

Performance effects of Allzyme® SSF vs a single microbial phytase in broiler chicks

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Introduction

The manufacturer-recommended level of Allzyme® SSF (Alltech Inc.) in broiler chick diet is 200 g/t of feed. Allzyme® SSF contains a guaranteed minimal phytase activity of 60 units per kg of supplemented feed. This enzyme level is about 8 to 10 times lower than dietary levels recommended for single-microbial phytases.

Objective

To compare the effect of Allzyme® SSF and a single microbial phytase at manufacturer-recommended levels on broiler chick performance.

Materials and methods

Location

Experiment farm, Instituto Internacional de Investigación Animal, Querétaro, México, in 2010.

Animals and experimental design

- 96 single-caged, 10-d-old chicks
- Completely randomized design
- 4 treatment groups
- 24 replicates of 1 chick/trt
- Duration of feeding trial = 25 d

Treatments

- Isoenergetic and isoamino acid diets of sorghum-SBM with varied calcium (Ca) and nonphytate phosphorus (nP) content:
- T1: 0.95% Ca, 0.45% nP, no enzyme
- T2: 0.75% Ca, 0.30% nP, no enzyme
- T3: T2 plus 185 g Allzyme® SSF per ton of feed
- T4: T2 plus 812 g of a single microbial phytase per ton of feed

Measurements

- Initial and final body weights, average daily gain, feed consumption, feed conversion, ash percentage in tibia bone, and mortality.

Data analysis

- ANOVA combined with Tukey's test.

Figure 1. Feed consumption did not differ (P>0.05) between treatments: T1 (control), T2 (low Ca and nP); T3 (T2 + Allzyme® SSF); T4 (T2 + single microbial phytase).

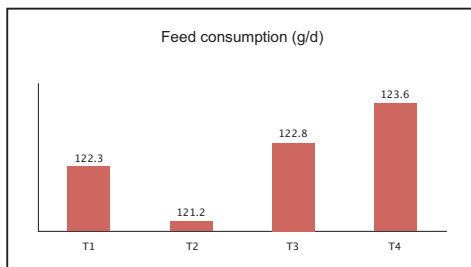


Figure 2. Average daily gain: T1 (control), T2 (low Ca and nP); T3 (T2 + Allzyme® SSF); T4 (T2 + single microbial phytase).

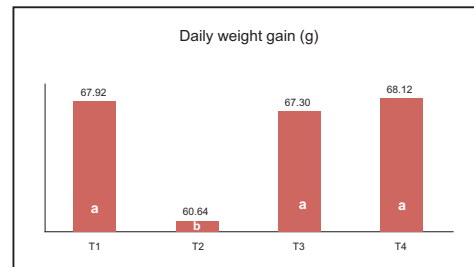


Figure 3. Final body weight: T1 (control), T2 (low Ca and nP); T3 (T2 + Allzyme® SSF); T4 (T2 + single microbial phytase).

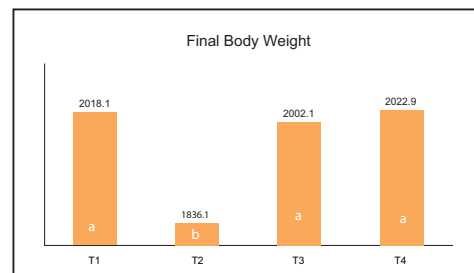


Figure 4. Ash content in tibia: T1 (control), T2 (low Ca and nP); T3 (T2 + Allzyme® SSF); T4 (T2 + single microbial phytase). (a,b differ P<0.05).

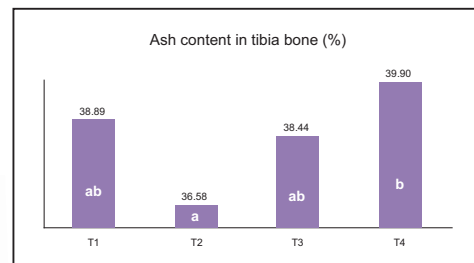


Figure 5. FCR: T1 (control), T2 (low Ca and nP); T3 (T2 + Allzyme® SSF); T4 (T2 + single microbial phytase). (a,b differ P<0.05)

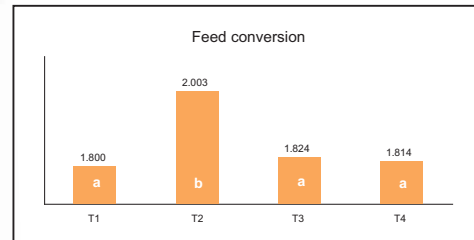


Table 1. Performance of growing broilers fed sorghum-soybean meal diets with varied Ca and nP content and with Allzyme® SSF supplementation.

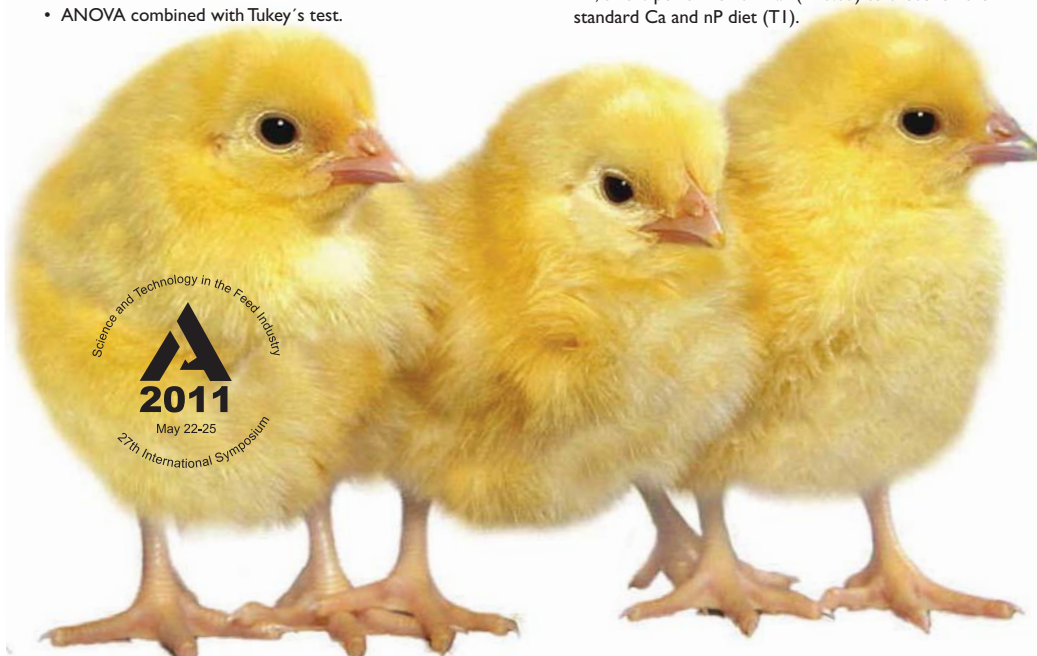
Performance parameters	T1	T2	T3	T4
	0.95/0.45* No enzyme	0.75/0.30* No enzyme	0.75/0.30* AZ SSF	0.75/0.30* Phytase
Initial weight, g	320.2	320.2	319.5	319.8
Average daily gain, g	67.9 ^a	60.6 ^b	67.3 ^a	68.1 ^a
Final weight, g	2018.1 ^a	1836.1 ^b	2002.1 ^a	2022.9 ^a
Feed consumed, g/d/bird	122.3	121.2	122.3	123.6
FCR	1.80 ^a	2.00 ^b	1.82 ^a	1.81 ^a
Tibia ash, %	38.89 ^{ab}	36.58 ^a	38.44 ^{ab}	39.90 ^b

a,b Means differ (P<0.05).

* % Ca / % nP

Results

- Initial weight and feed consumption (Figure 1) did not differ between treatments. No birds died during the experiment.
- In chicks fed the low-Ca, low-nP diet without enzyme (T2), daily gain (Figure 2), final BW (Figure 3), and ash content in tibia (Figure 4) were lower (P<0.05) compared with other treatments, while FCR (Figure 5) was higher (P<0.05) (Table 1).
- Supplementation with Allzyme® SSF or the single microbial phytase corrected the negative performance effects of T2; chicks performed similar (P<0.05) to those fed the standard Ca and nP diet (T1).



Conclusions

- Both Allzyme® SSF (60 PU/kg of feed) and single microbial phytase (600 PU/kg of feed) corrected the negative performance effects associated with the low-Ca, low-nP diet, with both enzyme products estimated at releasing 0.2% Ca and 0.15% nP.
- Allzyme® SSF at 60 PU/kg of feed provided performance improvement equal to that of the single microbial phytase at 600 PU/kg of feed.

