Southern Poultry Research, Athens, GA,* ²*Kemin AgriFoods, Des Moines, IA.*

Energy utilization by poultry is dependent on many factors including age of the animal, the amount of lipase and bile salt present in the gut and overall digestibility of the ration components. The use of emulsifiers (lecithin) has been shown to improve nutrient utilization. Studies were conducted to evaluate the effects of a lysophospholipid bio-surfactant and β mannanase based feed enzyme in the diets to improve growth performance in broilers during a 42 d period. Two separate experiments were conducted using a basal corn and soybean meal ration. In study 1, there were 6 treatments based on different fat levels and the presence of the surfactant (454 g/ton inclusion), enzyme (114g/ton inclusion) or both as adjusted on a caloric basis. In study 2, the enzyme was added to all treatments, but the level of fat and surfactant were adjusted based on either a caloric basis or on the feeding phase of the ration (starter, grower or withdrawal phase). Body weight gain, feed conversion and mortality were measured at each dietary feed change in both studies. In study 1, the control (no bio-surfactant, no enzyme) group had a similar ($P < 0.05$) feed conversion and gain response at both 21 and 42 d of age as compared with treatments with either enzyme alone, bio-surfactant alone or the combination. However, the cost per pound of live weight was reduced in the bio-surfactant alone treatment compared with all others. In study 2, the enzyme and bio-surfactant combination group (fed in all phases) had a significantly better ($P < 0.05$) feed conversion and gain response at both 19 (4.9 points, 0.01 kg) and 42 d (3.8 points, 0.073 kg) as compared with all other treatments. The data indicates that both phase of feeding (starter, grower, and withdrawal) as well as the presence of enzyme with a surfactant contribute to overall performance improvements.

Key Words: broilers, bio-surfactant, enzyme

T206 Impact of a new phytase on apparent phosphorus and calcium availability, bone mineralization and performance of broilers. R. Angel^{*1}, W. Saylor², and N. Ward³, ¹*University of Maryland, College Park,* ²*University of Delaware, Newark,* ³*DSM Nutritional Products, Parsippany, NY.*

To determine the impact of adding graded levels of a new phytase (IPA, DSM) to corn-soy starter diets and to compare it with an existing commercial phytase, 9 diets with Celite as marker were fed to 8 replicates of 5 broilers each (Ross 708) from 6 to 20d: Positive Control (PC, 0.45% non-phytate phosphorus (nPP); 0.9% Ca); Moderate and Low nPP (MP and NC, 0.30 and 0.15% nPP, respectively; 0.7% Ca); NC with 250, 500, 1000, 2000, and 4000 FYT IPA/kg; and NC with 1850 Ronozyme P CT (R) (DSM). At 20 d, performance was determined, tibias sampled and

ileal contents removed for apparent Ca and P availability determination. Body weight (20 d) and feed efficiency (FE) were similar ($P > 0.05$) for birds fed the PC, MP, and the NC plus 1000, 2000 and 4000 IPA diets. Broilers fed the NC+R diet had greater BW than those fed the NC diet and similar to those fed the NC+ 200 and 500 IPA. Tibia ash was lowest ($P < 0.01$) in broilers fed the NC diet (38.1%) followed by those fed the NC+R (41.7%) and highest ($P < 0.01$) for broilers fed the PC (50.85%) and NC+4000 IPA (50.45%). There was a quadratic effect ($P < 0.001$; R-squared 0.87) of IPA inclusion on tibia ash. Apparent Ca and nPP retentions were lowest ($P < 0.001$) for broilers fed the PC diet, followed by those fed the NC diet and the NC+R and highest for broilers fed the NC+4000 IPA (51.9 and 42.2; 57.8 and 49.4; 60.7 and 51.7; and 67.9 and 60.1% for Ca and nPP, respectively). IPA inclusion had a quadratic effect ($P < 0.01$) on Ca and nPP apparent availability. In vitro analysis of the pH optima curve of IPA showed it has a higher ($P < 0.001$) P release per unit at all pHs including the 2 to 3.5 pH range compared with phytases from *Peniophora l.*- and an *E. coli*.

Key Words: broilers, phytase, phosphorus availability

T207 Effects of co-administration of phytase and energy enzymes on broiler performance, tibia strength, bone ash, and processing parameters.

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An experiment was conducted to determine if dietary NSPase inclusion enhances phytase activity during a 40-d growout. The experimental design included 4 non-supplemented diets including an industry control (3090 kcal/kg ME and 0.40 available phosphorus (aP) in the starter diet), a low energy (LE) diet (-132 kcal/kg ME), a low phosphorus (LP) (- 0.10% aP), and a low energy and low phosphorus (LEP) diet (-132 kcal/kg of ME and -0.10% aP). Enzyme supplementation treatments included phytase inclusion of 200 or 250 FTU/kg into the LP diet. In addition, the LEP diet was supplemented with an NSPase enzyme and 200 or 250 FTU/kg phytase. Each of the 8 treatment groups contained 8 replicates of 48 chicks/ replicate. Broilers were fed a starter diet through 14 d of age, a grower diet from 15 - 26 d of age, and a finisher diet through termination of the study on d 40. Following each dietary phase, 3 broilers from each replicate were killed and tibias removed for bone strength and ash determination. On d 40, 10 male broilers from each replicate were processed for determination of carcass and breast yield. Average body weight was reduced ($P < 0.05$) and mortality rates increased ($P < 0.05$) in the LP and LEP diets. Phytase inclusion in the LP diets increased ($P < 0.05$) body weight, reduced mortality, improved feed conversion ratio, and increased bone strength and ash. Phytase inclusion of 250 FTU/kg outperformed the inclusion at 200 FTU/kg in most evaluated parameters regardless of NSPase inclusion. These data confirm that phytase inclusion improves growth performance and bone strength in diets containing inadequate levels of aP, but NSPase inclusion did not enhance phytase activity.

Key Words: broiler, phytase

T208 Effect of CTCZyme β -mannanase on broiler nutrient digestibility in corn-soybean meal diets.

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The possibility of improving digestibility of nonstarch polysaccharides present in broiler diets by the use of different carbohydrases appears as an opportunity to enhance feed utilization by the birds. In this study,

the effect of a β -Mannanase (CTCZyme) on nutrient digestibility in corn-soybean meal diets was investigated. One-day-old chicks received a nutritionally complete corn-soybean meal diet for 19 d. At that time birds were randomly allocated to 4 treatments, each of which had 6 replicates of 5 birds housed in battery brooders with wire floors. Aliquots of the basal diet were supplemented with 4 levels of CTCZyme (CTC Bio Inc., Seoul Korea): 0%, 0.025%, 0.05% (recommended level) and 0.1%. Chromic oxide was used as an indigestible marker. Feed was analyzed for gross energy, Chromium, and amino acid content. After 8 d of acclimation to the test diets, birds were sacrificed and ileal contents collected. Analysis of the ileal contents indicated that digestibility of Lys, Met, Thr, Trp, Arg, Leu, Ile, Cys, and Val were significantly ($P \leq 0.0001$) improved in a linear manner for each increment of CTCZyme inclusion. Lys digestion was increased 4.6% and Met by 3.1% by the highest level of enzyme. Ileal metabolizable energy also increased with each increment of CTCZyme level. These results show that the enzyme improves feed digestibility by making amino acids more available for the bird and increases energy utilization from the feed. These results suggest that lower levels of protein and energy could be used with the same results but further studies are required to estimate potential levels.

Key Words: broilers, mannanase, digestibility

T209 Effect of phytase supplementation on the digestibility of crude protein and amino acids of cowpea (*Vigna unguiculata*) in broilers.

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Cowpea (*Vigna unguiculata*) a tropical legume is being promoted for use in poultry feeding. To determine the digestibility of crude protein (CP) and amino acids (AAs) in the bean, 6 experimental diets were formulated containing 0, 150 or 300 g/ kg of heated cowpea in place of maize starch and 0 or 500 units of phytase enzyme (Natuphos), according to a 3×2 factorial arrangement. A total of 288 d old broiler chicks (Ross strain) were distributed into 36 cages on weight basis. Each diet was assigned to 6 cages containing 8 birds each. TiO₂ was added as an indigestible marker in the diets. Increase in cowpea level had no significant effect on the amount of dietary CP and AAs digested except for arginine, glutamic acid and phenylalanine which were significantly ($P < 0.05$) reduced. Supplementation with phytase caused a significant ($P < 0.05$) increase in the digestibility of dietary CP and AAs at the terminal ileum. Interaction between cowpea and phytase had no significant effect on CP and AA digestibility in the diets. Digestibility of CP and AAs in cowpea was increased with phytase supplementation. The results showed an increase in the digestibility coefficients. Nevertheless, regression analysis indicated that phytase in this study affected both the CP and AA losses on the basal level and the digestion of CP and AAs from cowpea.

Key Words: phytase, CP and AA digestibility, broilers

T210 Effect of phytase supplementation on the digestibility of phosphorus of cowpea (*Vigna unguiculata*) in broilers.

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Cowpea (*Vigna unguiculata*) a tropical legume rich in phytate phosphorus is being promoted for use in poultry feeding. To determine the digestibility of P in the bean and the performance of the birds, 6 experimental diets were formulated containing 0, 150 or 300 g kg⁻¹ of heated cowpea in place of maize starch and 0 or 500 units of phytase enzyme (Natuphos), according to a 3×2 factorial arrangement. A total of 288 d old broiler chicks (Ross strain) were distributed into 36 cages on weight basis. Each diet was assigned to 6 cages containing 8 birds each. TiO₂ was added as an indigestible marker in the diets. Phytase supplementa-

tion had a significant ($P < 0.05$) improvement on the digestibility of P in the diets and cowpea at the terminal ileum. The digestibility of P in the cowpea increased from 0.55 to 0.67 with phytase supplementation. The level of cowpea and its interaction with phytase had no significant effect on P digestibility and reduction in the loss of P at the basal level. Phytase supplementation caused a significant ($P < 0.05$) increase in feed intake and body weight in the birds but cowpea level or its interaction with phytase had no significant effect on the performance parameters. Results of the study showed that heated cowpea can be included at the rate of 300 g/kg in diets of broilers and that supplementation of such diets with phytase resulted increased P digestibility and better performance in broilers.

Key Words: phytase, phosphorus digestibility, broilers

T211 Effect of Ronozyme ProAct supplementation on growth and meat yield responses of broilers during a forty-two-day production period. W. A. Dozier III¹*, N. E. Ward², and S. L. Vieira³, ¹*Auburn University, Auburn, AL*, ²*DSM Nutritional Products, Inc., Parsippany, NJ*, ³*Universidade Federal do Rio Grande do Sul, Porto Alegre, RS, Brazil*.

Large price differentials between corn and soybean meal have resulted in strategies to reduce diet cost for broilers. A novel protease (Ronozyme ProAct) has been reported to improve amino acid digestibility of feed ingredients. This study examined growth and meat yield responses of broilers provided diets with reduced amino acid density supplemented with or without Ronozyme ProAct. Sixteen hundred Ross × Ross 708 male broilers were randomly distributed into 64 floor pens (25 birds per pen; 0.09 m²/bird) at 1 d of age and fed 5 dietary treatments until 42 d of age. Dietary treatments consisted of a 1) Positive Control (PC), 2) Negative Control (NC) 4% reduction in amino acid density, 3) Dietary treatment 2 + 200 ppm of Ronozyme ProAct, 4) NC 6% reduction in amino acid density, and 5) Dietary treatment 4 + 200 ppm of Ronozyme ProAct. Dietary amino acid specifications of the PC were considered to be moderate density. Dietary treatments 2 to 5 had amino acid density reduced by 4 or 6% compared with the PC. Primary ingredients consisted of corn, soybean meal, distillers dried grains with solubles (5% inclusion), and meat and bone meal (5% inclusion). Each treatment was represented by 12 replicate pens. BW gain, feed intake, feed conversion, mortality, and processing yields were assessed. Ronozyme ProAct supplementation increased ($P \leq 0.001$) BW gain from 1 to 14 d of age compared with NC with both 4% and 6% amino acid reductions and Ronozyme ProAct inclusion improved ($P \leq 0.02$) 14 d feed conversion with decreasing amino acid density by 4%. Broilers fed the PC diet had advantages ($P \leq 0.05$) in growth performance, carcass weight, and total breast meat weight compared with NC diets. In summary, Ronozyme ProAct supplementation improved growth performance during the starter period.

Key Words: amino acid, broiler, protease

T212 Influences of several enzyme containing β -glucanase and xylanase on meat yield of broilers fed barley-based diet. H. Shirzadi*, H. Moravej, M. Shivazad, and F. Fatehi, *Department of Animal Science, Faculty of Agriculture and Natural Resources, University of Tehran, Karaj, Tehran, Iran*.

The goal of this investigation was to compare the effects of 4 enzyme preparations containing β -glucanase and xylanase activities on performance and meat yield of broiler chicks fed barley-based diet with and without enzyme. Enzyme A provided per kilogram of diet (endo-1,4-

β -glucanase activity: min 800 units; endo-1,(4)- β -glucanase activity: min 1800 units; endo-1,4- β -xylanase activity: min 2600 units), enzyme B (endo-1,3(4)- β -glucanase: 100 AGL; endo-1,4- β -xylanase: 1100 visco units), enzyme C (endo-1,4- β -glucanase: 1500 BGU; endo-1,4- β -xylanase: 3600 FXU), enzyme D (1420 units; xylanases: 660 units). One hundred ninety-five male day-old broiler chicks (Ross 308) were randomly allocated to 5 treatment groups, with 3 replicates and 13 birds per replicate in floor pen. All data was analyzed through the General Linear Model procedure SAS for a randomized complete block design. The 5 dietary treatments consisted of the barley (60%) supplemented with and without enzyme (A, B, C, and D added on top to diets). All parameters were measured at 42d. Body weight was increased by addition all enzymes ($P < 0.05$), and significant differences were not observed among all enzymes. However, feed intake was not significantly affected by enzyme supplementation. Feed conversion ratio, carcass weight and relative weight of the abdominal fat were significantly improved by addition all enzymes compared with the barley-based diet without enzyme ($P < 0.05$), and no significant differences were found among all enzymes. Carcass yield, relative weight of the breast, legs, liver, and gizzard as percentage of live weight were not affected by enzyme supplementation ($P > 0.05$). The results of current study led to the conclusion that there were similar improvements on performance and meat yield of birds fed diets with enzyme supplementation. Therefore, choice preference of supplementation should be based on its economic value.

Key Words: xylanase, β -glucanase, meat yield

T213 Effect of high levels of phytase for broilers. C. Meneghetti, A. G. Bertechini*, J. A. G. Brito, and S. F. Castro, *Universidade Federal de Lavras, Lavras, MG, Brazil*.