Allzyme® SSF increased AMEn of the corn-soy diet and improved performance of broilers

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Introduction

Allzyme® SSF is:

- A naturally fermented enzyme complex by Alltech Inc.
- Produced through solid state fermentation using a selected strain of *Aspergillus niger* containing at least seven enzymes, which work in synergy for a greater release of nutrients from the diet.
- Numerous trials have been conducted to investigate the use of Allzyme® SSF in poultry diets (Wu *et al.*, 2003; 2004; Ribeiro *et al.*, 2003; Godwin *et al.*, 2004; Pierce *et al.*, 2007; 2009).
- Based on the results of these studies, the following recommendation was made for the usage of Allzyme® SSF in broiler diets:
 - Inclusion rate: 200 g/MT
 - Nutrients can be reduced in the corn-soy diet by: 75 Kcal/kg ME, 0.1% available P, 0.1% Ca

Objective

- To investigate the effect of supplementing different levels of Allzyme® SSF on:
 - AMEn of the diet
 - Retention of P and DM
 - Performance of broiler chicks

Materials and Methods

- Animals: 192 broiler chicks, 1 day old
- Replication: 8 replicate cages of 6 chicks per dietary treatment
- Housing: wire-floored starting cages in an environmentally controlled room
- Water & feed: *ad libitum*
- Duration: 21 days
- 1% Celite was included in the diet as a internal marker
- On day 20, fecal samples were collected after 24 h accumulation

Dietary treatments

1. Corn-soybean meal reference diet with 3150 Kcal/kg ME and 0.45% nonphytate P
2. Corn-soybean meal low nutrient diet with 3000 Kcal/kg ME and 0.25% nonphytate P
3. Diet 2 + 200 g Allzyme® SSF/T
4. Diet 2 + 400 g Allzyme® SSF/T

Statistical Analysis

- ANOVA used to identify treatment effects
- LSD used to determine mean differences
- P<0.05 required for significant difference

Results

- Dietary supplementation of Allzyme® SSF with 400 g/MT in corn-soy diet increased DM and P retention compared with chicks fed low nutrient diet or supplemented with 200 g/MT Allzyme® SSF (Figures 1 & 2).
- Compared with chicks fed low nutrient diets, birds fed the diet supplemented with 200 g/T Allzyme® SSF had higher AMEn, tibia ash content and weight gain (P<0.05). No further increases were observed when the dietary enzyme level was increased (Figures 3, 4 & 5).
- Compared with chicks fed low nutrient diets, enzyme supplementation did not increase feed intake significantly (Figure 6).
- Supplementation of a low nutrient corn-soy diet with Allzyme® SSF linearly decreased FCR of chicks compared with chicks fed low nutrient diet alone (Figure 7).

Conclusion

- Supplementing Allzyme® SSF with 200 g/T in corn-soy diet:
 - Increased AMEn of the diet by 84 kcal/kg
 - Increased tibia ash percentage
 - Improved performance of chicks
- No further increase of performance & AMEn was found at 400 g/T.

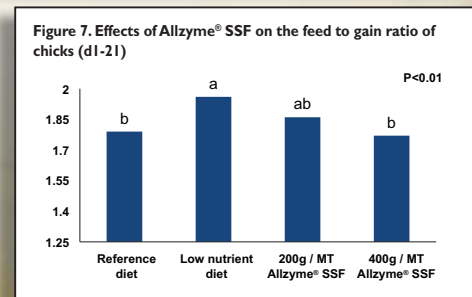
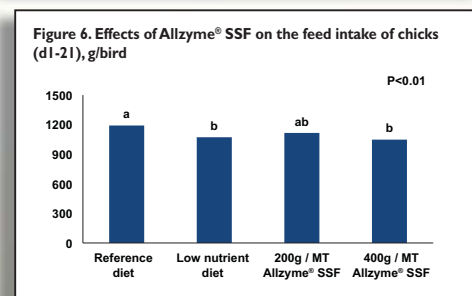
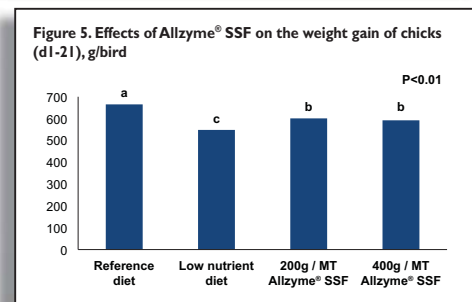
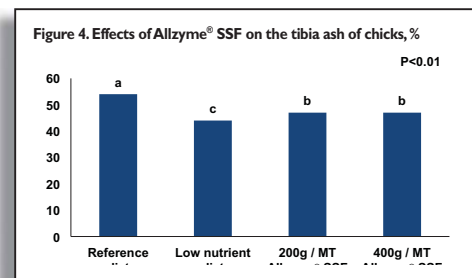
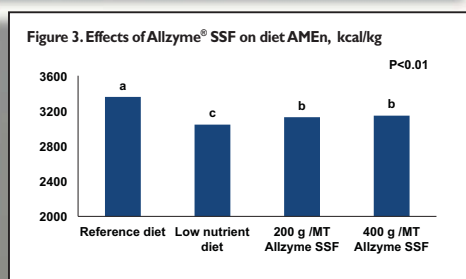
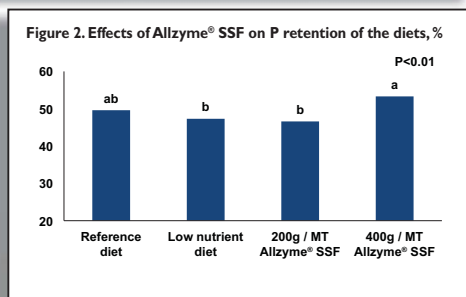
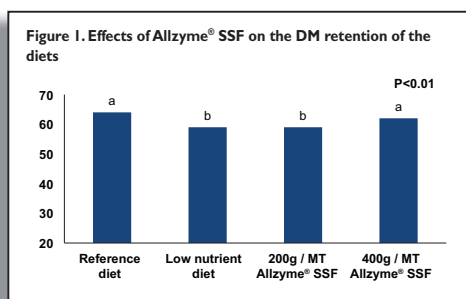


Table 1. Formulas and nutrient composition of the basal diets

Ingredient	Reference diet %	Low nutrient diet %
Corn	53.10	58.40
Soybean meal	36.50	35.80
Vegetable oil	5.20	1.70
Dical. Phosphate	1.79	0.66
Limestone	1.40	1.43
Salt, Iodized	0.45	0.45
DL-methionine	0.21	0.21
Vitamin-mineral premix	0.35	0.35
Celite	1.00	1.00
Nutrient		
ME, kcal/kg	3150	3000
Ca, %	1.00	0.80
Available P, %	0.45	0.25
CP, %	22	22
TSAA, %	0.92	0.92
Lysine, %	1.24	1.23
Na, %	0.21	0.21

Influence of Sel-Plex® on polymeric immunoglobulin receptor (plgR) in reovirus-challenged young broiler chickens

Rose F. Somody¹, F. W. Edens¹, C. M. Ashwell¹, and P. F. Cotter²
¹North Carolina State University, Raleigh, NC, ²Cotter Labs, Arlington, MA

Introduction

Avian reovirus (ARV) can infect joints and tendons, the respiratory tract, and the intestinal tract causing mortality, leg weakness, and poor feed conversions, which in turn cause depressed performance and productivity. Preliminary work from our laboratory demonstrated a relationship between selenium (Se) and expression of polymeric immunoglobulin receptor (plgR) in control and ARV-challenged broiler chickens (Read-Snyder et al., 2010). Previously, we had demonstrated that Sel-Plex-fed ARV-challenged broilers recovered intestinal integrity more rapidly than those fed no Se or sodium selenite (Read-Snyder et al., 2009).

PlgR, a transmembrane glycoprotein, is selectively expressed on the basolateral surface of both mucosal and glandular intestinal epithelial cells and transports dimeric (d) IgA, produced by plasma cells in the lamina propria, and dIgA-coated immune complexes to the apical surface where its secretory component with attached dIgA is cleaved and exocytosed into the lumen of the intestine where bacteria and viruses are neutralized

dIgA secreted by plasma cells attaches to the plgR receptors on the internal surface of intestinal epithelial cells. The plgR transports the dIgA into the cells and across the epithelium to the gut surface and intestinal lumen where the secretory component of plgR, which binds the dIgA, is cleaved and secreted into the lumen as sIgA (Figure 1).

Objective

To examine the influence of double-stranded (ds)RNA ARV-CU98 infection in broiler chickens fed different dietary sources of Se on bile and intestinal plgR expression.

Materials and methods

- Chicks: Feather-sexed Ross 708, 1 day of age
- Chicks housed in heated batteries
- Treatments: Isocaloric (3069 Kcal/kg ME; 16.37% CP; 2.06% fat; 1.57% fiber) Torula yeast diets with: No supplemental Se (<0.02 ppm), Sel-Plex® (Alltech, Inc.; 0.3 ppm Se), or Sodium selenite (0.3 ppm Se).
- ARV-CU98 challenge: At 5 d post-hatch by oral gavage (10¹² pfu/chick)
- Bile, tissue, and lavage: Chicks were euthanized, gall bladder bile collected, and duodenum, jejunum and ileum were dissected and flushed with PBS, and ~500 mg of the liver, duodenum, jejunum and ileum were placed into cold RNALater and frozen (-80 °C) until processed for RNA extraction.
- IgA analysis: Gut lavages processed for ELISA; bile quantified by radial immunodiffusion (Cotter, 2000, 2001)
- RNA extracted and checked for quality control.
- RNA to cDNA - Applied Biosystems High Capacity cDNA Reverse Transcription Kit
- plgR expression analyzed by qRT-PCR using the BioRad icycler iQ Real Time PCR system.
- Statistical analysis: All statistical analyses were conducted using SAS JMP® software. Differences in Ct ratios were analyzed for significance using the Pfaffl method. Data for the Ct ratio were subjected to one way ANOVA. Statements of significance were based on P≤0.05.

Results

Liver plgR gene expression increased daily post challenge with all treatments, and was maximized in Sel-Plex-fed chickens at 7 d post challenge; all other treatments showed maximum expression at 16 d post challenge (Figure 2A).

Fold changes in liver plgR gene expression were maximized at 16 d post challenge in Control-no challenge, Sel-Plex-no challenge and Selenite-Infected, but there was no change in Selenite-no challenge, and a decreasing profile for Sel-Plex-Infected at 16 d (Figure 2B).

Duodenum, jejunum, and ileum plgR gene expression generally showed maximum expression at 7 d post challenge or at 12 d of age (Figures 3A, 4A, 5A).

Intestinal fold changes in plgR gene expression were greatest in Se-fed chickens compared with non supplemented controls; changes in plgR gene expression were transitory showing an increase followed by a decrease (Figures 3B, 4B, 5B).

Conclusion

Sel-Plex® facilitates sIgA secretion by increasing plgR transport thus helping to explain how Se exerts its antiviral effect and helps to maintain the integrity of the intestinal tract through the innate immune system involving both plgR and sIgA.

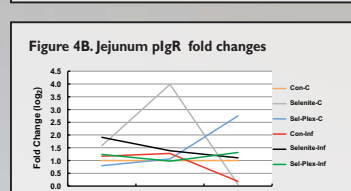
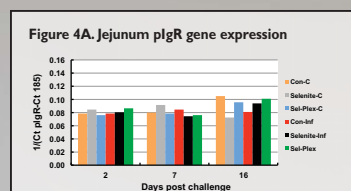
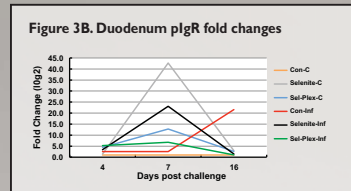
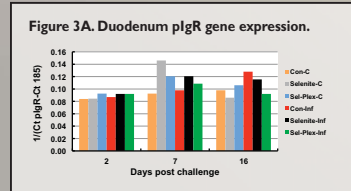
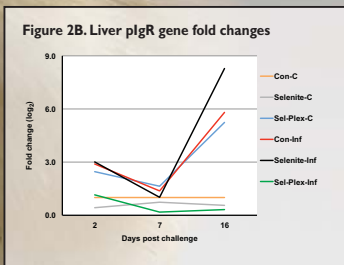
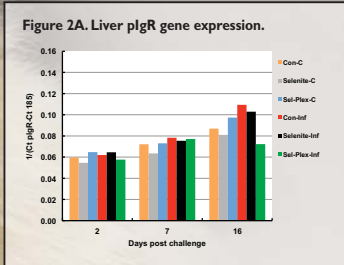
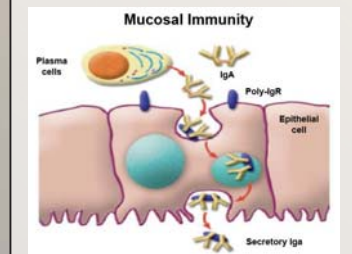
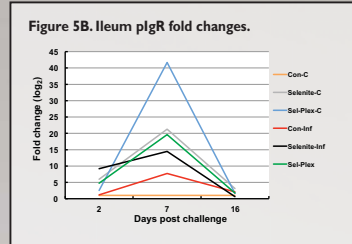
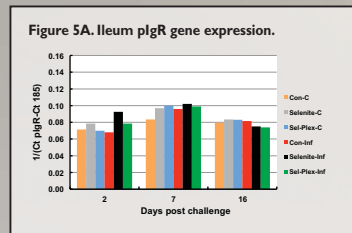


Figure 1. Interaction between plgR and sIgA in the intestinal tract.



The IgA secreted by the plasmatic cells, attaches to the receptors on the internal surface of intestinal epithelial cells. It moves into the cell and then to the gut surface and the intestinal lumen.



Influence of Sel-Plex® on secretory IgA in reovirus-challenged young broiler chickens

Rose F. Somody¹, F. W. Edens¹, C. M. Ashwell¹, and P. F. Cotter²

¹ North Carolina State University, Raleigh, NC, ² Cotter Labs, Arlington, MA

Introduction

At mucosal surfaces, secretory IgA (sIgA) is the primary defense mechanism that guards against invasion of bacteria and viruses. This first line of defense is established through interaction between the mucosal plasma cells, derived from transformed B cells in cooperation with T cells in the lamina propria, and pIgR with its secretory component in the epithelial cells on the intestine.

The dimeric IgA secreted by plasma cells attaches to the pIgR receptors on the internal surface of the intestinal epithelial cells. The pIgR transports the dIgA into the cells and across the epithelium to the gut surface and intestinal lumen where the secretory component of the pIgR, which binds the dIgA, is cleaved and secreted into the lumen as sIgA (Figure 1).

Objective

To examine the influence of double-stranded (ds)RNA ARV-CU98 infection in broiler chickens fed different dietary sources of Se on bile and intestinal sIgA expression.

Materials and methods

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- ARV-CU98 challenge: At 5 d post-hatch by oral gavage ($10^{1.2}$ pfu/chick)
- Bile, tissue, and lavage: Chicks were euthanized, gall bladder bile collected, and duodenum, jejunum and ileum dissected and flushed with PBS, and ~500 mg of liver, duodenum, jejunum and ileum placed into cold RNAlater and frozen (-80 °C) until RNA extraction.
- IgA analysis: Gut lavages processed for ELISA; bile quantified by radial immunodiffusion (Cotter, 2000, 2001).
- RNA extracted and checked for quality control.
- RNA to cDNA - Applied Biosystems High Capacity cDNA Reverse Transcription Kit
- Statistical analysis: All statistical analyses were conducted using SAS JMP® software. Differences in Ct ratios were analyzed for significance using the Pfaffl method. Data for the Ct ratio were subjected to one way ANOVA. Statements of significance were based on $P \leq 0.05$.

Results

- Bile sIgA increased daily post challenge with increases at 7d and 16 d post challenge; sIgA increase was greatest in Sel-Plex-fed birds in both control and infected groups (Figure 2).
- Se supplementation increased sIgA levels in the intestinal tract more quickly than no supplementation; sIgA levels in the intestine were time dependent, but Se source effect was equivocal.
- Duodenum, jejunum and ileum sections of the small intestine contained similar levels of sIgA and showed time-dependent increases in sIgA (Figure 3).
- In all segments, Se-fed birds generally had higher sIgA levels than did non-supplemented chickens.

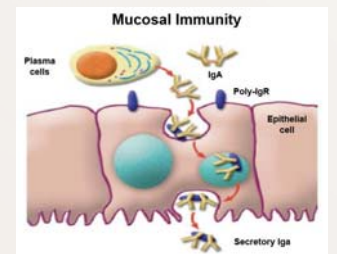
Conclusion

Sel-Plex® facilitates sIgA secretion and thus helps to exert its antiviral effect and to maintain the integrity of the intestinal tract through the innate immune system involving both pIgR and sIgA.