Three trials were carried out to evaluate the effects of supplementation of *Citrobacter braaki*, an *E. coli* derived phytase preparation (Genophos), on the performance, AMEn, retention and excretion of minerals and bone ash of broiler chickens. Broiler performance was evaluated using 3-phase feeding program (1–21 d, 22–35 d and 36–42 d of age). In trial 1, a total of 1456 d-old, male Cobb 500 broiler chicks were placed in 56 litter floored pens in a complete randomized design with 8 replicates/treatment. The treatment diets were: (1) Control (C-SBM based diet and no added phytase); (2) 1,500; (3) 3,000; (4) 4,500; (5) 6,000; (6) 8,000 and (7) 10,000 FTU/kg of supplemented phytase. All treatments with phytase had reduction in ME of 85 kcal/kg, Ca of 0.2% and available P 0.12%. In trial 2, 280 10-d-old male Cobb 500 broiler chicks were allotted in 56 metabolism cages to obtain the apparent digestibility of DM, CP, AMEn, Ca and P retention using the same diet from 1 to 21 d of the trial 1, and the excreta collection from 15 to 17 d of age. For trial 3, 168 28-d-old male Cobb 500 broiler chicks were placed in the same diet from 22 to 35 d of trial 1 and excreta collection between 33 and 35 d of age. Results showed that bird performance was similar of controls when 4,500 FTU/kg or higher of phytase inclusion occurred from 1 to 21 d. From 1 to 35 and 1–42 d, the treatments were comparable to control. A quadratic effect ($P < 0.05$) was observed (10–17 d) for AMEn and Ca retention at maximum values obtained with 7,727 and 5,500 FTU/kg of diet, respectively. A linear ($P < 0.05$) improvement in P retention was observed with increased phytase supplementation. At 32–35 d, a quadratic effect ($P < 0.05$) was observed on Ca retention with maximum values at 5,000 FTU/kg of diet. P retention increased (linear effect) with phytase supplementation ($P < 0.05$). Ash, Ca and P in the tibia (42 d) were not affected by treatments. Phytase supplementation starting at 4,500 FTU/kg improved broiler performance challenged in ME, Ca and P.

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Key Words: available P, Ca retention, nutrient digestibility

T214 Effect of enzymes in the diet of hens on egg quality. F. G. P. Costa^{*1}, M. L. Ceccantini², C. S. Santos¹, C. C. Goulart¹, C. F. S. Oliveira¹, G. B. V. Lobato¹, J. M. Freire¹, V. P. Rodrigues¹, R. C. Lima¹, I. S. Nobre¹, and R. C. L. Neto¹, ¹Federal University of Paraiba, Areia, PB, Brazil, ²Adisseo Brazil Animal Nutrition, Sao Paulo, SP, Brazil.

This study aims to demonstrate the technical feasibility and economic use of the set of a 6-phytase, 17 Carbohidrolases and 2 proteases (from the supply Rovabio Max AP) in diets for laying hens. The experiment was divided in 5 periods of 28 d each, which were used 360 hens, distributed in a completely randomized design with 5 treatments and 8 replicates of 9 birds per experimental unit. The treatments consisted of 2 diets without enzyme and 3 with the addition of the enzyme. Diets without enzymes were negative and positive controls. The positive control diet (PC) was formulated to meet the requirements of hens. The negative control diet (NC) was formulated with nutrient reduction (100 kcal / kg and 0.36, 0.014, 0.012, 0.12, and 0.153% in the levels of CP, lysine, methionine + cystine, calcium and phosphorus available, respectively). Diets supplemented with 50 g / ton of Rovabio Max AP were formulated to address different levels of linoleic acid, one with the linoleic acid diet equal to CN, the other with an intermediate value and the last with linoleic acid equal to diet PC. These diets have been reformulated considering the nutritional enzyme formulation. Thus, the three diets supplemented with enzymes have been reformulated to meet the nutritional levels of the control diet, except for linoleic acid.). All diets based on corn and soybeans. The data were subjected to analysis of variance with means tested with Tukey and orthogonal contrasts. The weight and percentage of egg specific gravity and pigmentation of the eggs were influenced ($P < 0.05$) by nutrition and supplementation with enzymes. Enzyme supplementation Rovabio Max AP proves efficient in the availability of nutrients to improve the quality of eggs.

Key Words: additive, linoleic acid, production

T215 Use of enzyme complex on the performance of layer hens. F. G. P. Costa^{*1}, M. L. Ceccantini², C. S. Santos¹, C. C. Goulart¹, C. F. S. Oliveira¹, G. B. V. Lobato¹, and J. M. Freire¹, ¹Federal University of Paraiba, Areia, PB, Brazil, ²Adisseo Brazil Animal Nutrition, Sao Paulo, SP, Brazil.

This study aims to demonstrate the technical feasibility and economic use of the set of a 6-phytase, 17 Carbohidrolases and 2 proteases (from the supply Rovabio Max AP) in diets for laying hens. The experiment was divided in 5 periods of 28 d each, which were used 360 hens, distributed in a completely randomized design with 5 treatments and 8 replicates of 9 birds per experimental unit. The treatments consisted of 2 diets without enzyme and 3 with the addition of the enzyme. Diets without enzymes were negative and positive controls. The positive control diet (PC) was formulated to meet the requirements of hens. The negative control diet (NC) was formulated with nutrient reduction (100 kcal / kg and 0.36, 0.014, 0.012, 0.12, and 0.153% in the levels of CP, lysine, methionine + cystine, calcium and phosphorus available, respectively). Diets supplemented with 50 g / ton of Rovabio Max AP were formulated to address different levels of linoleic acid, one with the linoleic acid diet equal to CN, the other with an intermediate value and the last with linoleic acid equal to diet PC. These diets have been reformulated considering the nutritional enzyme formulation. Thus, the three diets supplemented with enzymes have been reformulated to meet the nutritional levels of the control diet, except for linoleic acid. The production and feed conversion per dozen eggs were affected ($P < 0.05$) by nutrition and supplementation with enzymes. The enzyme Rovabio Max AP was effective in providing nutrients from food for birds. Despite reductions in nutrient levels are higher than recommended by

the manufacturer, the use of carbohidrolase, phytase e protease ensures the same productivity in the control group, confirming the effectiveness of the enzyme complex.

Key Words: additive, linoleic acid, production

T216 Dietary supplementation with two types of enzyme preparations improves nutrient digestibility in growing pigs. X. Ao^{*1}, S. M. Hong¹, H. Y. Park², K. H. Son³, B. H. Ku³, D. H. Shin³, and I. H. Kim¹, ¹Department of Animal Resource and Science, Dankook University, Cheonan, Choongnam, Korea, ²Korea Research Institute of Bioscience and Biotechnology, Daejeon, Korea, ³Insect Biotech Co. Ltd., Daejeon, Korea.