References

- Cotter, P., 2000. Analysis of chicken bile by gel precipitation reactions using a lectin in the place of antibody. *Poult Sci*, 79: 1276-1281.
- Cotter, P., 2001. Quantitative and qualitative heterogeneity of bile in commercial laying hens. Proceedings of the 6th Avian Immunology Research Group K.A. Schat, Ed. Cornell University.

Figure 1. Interaction between pIgR and sIgA in the intestinal tract.



The IgA secreted by the plasma cells attaches to the receptors on the internal surface of intestinal epithelial cells. It moves into the cell and then to the gut surface and the intestinal lumen.

Figure 2. Influence of Se source and ARV infection on bile sIgA.

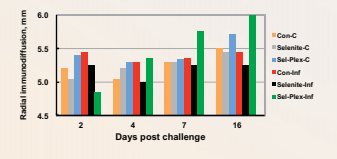
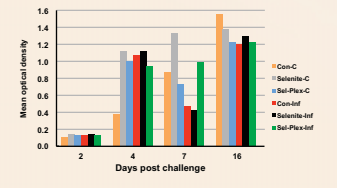


Figure 3. Influence of Se source and ARV infection on intestinal sIgA.



Evaluation of organic copper (Bioplex® Cu) as a copper source for chicks

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Objective

To evaluate the relative bioavailability value (RBV) of Bioplex® Cu compared with reagent-grade $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ in chicks.

Materials and Methods

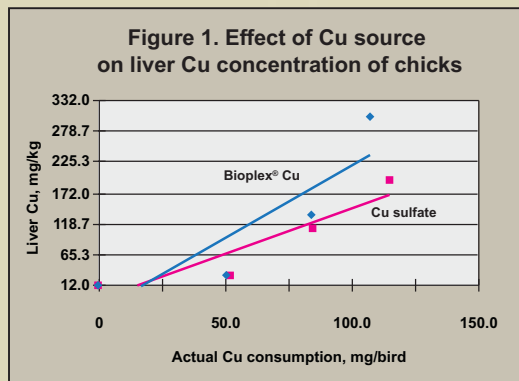
- Animals: 336 broiler chicks, 1 day old
- Replication: 8 replicate cages of 6 chicks per dietary treatment
- Housing: wire-floored starting cages in an environmentally controlled room
- Water & feed: *ad libitum*
- Duration: 14 days
- Feeding: from day 1 to day 5, all chicks fed basal diet (no supplemental Cu). Then, from day 6 to day 14, chicks fed treatment diets
- Sampling: at day 14, two birds per pen with a total of 16 birds per treatment sacrificed for sampling blood and liver
- Bioplex® Cu, a chelated proteinate, containing 10% Cu was supplied by Alltech Inc.

Dietary treatments

- Corn-soybean meal basal diet with no supplementation of Cu (Basal, Table 1)
- Basal + 150 mg Cu as Bioplex® Cu / kg diet
- Basal + 250 mg Cu as Bioplex® Cu / kg diet
- Basal + 350 mg Cu as Bioplex® Cu / kg diet
- Basal + 150 mg Cu as $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ / kg diet
- Basal + 250 mg Cu as $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ / kg diet
- Basal + 350 mg Cu as $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ / kg diet

Statistical Analysis

- ANOVA for completely randomized design
- Multiple linear regression was used to examine linear relationship between dependent and independent variables
- The relative bioavailability value of Cu as Bioplex® Cu was estimated using slope ratio methodology



Results

- Analyzed Cu concentrations (Table 2) in the treatment diets were close to the formulated levels.
- Feed intake and weight gain:** Chicks fed diets with highest Cu levels had lower ($P < 0.05$) intake and weight gain than chicks fed basal diet or basal plus 50 mg Cu as sulfate/kg (Table 3).
- Liver Cu concentration (Table 3):** Since feed intake differed among treatments, the actual consumption of Cu was corrected by feed intake. As dietary Cu increased from both sources, liver Cu linearly increased (Figure 1). Multiple linear regression of liver Cu on actual Cu consumption of Bioplex® Cu and Cu sulfate yielded the following equation: $Y = 2.66 X_1 + 1.93 X_2 - 45.3$ ($r^2 = 0.79$, $P < 0.05$), in which Y represents liver Cu, X_1 represents actual consumption of Cu as Bioplex® Cu and X_2 represents actual consumption of Cu as Cu sulfate. The slope ratio of X_1 vs. X_2 is 138%.

Conclusion

The relative bioavailability of Bioplex® Cu is 138% that of copper sulfate based on the liver Cu concentration.

Table 2. Effects of dietary supplementation of Bioplex® Cu and reagent grade copper sulfate on growth performance of chicks¹.

Cu source	Added Cu (mg/kg)	Feed intake (g/bird)	Weight gain (g/bird)	Gain:feed (g/g)
Basal	0	343 ^a	254 ^{ab}	0.740 ^{ab}
Bioplex® Cu	150	338 ^{ab}	246 ^c	0.728 ^b
	250	337 ^{ab}	244 ^c	0.722 ^{bc}
	350	307 ^c	213 ^d	0.694 ^c
Cu sulfate	150	348 ^a	264 ^a	0.760 ^a
	250	339 ^{ab}	248 ^{bc}	0.733 ^{ab}
	350	329 ^b	234 ^c	0.712 ^{bc}
SEM ²	4.86	5.12	0.01	

¹Data presented are means from 8 groups of six chicks.

²Standard error of the mean.

^{a,b,c,d} Means differ, $P < 0.01$.

Table 3. Effects of dietary supplementation of Bioplex® Cu and reagent grade copper sulfate on liver Cu concentration of chicks¹.

Cu source	Added Cu (mg/kg)	Feed intake (g/bird)	Actual Cu consumption (mg)	Liver Cu concentration (mg/kg DM)
Basal	0	343	0	12 ^a
Bioplex® Cu	150	338	50.7	30 ^a
	250	337	84.3	134 ^d
	350	307	107.5	304 ^d
Cu sulfate	150	348	52.2	29 ^a
	250	339	84.8	111 ^c
	350	329	115.2	195 ^b
SEM ²			9.3	

¹Data presented are means from 8 groups of two chicks for liver Cu concentration.

²Standard error of the mean.

^{a,b,c,d} Means differ, $P < 0.01$.

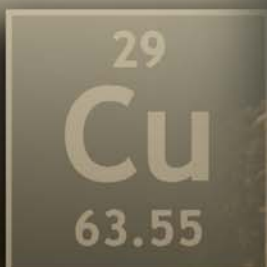
Table 1. Ingredient and composition (as fed basis) of the basal diet¹

Ingredient	% of Diet	Nutrient ¹	Calculated value
Corn	54.53	AMEn, kcal/kg	3144.00
Soybean meal (48%)	36.67	Protein, %	22.00
Corn oil	4.60	Calcium, %	1.00
Salt	0.46	Available P, %	0.45
Limestone	1.33	TSAA, %	0.91
Dicalcium phosphate	1.76	Lysine, %	1.25
Vitamin premix ²	0.25	Sodium, %	0.20
Cu free mineral premix ³	0.25		
DL-Methionine	0.15		

¹Contained 6.8 mg Cu/kg diet by analysis.

²Supplied per kg diet: 3.793 mg vitamin A (retinyl acetate), 0.0882 mg vitamin D₃ (cholecalciferol), 33 mg vitamin E (DL- α -tocopheryl acetate), 0.91 mg vitamin K₃ (2-methyl-1,4-naphthoquinone), 2 mg thiamin, 8 mg riboflavin, 55 mg niacin, 18 mg Ca pantothenate, 5 mg vitamin B₆ (pyridoxines), 0.221 mg biotin, 1 mg folic acid, 478 mg choline, 28 μ g vitamin B₁₂ (cyanocobalamin).

³Supplied per kg diet: 80 mg iron, 60 mg manganese, 60 mg Zn and 0.15 mg selenium.



Performance and gut health of poultry in the post-antibiotic era when feeding a novel yeast cell wall component

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Introduction

The ban on in-feed antibiotics within the European Union has increased interest in other feed supplements to support bird performance. Mannan oligosaccharides have been investigated as to impact on bird production and their mode of action. Actigen™ (Alltech Inc.) is a specific carbohydrate fraction isolated from yeast cell wall oligosaccharides.

Objective

To investigate the effect of Actigen™ on performance and gut development in broilers.

Materials and methods

Animals

- 240 male Ross 308 chicks
- 4 treatment groups
- 12 replicates per treatment of 5 broilers per pen
- Commercial basal diets for each growth phase as per Aviagen guidelines for bird age and strain.
- Duration: 0 — 42 d of age

Treatments

- Control – wheat/soya bean meal-based basal diet
- 200 – Control + Actigen™ at 200 g/t
- 400 – Control + Actigen™ at 400 g/t
- 800 – Control + Actigen™ at 800 g/t

Measurements

- Live weights, feed intakes, and feed conversion ratio (FCR) on a weekly basis.
- On d 42, the gastrointestinal (GI) tract in full was taken from 1 bird per pen with weight closest to the pen average. Weight of duodenum, jejunum and ileum were measured.

Data analysis

ANOVA and Bonferroni post hoc tests to examine the effects of Actigen™ and rate of dietary inclusion on bird performance and GI tract development.

Results

- Actigen™ supplementation was associated with increased live weight compared with the control.
- As in work with Bio-Mos® mannan oligosaccharides (Zhang *et al.* 2005; Chee, 2008), Actigen™ (200 g/t or 800 g/t) in weeks 1 and 2 was associated with improved ($P < 0.05$) FCR compared with the control.
- Actigen™ (200 g/t) was associated with lower ($P < 0.05$) relative duodenum weight compared with the control.
- The performance effects of Actigen™ were age-dependent with younger birds responding more than older birds, possibly because the gut microflora of younger birds is more transient and less mature.
- Dietary supplementation with Actigen™ may increase the rate of gut maturation leading to improved performance whilst concurrently reducing the relative weight of the small intestine.

Conclusions

- Actigen™ had a significant, positive effect on broiler performance.
- Performance effects were age-dependent with younger birds responding more than older birds.

Acknowledgements

- The authors gratefully acknowledge funding from the UK Biotechnology and Biological Sciences Research Council (BBSRC) and Alltech Inc.

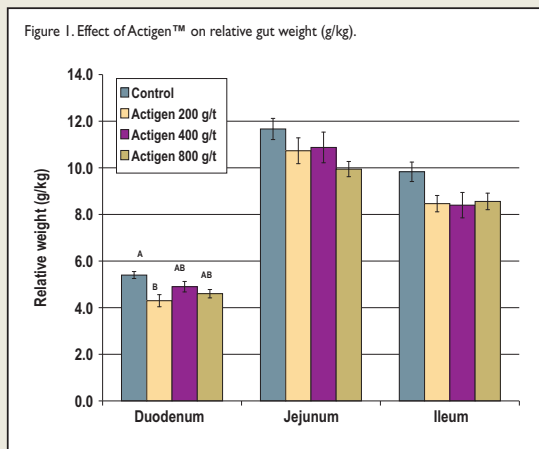
Table 1. FCR in response to Actigen™ treatments.

Week	Rate of Actigen™ in diet				P value
	0 g/t	200 g/t	400 g/t	800 g/t	
1 + 2	2.04	1.58	1.91	1.61	0.004
3 + 4	1.85	1.66	1.65	1.69	0.185
5 + 6	1.69	1.81	1.83	1.89	0.135
1 – 6	1.74	1.73	1.76	1.76	0.923

Table 2. Average body weight (g) in response to Actigen™.

Day	Rate of Actigen™ in diet				P value
	0 g/t	200 g/t	400 g/t	800 g/t	
1	43	44	44	43	0.851
14	217 ^A	305 ^B	256 ^{AB}	291 ^B	0.000
28	1024 ^A	1352 ^B	1243 ^B	1287 ^B	0.001
42	2515 ^A	2847 ^B	2677 ^C	2749 ^{BC}	0.010

^{ABC} Means differ $P < 0.05$



Actigen™ impact on broiler growth and production economics

JUDD CULVER¹ AND ZOE KAY² AND LODE NOLLET², ¹ALLTECH INC., DUNBOYNE, CO., MEATH, IRELAND; ²ALLTECH NETHERLANDS

Introduction & Objectives

Ability of broilers to grow to meet genetic potential is largely dependent on the preservation of good gut health and improvement of disease resistance (immunity). To obtain these effects, Alltech's experience in yeast cell wall technology has led to the development of Actigen™. Validation of the effect of Actigen™ on broiler performance, using scientific trials, is thereby an essential step.

Materials and methods

Experimental design

- Location – Scottish Agricultural College, UK with a top UK feed company
- 2912 1-d-old Ross 308 chicks: male (n=1344), female (n=1568)
- 4 treatment groups of 8 replicates/trt (male 42/pen; female 49/pen)
- Low stocking density: 33 kg/m²
- 3-phase feeding – starter (0 – 10 d), grower (11 – 25 d), finisher (35 – 40 d)
- Duration of feeding trial – 40 d

Treatments

- Negative control – wheat/soy-based (starter crumbled, others pelleted)
- Actigen™ – Control + Actigen™ (800 g/t in starter; 400 g/t in grower; 200 g/t in finisher)

Measurements

- Weight and FCR at 10, 25, 35, and 40 d.
- Litter score and humidity, and scores for hock, foot pad, and lesions at 25 and 40 d.

Data analysis

- ANOVA

Table 1. Broiler weight (kg) in response to Actigen™

Treatment	d 10	d 25	d 36	d 40
Control	0.313	1.240	2.080 ^a	2.521 ^a
Actigen™	0.318	1.263	2.189 ^b	2.657 ^b

^{a,b}Means differ P<0.05

Table 2. Feed conversion in response to Actigen™

Treatment	d 1 – 10	d 10 – 25	d 25 – 35	d 35 – 40	d 1 – 40
Control	1.109	1.443	1.823 ^a	1.894	1.636
Actigen™	1.082	1.444	1.713 ^b	1.821	1.603

^{a,b}Means differ P<0.05

Table 3. Production economics

Extra profit at different broiler prices		Extra cost at different feed prices	
Broiler price (€/kg)	Extra profit* (€)	Feed price (€/kg)	Extra feed cost (€)**
0.7	95.2	0.25	33.7
0.8	108.8	0.275	37.1
0.9	122.4	0.3	40.4

* Due to higher end weight of birds fed Actigen™.

** Due to higher intake and better growth of birds fed Actigen™ (1.09 kg Actigen™/4000 kg feed)

Results

- Performance was good for both treatments. Treatments did not differ (P<0.05) for litter score, humidity, hock or foot pad scores, or mortality.
- Addition of Actigen™ improved average end weight by +136 g and reduced FCR by -3.3 pts (Tables 1 and 2, Figures 1 and 2).

• Profits ranged from 60 – 90 € per 1000 broilers, depending on prices of broilers and feed, using 1.09 kg Actigen™ (Table 3).

Conclusions

Actigen™ supplementation resulted in improved broiler performance and profit.



Figure 1. Effect of Actigen™ on broiler weight (g).

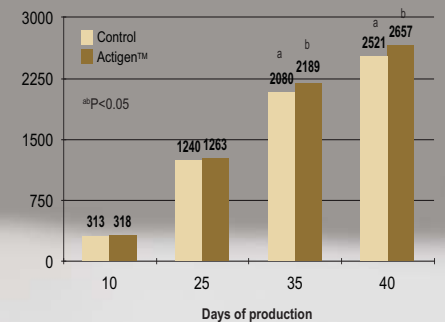
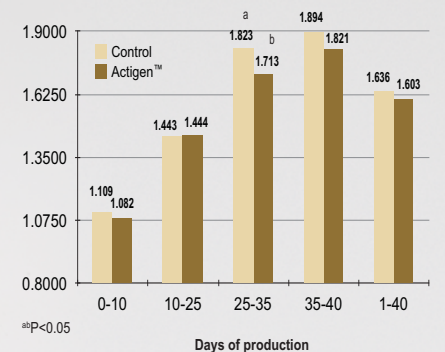


Figure 2. Effect of Actigen™ on FCR.



The effect of Synergen™ on the performance of broilers fed reformulated diets based on two varieties of wheat

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Introduction

Low and high viscosity wheat have different effects on diet digestibility, gut microflora and the intestinal environment of poultry. High gut viscosity can reduce the amount of nutrients absorbed from the diet and thus adversely affect growth and feed efficiency.

Objective

To evaluate the effect of Synergen™ (Alltech Inc.) on broiler performance using treatments containing two different wheat varieties and reformulated to contain a commercial phytase.

Materials and methods

Experimental design

- 1-d-old Ross 308 broilers, mixed sex
- 960 broilers in randomized block design
- 6 treatments (10 replicates of 16 birds each)
- Duration – 37 days
- Diet – Pelleted feed: soya plus wheat
- Wheat varieties (*in vitro* viscosities):
 - V1 – Sahara (cP 1.38 + 0.008)
 - V2 – Altigo (cP 1.30 + 0.003)
- Feeding program – 3 phases (d 1 – 14, d 15 – 28, d 28 – 37)
- All diets reformulated with 0.1% less Ca and 0.1% less available P
- Diets 2, 3, 5 and 6 had an ME reduction of 75 kcal.