The objective of this experiment was to determine the effects of 2 types of enzyme preparations (single or complex) on fecal digestibility and ileal apparent digestibility (AID) in the hindgut of growing pigs. Three ileal-cannulated growing barrows (28.56 ± 0.58 kg) were housed in individual metabolism crates and randomly assigned to 1 of 3 treatments within a 3×3 Latin square design. The treatments were as follows: 1) CON (basal diet), 2) P1 (basal diet + 0.1% protease), and 3) P2 (basal diet + 0.1% complex enzyme including protease, xylanase and lipase). The 3 feeding periods consisted of 4 d of acclimation to the diet followed by 5 d of fresh feces samples collection and 6 and 7 d of ileal-digesta collection. Ileal effluents were continuously collected for the same 12-h interval each day. Pigs fed the P1 diet had a higher DM and energy digestibility ($P < 0.05$) than pigs fed the CON and P2 diets, while the N digestibility was greater ($P < 0.05$) in the CON and P1 groups than in the P2 group. The AID of DM and energy was increased ($P < 0.05$) in the P1 and P2 groups when compared with the CON group, with the P1 treatment showing the highest levels. The AID of N digestibility was also greater ($P < 0.05$) for pigs fed the P1 diet than for those fed other diets. Pigs fed the P1 diet had a higher AID of total essential AA and non-essential AA ($P < 0.05$) than those fed the CON and P2 diets. A similar tendency was also observed in individual AA ileal digestibility. In conclusion, these results showed that the single protease addition improved nutrient digestibility.

Key Words: enzyme, nutrient digestibility, growing pigs

T217 Effects of dietary Tylan inclusion level on the growth performance and carcass characteristics of growing-finishing pigs. C. L. Puls^{*1}, M. Mercedes¹, M. Ellis¹, A. M. Gaines², B. A. Peterson², B. F. Wolter², and M. Kocher², ¹University of Illinois, Urbana, ²The Maschhoffs, Carlyle, IL.

This study was carried out to evaluate the effect of including Tylan in the diet of growing-finishing pigs on growth performance and carcass characteristics. The study was carried out from 39.9 ± 1.45 kg to 127.2 ± 1.13 kg BW and compared 2 Tylan inclusion levels (0 vs. 10 g/ton). Diets were formulated to meet or exceed the nutrient requirements proposed by NRC (1998). The study involved 144 barrows housed in pens of 9 with 8 pens per inclusion level. Pigs had ad libitum access to feed and water throughout the study period. At the end of the study period, pigs were sent to a commercial facility for harvest and carcass evaluation. There was no effect ($P > 0.05$) of Tylan inclusion level on overall ADG, ADFI, or G:F ratio. Mortality levels were lower for pigs fed Tylan compared with the control (0.00 vs. 2.82%, respectively); however, this difference was not significant ($P = 0.15$). There was a trend ($P = 0.06$) for carcass yield to be greater for pigs fed Tylan compared with the control (74.9 vs. 74.3%, respectively; SEM 0.42) and this resulted in an improvement ($P < 0.01$) in carcass G:F ratio for pigs fed Tylan (0.26 vs. 0.25 kg:kg, respectively; SEM 0.004). There was no

effect ($P > 0.05$) of Tylan inclusion level on back fat thickness, *Longissimus* muscle depth, or predicted carcass lean content. In conclusion, this study suggests that feeding Tylan (at 10 g/ton) to growing-finishing pigs does not affect live weight growth performance but could reduce mortality, and improve carcass yield and carcass feed efficiency, findings that warrant validation.

Key Words: pigs, growth, Tylan

T218 Effect of a protease enzyme on performance of weanling piglets fed corn-soybean diets with different protein levels. D. Wang¹, X. Piao¹, F. C. Guo², H. Cao², J. Zhao², and R. J. Harrell*², ¹China Agricultural University, Beijing, China, ²Novus International Inc., St Charles, MO.

Application of exogenous enzymes can improve the digestibility of feedstuffs, lower dietary costs, and improve animal performance. The objective of the present study was to examine the benefits of supplementing nursery pig diets with a protease enzyme (NZ) at 2 levels of dietary protein. A total of 190 pigs (8.31 ± 0.63 kg of BW) were allotted by weight and sex to one of 4 treatments in a 2 × 2 factorial arrangement with the factors being high (HP, 20.3% CP) vs. low dietary protein (LP, 18.3% CP) and 0 vs. 500 mg/kg protease (Cibenza DP100, Novus International Inc., St. Charles, MO) for a period of 21 d with 6 replicates (n = 7 or 8 pigs/pen). All the diets were formulated according to an ideal amino acid pattern. CP level did not alter ADFI ($P > 0.83$), ADG ($P > 0.73$), or GF ($P > 0.57$). No differences were observed for the NZ on ADG ($P > 0.20$) or ADFI ($P > 0.22$). The GF was improved by 17% with the addition of NZ (0.623 vs. 0.728 ± 0.016; $P < 0.01$), regardless of dietary CP level. The digestibility of DM, OM, or energy was not affected by dietary CP level ($P > 0.45$). The addition of NZ increased CP digestibility by 3.5% (84.3 vs. 87.2 ± 0.5%, $P < 0.01$), increased DM digestibility by 2.2% (86.1 vs. 88.0 ± 0.4%, $P < 0.01$), and energy by 2.3% (86.5 vs. 88.5 ± 0.4%, $P < 0.01$), regardless of dietary CP level. Blood urea nitrogen levels were higher in pigs fed HP compared with LP (4.0 vs. 2.58 ± 0.21 mmol; $P < 0.01$). The addition of NZ interacted with CP level on BUN levels by increasing BUN in HP, but not in LP dietary CP ($P < 0.02$). These results indicate that the addition of a protease enzyme can improve digestibility of feedstuffs and improve feed efficiency in nursery pigs.

Key Words: swine, nursery, protease

T219 Effects of supplementing different enzymes on performance, nutrient digestibility and blood metabolites in growing pigs. J. K. Jo¹, P. L. Shinde¹, J. S. Kim¹, Y. W. Kim¹, K. H. Kim¹, J. D. Lohakare¹, C. S. Ra¹, J. H. Lee², and B. J. Chae*¹, ¹Kangwon National University, Kangwon National University, Chuncheon, Rep. of Korea, ²CTC Bio. Inc., CTC Bio. Inc., Seoul, Rep. of Korea.

A 28-d growth study was conducted to investigate the effects of mannanase (M), amylase + mannanase (AM), mannanase + protease (MP) and amylase + mannanase + protease (AMP) supplementation on the performance, apparent total tract digestibility (ATTD) of nutrients and blood metabolites in growing pigs. A total of 240 growing pigs (initial BW 55.58 ± 0.85 kg) were randomly allotted to 5 treatments on the basis of BW. Each treatment was comprised of 4 replicates with 12 pigs in each. A corn-soybean meal based diet (control) was supplemented with 0.05% M, AM, MP or AMP as dietary treatments. Pigs fed enzyme supplemented diets had greater ($P < 0.05$) ADG and ATTD of DM than pigs fed control diet. Moreover, pigs fed AMP diet had higher ($P < 0.05$; 949 vs. 918, 933 and 929 g) ADG than pigs fed M, MP and AM diets. Pigs offered AMP and MP diets had better F/G ($P < 0.05$; 2.91 and 2.95

vs. 3.05 g/g) than pigs fed control diet. In addition, pigs fed MP diet gained more ($P < 0.05$; 933 vs. 918 g) when compared with pigs offered M diet. The ATTD of DM was higher ($P < 0.05$; 81.29 vs. 80.50%) in pigs fed AMP diet when compared with pigs fed M diet; while the ATTD of CP was higher ($P < 0.05$; 76.44 vs. 75.24 and 75.21%) in pigs fed AMP diet when compared with pigs fed AM and control diets. Additionally, the blood urea nitrogen concentration was greater ($P < 0.05$; 17.75 vs. 14.15 and 13.50 g/dl) in pigs fed MP diet when compared with pigs fed AM and control diet. These results indicate supplementation of different enzymes in combination (mannanase + protease and amylase + mannanase + protease) to be more efficient in improving the performance and nutrient digestibility in growing pigs.