Treatments

- T1** Positive control
60% V1 wheat + commercial phytase
- T2** Reformulated
(V1) + commercial phytase
- T3** Reformulated
(V1) + Synergen™ at 200 g/t

- T4** Positive control
60% V2 wheat control + commercial phytase
- T5** Reformulated
(V2) + commercial phytase
- T6** Reformulated
(V2) + Synergen™ at 200 g/t

Measurements

- Technical performance, gut viscosity, and bone minerals.

Data analysis

- ANOVA

Results

- Synergen™ added to the V2 diet improved BW at d 28 compared with the reformulated diet (1161 vs 1236 g; P<0.05). At 37 d, Synergen™ tended to improve BW (+23 g, P>0.05).
- Overall, birds fed Synergen™ had the same rate of growth as positive and reformulated controls.

Table 1. Effect of Synergen™ on broiler body weight.

Bird age	Body weight (g)					
	T1 control (V1)	T1 Reform (V1)	T3 Reform (V1) Synergen™	T4 control (V2)	T5 Reform (V2)	T6 Reform (V2) Synergen™
14 ^d	348 ^a	338 ^{ac}	317 ^d	343 ^a	328 ^b	332 ^{bc}
28 ^d	1198 ^{ab}	1165 ^a	1160 ^a	1240 ^b	1161 ^a	1236 ^b
37 ^d	1973 ^{ab}	1932 ^{ab}	1929 ^a	2003 ^b	1952 ^{ab}	1975 ^{ab}

^{a,b} Means differ (P<0.05).

Table 2. Effect of Synergen™ on broiler average daily gain (ADG).

Period (days)	Average daily gain (g)					
	T1 control (V1)	T1 Reform (V1)	T3 Reform (V1) Synergen™	T4 control (V2)	T5 Reform (V2)	T6 Reform (V2) Synergen™
0 – 14	22.2 ^a	21.5 ^{ac}	20.0 ^d	21.8 ^a	20.8 ^b	21.1 ^{bc}
14 – 28	60.5 ^{ab}	58.6 ^a	60.0 ^a	64.0 ^{ab}	59.6 ^{ab}	64.1 ^{ab}
28 – 37	86.1 ^a	85.2 ^a	85.2 ^a	84.8 ^a	87.9 ^a	82.1 ^a
0 – 37	52.3 ^{ab}	51.2 ^{ab}	51.0 ^a	53.1 ^b	51.8 ^{ab}	52.4 ^{ab}

^{a,b} Means differ (P<0.05).

Table 3. Effect of Synergen™ on broiler feed conversion ratio (FCR).

Period (days)	Average daily gain (g)					
	T1 control (V1)	T1 Reform (V1)	T3 Reform (V1) Synergen™	T4 control (V2)	T5 Reform (V2)	T6 Reform (V2) Synergen™
0 – 14	1.307 ^{ab}	1.300 ^{ab}	1.319 ^a	1.289 ^b	1.320 ^a	1.315 ^{ab}
14 – 28	1.589 ^{ab}	1.616 ^a	1.536 ^{ab}	1.533 ^{ab}	1.604 ^a	1.513 ^b
28 – 37	1.796 ^{ab}	1.81 ^{5ab}	1.825 ^{ab}	1.917 ^a	1.929 ^a	1.915 ^a
0 – 37	1.619 ^a	1.634 ^a	1.614 ^a	1.642 ^a	1.664 ^a	1.629 ^a

^{a,b} Means differ (P<0.05).

- FCR was improved (P<0.05) with Synergen™ compared with the reformulated V2 treatment in the grower and finisher stages.
- Over the entire trial, reformulation improved FCR by 0.02; this effect was corrected by adding Synergen™.
- Synergen™ was effective in reducing gut viscosity when diets were made with V2 wheat, thus preventing impaired digestion.
- Bone breaking strength was unaffected by treatment, demonstrating the ability of Synergen™ to release minerals from diets.

Conclusion

Synergen™ allows for a more flexible approach to feed formulation by reducing nutrient constraints in poultry diet formulation whilst maintaining performance.

Table 4. Effect of Synergen™ on gut viscosity.

Treatment	Viscosity (ηrel)
T1 Control (V1)	3.50 ^a
T2 Reform (V1)	3.71 ^{ab}
T3 Reform (V1) Synergen™	3.94 ^{ab}
T4 Control (V2)	4.77 ^c
T5 Reform (V2)	4.28 ^{bc}
T6 Reform (V2) Synergen™	4.32 ^{bc}
T1 Control (V1)	3.50 ^a

^{a,b} Means differ (P<0.05).

Table 5. Effect of Synergen™ on broiler bone strength and mineralization.

Parameter	Average daily gain (g)					
	T1 control (V1)	T1 Reform (V1)	T3 Reform (V1) Synergen™	T4 control (V2)	T5 Reform (V2)	T6 Reform (V2) Synergen™
Bone weight, g	11.44 ^a	11.98 ^{ab}	12.03 ^{ab}	13.45 ^b	13.07 ^b	12.55
Breaking strength, N	408.12	401.80	404.41	397.33	473.17	400.72
Distance, mm	1.89	2.27	2.17	2.14	1.8	2.35
Mean ash content, %	42.9	42.2	42.3	41.2	42.0	41.7

a, b Means differ (P<0.05).



Effect of Synergen™ in wheat diets on broiler performance

JUDD CULVER, ALLTECH UK, STAMFORD, LINCS., UK; & ZOE KAY, ALLTECH INC., CO. MEATH, IRELAND

Introduction and Objective

Synergen™ is a product of solid state fermentation of *Aspergillus niger* that contains residual enzyme activity. Synergen allows for a more flexible approach to feed formulation through the inclusion of by-products or by reducing nutrient constraints in the diet. The objective of this study was to assess the effect of Synergen™ (Alltech Inc.) in wheat diets on broiler performance.

Materials and methods

Experimental design

- 688 Ross 308 broilers at hatch
- 2 treatment groups with 8 replicates/trt (4 male at 40/pen; 4 female at 46/pen)
- Starter feed in crumble form, all other feeds pelleted

Treatments

- Control – reformulated commercial wheat-soy based ration, containing phytase and non-starch polysaccharide (NSP) enzyme, reduced by 0.1% Ca, 0.1% available P, 0.2 MJ/kg ME
- Synergen – control + Synergen™ (200 g/t of feed)

Measurements

- Live weight, daily gain, feed intake, and FCR at the end of each feeding period
- Mortality and litter score

Results

- Feed intake and weight gain were unaffected by treatment.
- Synergen™ treatment was associated with numerically higher body weight at 40 d and improved FCR.
- Mortality and litter score were unaffected by treatment.

Conclusion

Synergen™ maintained performance of broilers fed a reformulated wheat-soy ration.

Figure 1. Effect of Synergen™ on body weight (kg).

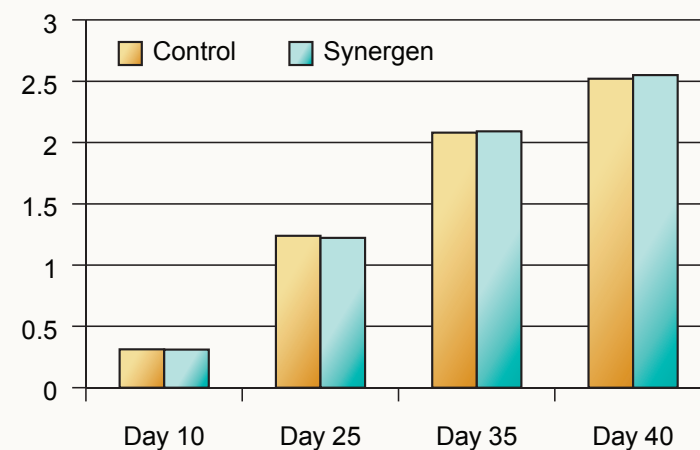


Figure 2. Effect of Synergen™ on FCR.

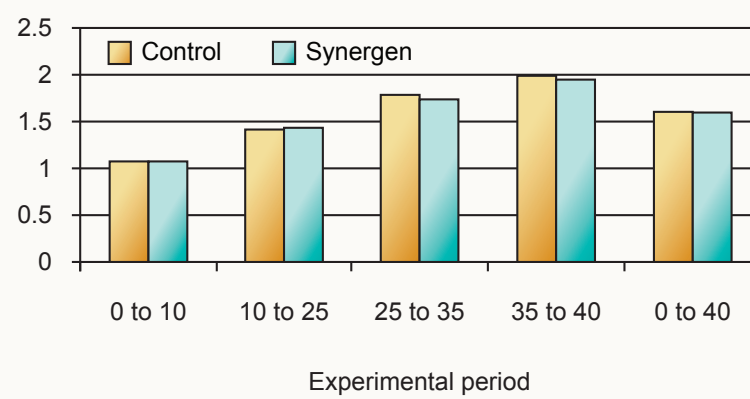


Table 1. Effect of Synergen™ on ADG (g).

Treatment	Experimental period (days)				
	Starter (0 – 10)	Grower (10 – 25)	Finisher (25 – 35)	Withdrawal (35 – 40)	Total (0 – 40)
Control	0.28	0.93	0.84	0.44	2.48
Synergen™	0.27	0.91	0.87	0.46	2.51

Table 2. Effect of Synergen™ on average feed intake.

Treatment	Experimental period (days)				
	Starter (0 – 10)	Grower (10 – 25)	Finisher (25 – 35)	Withdrawal (35 – 40)	Total (0 – 40)
Control	0.30	1.33	1.51	0.90	4.04
Synergen™	0.30	1.33	1.53	0.90	4.05

Bioplex® Zn effects on broiler carcass quality and performance



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Objective

To investigate various levels of organic zinc (Bioplex® Zn, Alltech Inc.) supplementation on performance and carcass quality of female broiler chickens.

Materials and methods

Experimental design

- 3,200 1-d-old female broilers (Ross X Ross 308)
- 16 floor pens (200 birds/pen)
- 4 treatments with 4 replicates (pens)/trt
- All birds fed the basal diet d 1 – 7
- Food and water *ad libitum*
- Duration of feeding trial = 5 wk

Treatments

- Control – Corn-wheat-soy basal diet (no added Zn, background Zn = 30 mg/kg)
- BP 20 – Control + 20 ppm Zn as Bioplex® Zn
- BP 40 – Control + 40 ppm Zn as Bioplex® Zn
- BP 80 – Control + 80 ppm Zn as Bioplex® Zn

Measurements

- Body weight, feed consumption, and mortality were measured throughout the trial.
- At the end of the feeding trial, 3 birds/pen were killed; skin and meat were sampled.
- **Skin thickness.** 1-cm samples from the outer side of thighs and pelvic back region were fixed in 10% neutral formalin (pH 7.4), dehydrated in ethanol, and embedded in paraffin, after which 4-µm-thick sections were stained with hematoxylin and eosin. Thickness of epidermis and dermis was determined under a light microscope (100x) and photographed.
- **Collagen.** Skin and meat samples were analyzed for hydroxyproline (Ignat'eva et al. (2007) with some modifications); total collagen was calculated (Cross et al. 1973).
- **Shear force.** Determined using a texture analyser as per Gwartney et al. (1992).
- **Zinc.** Five-gram ground samples (thigh meat and skin) were ignited, then heated at 600 °C for 2 h, and digested in HCl. Solutions were prepared and Zn determined using atomic absorption spectrometry.

Data analysis

ANOVA using GLM procedure of SAS.



Table 1. Skin layer thickness (µm) of broilers fed various levels of Bioplex® Zn.

Treatments	Back skin		Thigh skin	
	Epidermis	Dermis	Epidermis	Dermis
Control	34.93	234.3	35.52 ^b	160.0 ^b
Bioplex® 20	35.92	249.8	35.83 ^b	258.2 ^a
Bioplex® 40	37.97	304.9	38.77 ^{ab}	250.5 ^a
Bioplex® 80	40.11	252.1	45.82 ^a	277.5 ^a
P value	0.134	0.184	0.033	0.011

^{a,b}Means differ P<0.05.

Table 2. Collagen content in skin and meats of broilers fed various levels of Zn in Bioplex® form.

Treatments	Back skin		Thigh skin	
	Breast meat	Thigh meat	Back skin	Thigh skin
	(mg/g, wet weight)			
Control	1.12	1.19	13.43 ^c	9.28 ^c
Bioplex® 20	1.24	1.25	19.13 ^b	15.71 ^b
Bioplex® 40	1.27	1.49	25.10 ^a	19.01 ^{ab}
Bioplex® 80	1.15	1.42	29.60 ^a	24.88 ^a
P value	0.491	0.281	0.029	0.013

^{a,b,c}Means differ P<0.05

Results

- Performance did not differ between treatments. This was expected as research showed Zn requirement for performance, when supplemented as Bioplex® Zn, is below 20 mg/kg.
- Epidermal and dermal thicknesses of thigh skin increased (P<0.05) with Bioplex® Zn treatments; thickness of back skin remained unchanged.
- Collagen content in back and thigh skin increased (P<0.05) with Bioplex® Zn, however the collagen contents of breast and thigh meats were unaffected.
- Increases in collagen content were greater (P<0.05) in the back and thigh skin using BP 80 compared with BP 20.
- Shear force values and Zn concentrations in skins and meats were unaffected with treatments.

Conclusion

Bioplex® Zn increased skin thickness by increasing skin collagen content, thereby improving carcass quality.

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Performance responses of broilers to Actigen™, a probiotic, or butanoic acid

PER LAUSTSEN, ALLTECH DENMARK AND LODE NOLLET, ALLTECH NETHERLANDS BV

Introduction

Foot pad lesions are an important indicator of bird welfare and as such are included as a measure in the EU broiler welfare regulations. A trial was conducted with a commercial broiler producer to compare performance response and foot pad lesion scores of commercial broilers to three non-antibiotic dietary supplements.

Objective

To compare the performance response of commercial broilers to Actigen™, a specific carbohydrate fraction isolated from yeast cell wall oligosaccharides; Bioacton, a lactic acid bacterial probiotic; or CalSu, calcium butanoic acid.

Materials and methods

Experimental design

- 720 broilers
- 3 treatments with 4 replicates (pens of 60 birds)/trt
- Birds vaccinated on d 5 with MR5.
- Diet – standard pelleted feed to 11 d of age; feed mixed 80:20 with whole wheat to finish

Treatments

- Control: Biacton (d 0 – 11, 500 g/t)
- Actigen™: Actigen™ (d 0 – 11, 800 g/t; d 11 – slaughter, 250 g/t)
- CalSu : CalSu (d 0 – 42)

Measurements

- Body weight, feed consumption, and burning pad index

Results

- Feeding Actigen™ was associated with improved weight gain and improved FCR from 11 – 21 d of age and to 34 d of age compared with other treatments.
- Based on a feed per tonne cost of 250 €, the cost benefit of switching to Actigen™ was 4 €/t.
- Burning pad index (indicating litter quality, foot pad lesions and bird welfare) improved 30% with Actigen™.

Conclusion

Actigen™ improved performance and bird health, compared with Biacton or CalSu.

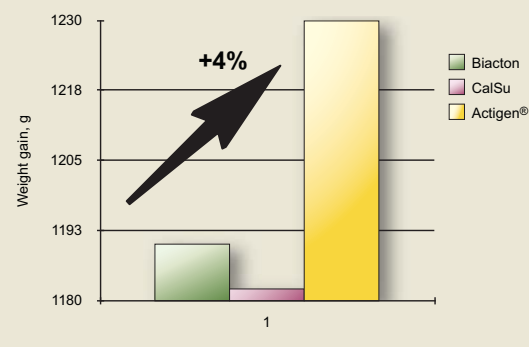
Table 1. Response of commercial broilers to supplementation with Actigen™, Biacton, or CalSu.

Day	Parameter	Biacton	Actigen™	CalSu
d 0 – 11	Growth, g	415	391	392
	FCR	1.03	1.09	1.06
d 11 – 20	Growth, g	443	453	438
	FCR	1.71	1.66	1.72
d 21 – 34	Growth, g	1190	1230	1182
	FCR	1.77	1.76	1.78
d 0 – 34	Growth, g	2048	2074	2012
	FCR	1.61	1.61	1.63

Figure 2. Burning pad index improved 30% with Actigen™ treatment.



Figure 1. Broiler average weight gain (d 14 – 34) in response to supplementation with Actigen™, Biacton, or CalSu.



Effect of Synergen™ in layer diets on egg production and profitability

Piotr Cierpinski, Alltech Poland & Wim Beeks, Alltech Netherlands BV

Objective

To evaluate the effect of Synergen™ (Alltech Inc.) in layer diet on egg production and profitability.