Key Words: enzymes, performance and nutrient digestibility, growing pigs

T220 Evaluation of the effects of dietary enzyme on growth performance, nutrient digestibility, blood characteristics and ileal digestibility in growing pigs. L. Yan*, H. D. Jang, T. X. Zhou, X. Ao, J. H. Jung, and I. H. Kim, Department of Animal Resource and Science, Dankook University, Cheonan, Choongnam, Korea.

Two experiments with growing pigs were conducted to investigate the effects of 2 distinct multi-enzyme preparations on nutrient digestibility, growth performance and blood profiles. In Exp.1, a total of 96 pigs (29.7 ± 0.69 kg) were used in a 42-d performance and digestibility trial using 4 dietary treatments: CON (control diet), E (control+0.10% Endopower), N1 (control+0.10% NSPase) and N2 (control+0.20% NSPase). Endopower is a commercial multi-enzyme preparation that contains α-galactosidase, galactomannase, xylanase and β-glucanase. NSPase primarily contained α-1,6-β-galactosidase, β-1,4-mannanase, and β-1,4-mannosidase. There were 6 replication pens per treatment with 4 pigs per pen. Pigs fed the N1 diet had a higher ADG ($P < 0.05$) and G:F ($P < 0.05$) than those fed the control diet. There were no significant differences in the ADG and G:F among the multi-enzyme treatments ($P > 0.05$). When compared with the control, the apparent digestibility of DM was increased ($P < 0.05$) by E treatment. The N digestibility was improved ($P < 0.05$) in response to multi-enzyme treatments during the experimental period. In addition, BUN was higher ($P < 0.05$) in the E treatment group than in the CON and N1 treatment groups at the end of the experiment, while the level of glucose was improved ($P < 0.05$) by E and N2 treatments. In Exp.2, 4 ileal-cannulated growing barrows (20.17 ± 1.31 kg) were housed in individual metabolism crates and randomly assigned to 1 of 4 treatments (same as Exp.1) within a 4 × 4 Latin square design. Enzyme supplementation improved the majority of apparent ileal AA digestibility ($P < 0.05$). These findings demonstrated that supplementation of the diet of these enzymes could improve the feeding values of growing pigs fed a diet based on corn and soybean meal.

Key Words: enzyme, digestibility, growing pigs

T221 Protease increased in vitro digestibility of various feed ingredients. F. Yan*, P. Disbennett, M. Schulz, M. Vazquez-Anon, N. Odettallah, S. Carter, and D. Dowell, Novus International Inc., St. Charles, MO.

A protease (CIBENZA DP100, Novus International Inc.) was evaluated in an in vitro system to evaluate its ability to degrade proteins in a variety of feed ingredients and it was also compared with a commercial protease, denoted as Protease B. Soybean meal, canola meal, cottonseed meal, DDGS, corn, wheat, lupin, poultry meal, meat and bone meal, fish meal, feather meal, and blood meal were tested in the study. Each

test ingredient was solubilized for 4 h at pH 2.4 and protein concentration of 8 mg/ml, buffered to pH 7.5, and then incubated with either DP100 or Protease B for 18 h at 37°C, at an enzyme concentration that it would encounter in the digesta in vivo when it is used at the recommendation dose based on the assumption of water intake being twice of feed intake. For negative control, deionized water was added. After 18 h of incubation, OPA (o-phthalaldehyde) analysis with UV-vis spectroscopy was performed to measure β \pm -amino groups. All tests were performed in quadruplicate. Both DP100 and Protease B demonstrated proteolytic activity for all ingredients tested ($P < 0.0001$) compared with the negative control, which indicated broad substrate specificity of these proteases. Their ability to hydrolyze proteins from different feed ingredients varied where they were most effective in hydrolyzing blood meal proteins and less efficient for corn and feather meal proteins. Cibenza DP100 outperformed Protease B for all ingredients ($P < 0.05$) except for corn. The percent increase in absorbance of DP100 over Protease B was 23.0% for soybean meal, 27.3% for canola meal, 42.0% for cottonseed meal, 59.4% for DDGS, 39.5% for wheat, 51.6% for lupin, 13.4% for poultry meal, 20.9% for meat and bone meal, 12.8% for fish meal, 7.3% for feather meal, and 32.1% for blood meal. These results demonstrated efficacy of CIBENZA DP100 in digesting proteins from various feed ingredients and it can be beneficial in improving protein digestion of animals fed diets containing these ingredients.

Key Words: protease enzyme, in vitro, digestibility

T222 Effects of graded levels of phytase on the apparent and standardized total tract digestibility of phosphorus in corn and corn co-products. F. N. Almeida* and H. H. Stein, *University of Illinois, Urbana.*

An experiment was conducted to measure the effects of graded levels of microbial phytase on the standardized total tract digestibility (STTD) of P in corn, distillers dried grains with solubles (DDGS), high protein distillers dried grains (HP-DDG), and corn germ. A second objective was to develop regression equations to predict the response of adding phytase to each of these ingredients. Four corn based diets, 4 DDGS based diets, 4 HP-DDG based diets, and 4 corn germ based diets were formulated. The 4 diets with each ingredient contained 0, 500, 1,000, or 1,500 phytase units (FTU) per kg (Optiphos 2000, Enzyvia, Sheridan, IN). A P-free diet was also formulated to measure the basal endogenous losses of P. A total of 102 pigs (initial BW: 18.2 ± 2.1 kg) were individually housed in metabolism cages equipped with a feeder and a nipple drinker and a screen floor that allowed for total collection of feces. Pigs were allotted to the 17 diets in a randomized complete block design with 6 replicates per diet. Supplementation of microbial phytase increased (linear, $P < 0.01$; quadratic, $P < 0.05$) the STTD of P in corn from 40.9 to 67.5, 64.5, and 74.9%, tended to increase (linear, $P = 0.07$) the STTD of P in DDGS from 76.9 to 82.9, 82.5, and 83.0%, increased (linear, $P < 0.01$; quadratic, $P < 0.05$) the STTD of P in HP-DDG from 77.1 to 88.0, 84.1, and 86.9%, and increased (linear and quadratic, $P < 0.01$) the STTD of P in corn germ from 40.7 to 59.0, 64.4, and 63.2% in diets supplemented with 0, 500, 1,000, or 1,500 FTU/kg of phytase, respectively. Regression equations were developed to allow the calculation of the STTD of P with any level of phytase (Optiphos 2000, Enzyvia, Sheridan, IN) for each of the test ingredients. Therefore, results of this experiment allow the prediction of the amount of digestible P in these ingredients containing any level of phytase between 0 and 1,500 FTU.

Key Words: digestibility, phosphorus, phytase

T223 Effects of multi-enzyme on nutrients digestibility and metabolizable energy values of pure corn and wheat diets. G. G. Zhang*, Z. B. Yang, Q. Q. Zhang, W. R. Yang, and S. Z. Jiang, *Shandong Agricultural University, Taian, China.*

Objectives of this study were to determine effects of enzyme supplementation in pure corn- and wheat-based diets on energy efficiency and nutrients utilization in poultry. By force-fed true metabolizable energy bioassay (TME Bioassay) method, a metabolic trial was conducted, 90 healthy adult roosters with similar weight were randomly assigned to 10 dietary groups with 3 replicates of 3 roosters for each, 2 of the groups (controls) were not force-fed during experiment for collecting endogenous excreta, other groups were force-fed corn with different levels multi-enzyme (0, 50, 100, and 150 mg/kg of diet), or force-fed wheat with different levels multi-enzyme (0, 50, 100, and 150 mg/kg of diet), respectively. The results showed the use of multi-enzyme preparation in corn enhanced the apparent/true digestibility (AD/TD) of neutral detergent fiber (NDF) ($P < 0.05$), dry matter (DM) ($P < 0.05$), and starch ($P < 0.05$), the apparent metabolizable energy (AME) value and true metabolizable energy (TME) value were increased by 6.76%, and 5.86% respectively. Inclusion of multi-enzyme in wheat improved the AD/TD of NDF ($P < 0.05$), DM ($P < 0.05$) and starch ($P < 0.05$), AME and TME were enhanced by 9.75% and 8.88% respectively. Nutrients digestibility and metabolizable energy values showed linear or quadric increasing trend, with the amount of multi-enzyme added in corn and wheat. It appears from this study that the nutrients utilization of pure corn or wheat diet could be enhanced by using appropriate multi-enzyme supplement. However, an proper addition is necessary in corn to maximize the nutrients utilization and energy conversion, and higher rates of supplementation did not lead to further increase in nutrients digestibility and ME values.

Key Words: multi-enzyme, digestibility, metabolizable energy

T224 Effect of Rovabio Max on energy and nitrogen utilization in diets high in distillers dried grains with solubles. A. J. Karimi*², Y. Min¹, J. H. Park¹, C. A. Coto¹, C. Lu¹, F. Yan¹, and P. W. Waldroup¹, ¹*University of Arkansas, Fayetteville,* ²*University of Kurdistan, Kurdistan, Iran.*

The inclusion of DDGS in poultry diets is limited due the reduced content of starch and high levels of Non Starch Polysaccharides (NSP). An improvement on the nutrient availability through the use of exogenous enzymes represents an alternative to increase its utilization. Rovabio Max is a preparation containing xylanases, β -glucanases, pectinases, mannanases, phytase and α -galactosidase. A study was conducted to evaluate the use of Rovabio Max on utilization of diets containing high levels of DDGS. The experimental design consisted of a 2×4 factorial arrangement. Two isocaloric (ME = 3020 kcal/kg) basal diets were formulated, one with no DDGS and the second with 30% DDGS of known composition. Chromic oxide was used as an indigestible marker. Aliquots of the 2 basal diets were supplemented with no enzyme or 3 levels of Rovabio: The recommended level (1X), twice (2X) and 4 times (4X) the recommended level. One hundred and 90 2 male chicks of a commercial strain (Cobb 500) were fed a common nutritionally complete diet to 18 d at which time they were placed on the study. Each experimental diet was fed to 4 pen replicates of 6 male chicks in wire floor battery cages. After a 5-d adaptation period, excreta samples were collected and freeze-dried to determine GE, AME and N retention. Birds were weighed and feed consumption determined. The ANOVA considered DDGS level, enzyme, and interaction. No significant ($P > 0.05$) effect of Rovabio, DDGS and their interactions was found on body weight, feed intake, feed conversion and mortality. No effect of the enzyme was found for

GE, N in excreta, N retention, AME and AMEn. No interaction between DDGS and Rovabio was found for nitrogen and energy utilization. The inclusion of 30% DDGS in the diet significantly ($P < 0.05$) increased the GE and N in excreta with no effect on AME and AMEn values. The increased concentration of nutrients in excreta deserves consideration due to environmental implications.

Key Words: DDGS, enzymes, digestibility

T225 Effect feed processing method and enzyme supplementation of wheat-based diets on performance, gastrointestinal and carcass characteristics in broiler chicks. Z. Qobadi and A. Karimi*, *University of Kurdistan, Sanandaj, Kurdistan, Iran.*

This study was carried out to compare the effects of feed processing (pelleted vs. mash) and enzyme supplementation (with and without 0.3g Grindazym GP 15000 /kg of wheat in complete diet) in a wheat-based diet on performance, gastrointestinal and carcass characteristics of broiler chicks to 36 d of age. Ross 308 straight-run broiler chicks ($n = 336$) were randomly allocated to 4 dietary treatments, each replicated 4 times (21 chicks per pen) in a completely randomized design in a 2×2 factorial arrangements. Measurements included body weight (BW), daily gain (DG), feed intake (FI) and feed conversion ratio (FCR) at 20 and 36 d of ages. The relative weights of gastrointestinal organs to body weight were determined at 20 and 36 d of ages. The pH of ileum digesta content was also determined at 20 d of age. The results showed that the broiler chicks fed pelleted diets had significantly improved BW at 36 d of age, DG during 20 to 36 and 0 to 36 d of ages and FI during 0 to 20, 20 to 36 and 0 to 36 d growth period. Feed conversion ratio was significantly increased in pelleted fed treatment during 0–20 and 0–36 d growth period. Enzyme supplementation had significantly improved BW at 20 and 36 d of age, DG during 0–20, 20–36 and 0–36 d of ages, FI during 0 to 20 and 0 to 36 d of ages and FCR during 0–36 d of ages. The interaction between feed processing and enzyme supplementation was only significant on FCR during 20–36d. Neither feed processing nor enzyme supplementation had significant effects on ileum digesta pH measurements, carcass and gastrointestinal characteristics, except gizzard relative weight at 36 d of age. In conclusion, the results of this experiment confirmed the beneficial effects of both pelleting and feed enzyme supplementation on broiler chicks' performance.

Key Words: wheat, processing, enzyme

T226 Calcium chloride reduces the negative impact of feeding high potassium and co-product containing diets to finishing pigs. J. Guimaraes*, C. L. Zhu, D. Wey, and C. F. M. de Lange, *University of Guelph, Guelph, Ontario, Canada.*

Co-products from the biofuel and human food industries may serve as alternative pig feed ingredients. Previously, we observed a reduction in animal performance when pigs were liquid-fed high potassium (K) diets containing corn steep water (CSW) and whey permeate (WP). This study was conducted to investigate the addition of calcium chloride (CaCl_2) to pig diets to reduce the negative impact of feeding high K levels. A total of 192 purebred Yorkshire pigs (average initial BW 45.5 kg; 4 gilts and 4 barrows per pen) were liquid-fed 1 of 6 diets over a 9 week period: (1) CSBM (Corn and soybean meal based diet with added 0.9% potassium carbonate; 0.98% K); (2) CSBM/ CaCl_2 -mEq (CSBM with added 0.84% CaCl_2); (3) CSBM/ CaCl_2 (CSBM diet with added 1.05% CaCl_2); (4) Co-prod (22% WP, 6% CSW, 1.28% K); (5) Co-prod/ CaCl_2 -mEq (Co-prod diet with added 0.87% CaCl_2); (6) Co-prod/ CaCl_2 (Co-prod diet with added 0.68% CaCl_2). Diets 2 and 5 were formulated to an electrolyte balance of 166 mEq/kg; diets 3 and 6 were designed

to maintain a target balance between K and Cl. Data were exposed to analyses of variance using GLM of SAS with treatment as the only source of variation; treatment means were compared using orthogonal contrasts. Diet did not influence feed intake ($P > 0.05$). For CSBM, adding CaCl_2 at both levels improved feed to gain (2.36 vs 2.57; $P < 0.01$); such response was not seen for Co-prod ($P > 0.10$). Hot carcass weight and carcass lean yield did not differ among treatments ($P > 0.05$). For Co-prod, adding CaCl_2 reduced plasma carbon dioxide levels ($P < 0.01$). Based on quantitative histology observations, the addition of CaCl_2 to Co-prod eliminated damage to walls of glomeruli capillaries. Optimal CaCl_2 additions to high K diets are still to be confirmed. These results suggest that some of the negative effects of feeding high K co-product containing diets to pigs can be reduced, reducing the reliance on traditional feed ingredients for pigs.

Key Words: pigs, calcium chloride, co-products

T227 Production and characterization of a thermostable beta-propeller phytase from *Bacillus licheniformis*. S. J. Fu*^{1,3}, J. Y. Sun¹, X. Y. Weng², L. C. Qian¹, and Z. Q. Shen⁴, ¹*Microbiology Division, Institute of Feed Science, College of Animal Science, Zhejiang University, Hangzhou Zhejiang, China,* ²*College of Life Science, Zhejiang University, Hangzhou Zhejiang, China,* ³*Binzhou Animal Husbandry and Veterinary Research Institute, Binzhou Shandong, China,* ⁴*Shandong Lydu Biological Technology Co., Ltd, Binzhou Shandong, China.*

A novel β -propeller phytase producing thermophilic strain of *Bacillus licheniformis* was isolated from soil. The optimal fermentation parameters for producing phytase by *B. licheniformis* under shake flask culture were determined by single factor test and the results were as follows: 1.0% dextrose used as carbon source, 0.1% $(\text{NH}_4)_2\text{SO}_4$ as nitrogen source, initially pH7.5, incubation temperature 55°C. After incubation for 36h under these conditions, the activity of neutral phytase reached 0.267 U/mL with specific activity 0.701U/mg. The optimum temperature and pH of the phytase from *B. licheniformis* (PhyC) were 55°C and 7.0, respectively. After treated at 80°C, pH 7.0 for 10 min, the residual activity of PhyC was 57.36%. Over 80% of PhyC activity was retained after treatment by preincubation over a pH range of 6.5–9.0 for 1 h at 25°C. As for substrate specificity, it was very specific for sodium phytate and showed no activity on other phosphate esters. Its activity was greatly inhibited by EDTA and metal ions such as Cd^{2+} , Mn^{2+} , Cu^{2+} and Ba^{2+} .

Key Words: *Bacillus licheniformis*, phytase, characterization

T228 A Lysozyme supplement for piglets: Weaned pigs responses to *Escherichia coli* K88⁺ (ETEC) oral challenge. E. Kiarie*¹, S. Bhandari¹, D. O. Krause¹, G. Zhang², and C. M. Nyachoti¹, ¹*University of Manitoba, Winnipeg, MB, Canada,* ²*Neova Technologies Inc., Abbotsford, BC, Canada.*

Lysozyme is a low-molecular-weight protein with antimicrobial properties. An experiment was conducted to investigate response of piglets receiving Entegard (EG, a water-soluble lysozyme antimicrobial blend) upon oral challenge with ETEC. A total of 36 individually housed weanlings were randomly allotted to 1 of the 4 treatments to give 9 pens per treatment. Treatments were control (C, no additive), antibiotic (AB, in-feed) and EG (EG1 and EG2, in-water). All pigs received a basal diet similar in composition and nutrients (NRC, 1998), except AB pigs which had an added Aureo SP 250. Entegard was delivered in the drinking water: EG1, 0.1% and EG2, 0.2%. Pigs were acclimatized to treatments for a 7-d period to monitor growth performance. On d 8, each pig was bled to obtain serum and gavaged with 6 mL (2×10^9 cfu/ml) of

ETEC. Pigs were monitored for another 7 d to assess severity of diarrhea using a fecal consistency scoring system and growth performance, subsequently all pigs were killed to obtain intestinal tissues and digesta samples. Treatments did not influence ($P > 0.10$) growth performance throughout the study. More ETEC counts were observed on the ileal ($P = 0.001$) and colon ($P = 0.025$) mucosal scrapings of the C pigs than AB and EG1 pigs which in turn showed numerically lower incidences on diarrhea than C. Pigs receiving AB and EG1 had higher small intestine weight and ileal villous height than those receiving C, however, ileal villi height to crypt depth ratio for EG1 (1.56) and EG2 (1.38) was similar to that of AB (1.68) pigs which was in turn higher than that of C (1.34) pigs. Pigs in the EG1 group showed higher ($P < 0.001$) serum tumor necrosis α (TNF- α) and interleukin 6 (IL-6) before ETEC challenge, however, 7-d post-challenge pigs receiving EG2 showed ($P < 0.05$) the least circulating TNF- α and IL-6. Overall, better intestinal growth and development as well as lower ETEC counts on the intestinal mucosal and serum pro-inflammatory cytokines suggest that Entegard can maintain gut health and function in piglets commensurate to antibiotics.

Key Words: lysozyme, piglet performance, gut health and function

T229 Effect of microbial phytase on growth performance, plasma phosphorus concentration and tibia mineralization of broilers according to dietary calcium and phosphorus concentrations. M. P. Letourneau Montminy^{*1}, N. Meme², M. Magnin³, and A. Narcy², ¹Agriculture and Agri-Food Canada, Sherbrooke, Qc, Canada, ²INRA UR83, Nouzilly, France, ³BNA Nutrition Animale, Chateau-Gontier, France.

One hundred ninety-two 4-d-old Ross broilers were used to investigate the effect of microbial phytase according to various dietary non-phytate P (NPP) and Ca concentrations on growth performance, plasma P concentration and bone mineralization. Broilers were fed maize-soybean meal diets from 4 to 21 d of age in a $4 \times 2 \times 2$ factorial arrangement with varying concentrations of Ca (0.5, 0.75, 1.0 and 1.25%), NPP (0.2, 0.3 and 0.4%) and microbial phytase (Natuphos; 0 and 500 FTU/kg). The effect of dietary NPP on ADG, ADFI, feed conversion ratio, plasma P concentration, tibia weight, ash weight and ash concentration was dependent on dietary concentration of Ca (NPP \times Ca, $P < 0.001$) and microbial phytase (NPP \times Phyt, $P < 0.001$). Thus, the negative impact of decreasing dietary NPP concentration on these variables was more important in high than in low Ca diets. Also, the positive impact of phytase on growth performance, plasma P concentration and tibia mineralization was increased when NPP was decreased. Additionally, the impact of phytase on growth performance, plasma P, tibia weight, ash weight ($P < 0.001$) and tibia ash concentration ($P < 0.05$) is affected by Ca concentration. These results show that broilers are sensitive to P deficiency which decreases growth performance, plasma P concentration and bone mineralization. However, the impact of P deficiency in animal responses is higher when birds are fed high Ca diets. Furthermore, the response of broilers to phytase was affected by dietary Ca and NPP concentrations, increasing phytase effects when Ca was increased or NPP was decreased. Thus, the more severe the P deficiency (high Ca or low NPP), the more the response of birds to phytase for the studied criteria was important. Optimal dietary concentration of NPP should be established according to dietary Ca and phytase addition. It is also worth noting that these results emphasize the needs to normalize dietary conditions in which P equivalency of phytase are determined.

Key Words: calcium, phytase, broilers

T230 Effect of phytase application on the calcium and phosphorus retention and balance of layers in the last third of the laying cycle. J. Tossenberger¹, L. Babinszky^{*1}, and I. Kühn², ¹Kaposvár University, Kaposvár, Hungary, ²AB Vista, Darmstadt, Germany.

Calcium (Ca) and phosphorus (P) retention (RET) and the amount and percentage of Ca and P release via eggs were studied in the last third of the laying cycle (wk 39 of egg production). A corn-soybean meal based diet was fed to 96 Hy-Line Brown hybrid layers (3 layers/cage) distributed over 4 treatments (Trts). Feeds had identical Ca levels (39.3 g/kg); P levels and phytase activities differed. P content of Trt1 was 2.0 g/kg non-phytate P (NPP) (positive control: PC). P content of Trt2 was 1.0 g/kg NPP (negative control: NC). Trts3 and 4 had same P content as Trt 2 but also had an added 6-phytase, (FINASE EC, from *Trichoderma reesei*), at 125 PPU/kg (Trt3) and 250 PPU/kg (Trt4). Data were analyzed by ANOVA (SAS, 2004). Ca and P RET was lowest in NC group (Ca: 1686 mg/d, P: 78 mg/d). Adding 125 PPU/kg phytase increased Ca and P RET to the PC group levels ($P \geq 0.05$), i.e. 1888 mg/d and 129 mg/d ($P \leq 0.05$). Ca and P RET increased only numerically in Trt4 ($P \geq 0.05$). While NC birds secreted 84.6% of retained Ca (1420 mg/d) and 84.9% of retained P (66 mg/d) with the egg, these values were 98% (1869 mg/d) and only 67.3% (87 mg/day) for Trt3 birds. In consequence Ca balance dropped from +6.9% (NC) to +0.3% (attributable to increased production of birds), while P balance grew from +3.9% to +12.5% (attributable to relatively low egg P level which offsets increased production) (data not shown). The higher phytase dosage (Trt4) did not lead to further Ca and P output in the egg, their amount and ratio being close to those found in Trt3. The Ca balance of PC birds was at an equilibrium, and their P balance showed a surplus of +6.3%. To conclude, adding the tested 6-phytase to low P (1.0 g NPP/kg) layer diets at a level of 125 PPU/kg already improves the Ca and P RET of layers also in the last third of the laying cycle, beside improving their P balance. This should be considered when choosing the P content of the diets and can be used as a tool to reduce P excretion by layers.

Key Words: layer, phosphorus/calcium, retention/balance

T231 Effect of enzyme preparation on nutrient digestibility, digestive enzyme activities and pancreatic enzyme mRNA expression of hens during late laying period. C. Wen^{*1}, L. Wang¹, T. Wang¹, Y. Zhou¹, G. Hou², and Z. Zhou², ¹Nanjing Agricultural University, Nanjing, Jiangsu, China, ²Guangdong VTR Bio-Tech Co., Ltd, Zhuhai, Guangdong, China.

This experiment was conducted to study the effect of enzyme preparation on nutrient digestibility, digestive enzyme activities and pancreatic enzyme mRNA expression of hens during late laying period. Thirty-six 58-wk-old ISA Brown hens were randomly allocated to 2 groups with 6 replicates (3 birds per replicate), and fed corn-soybean meal based diets with or without an enzyme preparation (including phytase, xylanase, cellulase, α -amylase and acid protease) for 4 weeks. The apparent digestibility coefficients of protein, fat and Ca were increased ($P < 0.05$) by enzyme supplementation. The birds fed diets containing enzyme preparation also had a higher ($P < 0.05$) protease activity in jejunal digesta compared with the control group. The pancreatic enzyme activities and their mRNA expression were reduced by enzyme supplementation, but the differences were not significant. The data show that the enzyme preparation is effective in improving nutrient digestibility, but may depress the synthesis of digestive enzymes in pancreas.

Key Words: hen, nutrient digestibility, digestive enzyme activity

T232 Effects of multi-enzyme and *Bacillus subtilis* on sow productivity. T. X. Zhou*, J. S. Yoo, H. J. Kim, Q. W. Meng, J. H. Jung, and I. H. Kim, *Department of Animal Resource and Science, Dankook University, Cheonan, Choongnam, Korea.*

The objective of the experiment was to evaluate the effect of multi-enzyme (Endopower: α -galactosidase, β -glucanase, galactamannanase and xylanase) and *Bacillus subtilis* on sow productivity. A total of 100 sows (Landrace \times Yorkshire) were randomly allotted into 4 dietary treatments and each treatment had 25 sows. The experiment was conducted from July to August 2009. Sows were fed experiment diets from 4 d before farrowing to 21 d of weaning. No crossfostering was done. Dietary treatments were as followed: 1) CON (basal diet), 2) E (basal diet + 1 g/kg Endopower), 3) B (basal diet + 0.4 g/kg *Bacillus subtilis*), and 4) EB (basal diet + 1 g/kg Endopower + 0.4 g/kg *Bacillus subtilis*). Differences among treatments were separated by Duncan's multiple range test. Sows fed multi-enzyme and *Bacillus subtilis* had a higher ($P < 0.05$) ADFI than that of sows in CON treatment (4.99 kg/d vs. 4.71 kg/d). The back fat difference of sows fed multi-enzyme and *Bacillus subtilis* was lower ($P < 0.05$) than that of sows in CON treatment (-5.64 mm vs. -3.68 mm). Sows fed multi-enzyme had a higher ($P < 0.05$) litter size than sows fed *Bacillus subtilis* at birth (11.3 vs. 9.6). Sows in E treatment had a greater ($P < 0.05$) number of litters at weaning than sows in B treatment (9.4 vs. 8.1). No difference was observed on litter performance among treatments. Estrus was not affected by dietary treatments. In conclusion, multi-enzyme and *Bacillus subtilis* increased the ADFI of sows and reduced the backfat loss. Besides, multi-enzyme reduced the mortality of piglets.

Key Words: multi-enzyme, Bioplus 2B, sow

T233 EconomasE decreases sterol carrier protein-2 (SCP2) gene expression levels in breast muscle from 6-week-old chickens. K. M. Brennan*, T. Ao, J. L. Pierce, R. F. Power, and K. A. Dawson, *Center for Animal Nutrigenomics and Applied Animal Nutrition, Alltech Inc., Nicholasville, KY.*

EconomasE (Alltech Inc.) is a proprietary blend of dietary ingredients designed to enhance antioxidant status. Previous studies have shown that supplemental EconomasE increases serum total antioxidant capacity and decreases breast meat drip loss compared with birds fed a control diet (Ao et al., 2009). Based on these data, breast muscle gene expression profiles were compared from birds fed supplemental vitamin E (VE) and EconomasE. Dietary treatments included 1) corn-soy control diet containing 0.3 ppm Se as selenite, but no VE; 2) Diet 1 plus 50 IU VE / kg; 3) Diet 1 plus 100 IU/kg VE; 4) Diet 1 plus 200 g EconomasE /tonne. Birds were house in pens of 22 birds, with 8 replicate pens per treatment. Seven chicks from each of 4 dietary treatments were randomly selected and killed after 42d on treatment. Total RNA was isolated from frozen breast muscle and gene expression was measured using the Affymetrix microarray system. Gene expression data showed that a potential target, sterol carrier protein 2 (SCP2), decreased with both 50 IU and 100 IU of VE (-1.92-fold and -1.59 fold, respectively) and with EconomasE supplementation (-1.67-fold, $P \leq 0.05$). SCP2 plays an important role in the cellular metabolism of lipids, but SCP2 overexpression leads to increased cellular lipid peroxide damage. The decrease in SCP2 mRNA levels was confirmed using real-time PCR. Relative levels of SCP2 mRNA were significantly decreased with both VE treatments (-1.36-fold and -1.34-fold, respectively) and EconomasE supplementation (-1.42-fold, $P \leq 0.05$). These data show that EconomasE mimics VE in breast muscle, potentially through reducing SCP2 levels and peroxidation of cellular membranes.

Key Words: gene expression, vitamin E, broiler