Materials and methods

Experimental design

- 30,000 Lohmann brown laying hens (48-54 wk of age at start)
- 2 treatment groups:
 - Control – 14,800 birds
 - Synergen™ – 15,200 birds
- 6,000 cages/house, 4 – 6 birds/cage
- Mash diet fed *ad libitum*.
- Trial duration = 6 wk
- No vaccinations administered during trial.
- Garlic essence fed to both groups.

Treatments (Tables 1 and 2)

- Control – standard commercial diet
- Synergen™ – reformulated control + Synergen™ at 150 g per tonne

Measurements

- Egg production, feed intake, FCR, and feeding costs.

Results

- Birds fed Synergen™ produced more eggs (Table 3) of similar weight compared with control.
- Although birds fed Synergen™ ate slightly more feed (Table 4), the cost per tonne of feed was less, resulting in lower feeding costs.
- Over the 6-wk trial, profit from birds fed Synergen™ was €176 more (or 0.04 €/egg) compared with the control.

Conclusion

Synergen™ in layer diet can reduce feeding costs and result in more eggs laid per hen with no decline in individual egg weight.

Table 1. Feed formulas.

Raw material	Control %	Synergen™ %
Corn	20.00	20.00
Wheat	30.00	25.00
Triticale	10.00	10.00
Barley	5.10	12.83
Soy oil	1.50	0.50
Canola meal, 35% c.p.	2.00	2.00
Sunflower meal, 29% c.p.	3.00	2.80
Soybean meal, 46% c.p.	17.82	16.70
l-Ca phosphate	1.00	0.60
Limestone	8.60	8.58
NaCl	0.34	0.35
l-Lysine 98%	0.05	0.04
DL-methionine 98%	0.09	0.09
Premix 0.5%	0.50	0.50
Synergen™ (Alltech Inc.)	0.00	0.02
Total	100	100

Table 2. Nutritional value of treatments.

Nutritional value	Control	Synergen™
ME kcal/kg	2727.304	2739.998
ME MJ/kg	11.420	11.474
Crude protein, %	16.801	16.806
Crude fiber, %	3.416	3.554
Lysine, %	0.830	0.834
Methionine, %	0.363	0.372
Methionine & cysteine, %	0.679	0.676
Threonine, %	0.612	0.618
Tryptophan, %	0.198	0.199
Arginine, %	1.055	1.054
Histidine, %	0.395	0.390
Isoleucine, %	0.672	0.685
Leucine, %	1.312	1.297
Phenylalanine & tyrosine, %	1.267	1.252
Valine, %	0.780	0.775
Vitamin B11, %	1.590	1.095
Ca total, %	3.850	3.850
P total, %	0.605	0.615
P available, %	0.350	0.363
Na total, %	0.157	0.159
NaCl, %	0.396	0.400

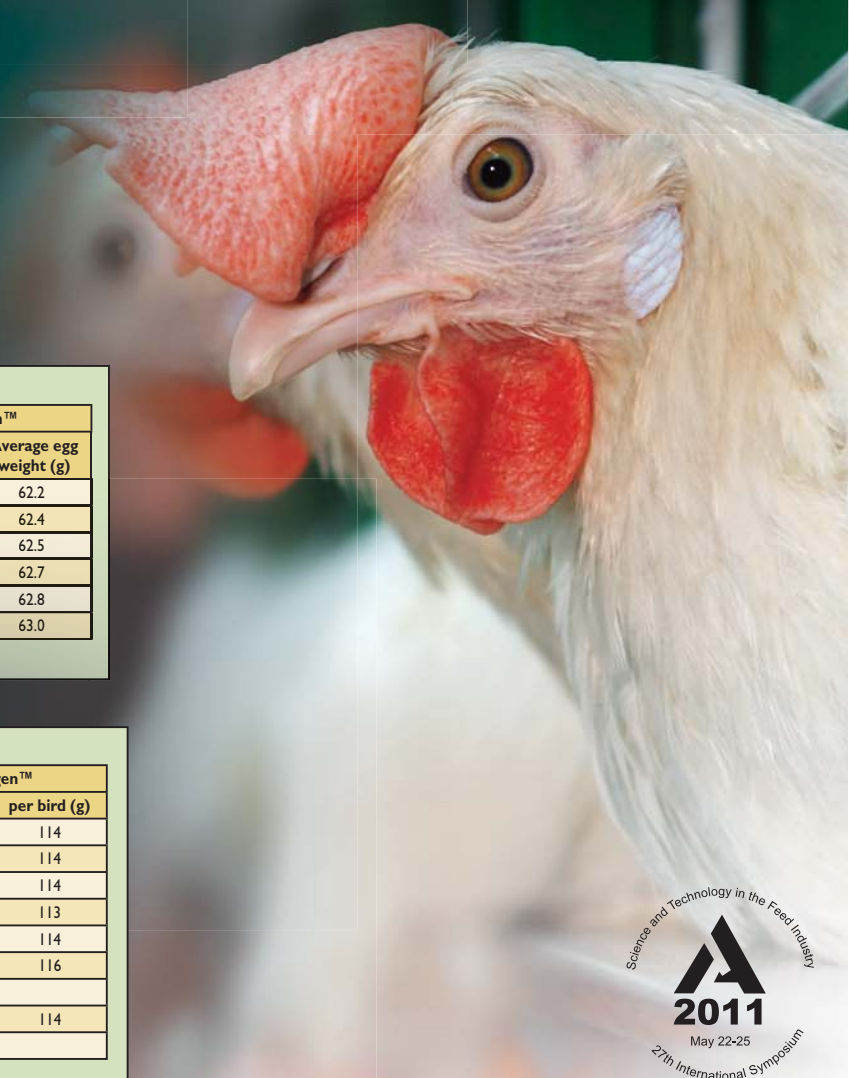
Table 3. Effect of Synergen™ on egg production.

Week	All layers		Control		Synergen™	
	Total number eggs	Average egg weight* (g)	Total number eggs	Average egg weight* (g)	Total number eggs	Average egg weight (g)
48	175,200	62.3	86,400	63.4	88,800	62.2
49	173,400	62.5	85,540	62.5	87,860	62.4
50	174,100	62.6	85,890	62.6	88,210	62.5
51	169,100	62.8	83,430	62.9	85,670	62.7
52	168,900	62.8	83,310	62.9	85,590	62.8
53	169,300	63.0	83,520	63.0	85,780	63.0

* Based on 50 – 100 eggs per day.

Table 4. Effect of Synergen™ on average daily feed intake.

Week	All layers		Control		Synergen™	
	Total (kg)	per bird (g)	Total (kg)	per bird (g)	Total (kg)	per bird (g)
48	23,730	113	11,600	112	12,130	114
49	23,740	113	11,610	112	12,130	114
50	23,940	114	11,800	114	12,140	114
51	23,520	112	11,500	111	12,020	113
52	23,760	113	11,620	112	12,140	114
53	24,110	114	11,710	113	12,400	116
Total	142,800		69,840		72,960	
Mean		113		112		114
Cost (€/t)			174.67		164.16	



Evaluation of the effectiveness of Actigen™ as a growth promoter in broilers

RENATA OLEJNICZAK, ALLTECH POLAND; LODE NOLLET, ALLTECH NETHERLANDS BV

Objective

To evaluate the effect of Actigen™ (Alltech Inc.) on broiler performance.

Materials and methods

Animals

- 3024 Ross 308 broiler chicks at hatch
- 2 treatment groups
- 7 replicates per treatment of 216 broilers per pen

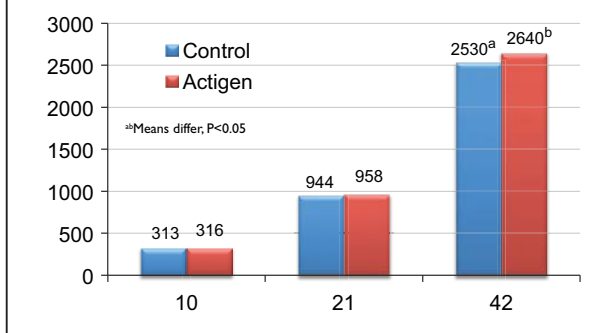
Treatments

- Control – Wheat/soy-based feed in phases: (starter 0-10 d), grower I (11-21 d), grower II (21-30 d), finisher (31-42 d)
- Actigen™ – Control diet plus Actigen™ at 600 g/t in starter and grower I, 400 g/t in grower II, 200 g/t in finisher

Measurements

- Body weight at days 10, 21, and 42; feed consumption; calculated values for average daily gain (ADG) and feed conversion ratio (FCR).

Figure 1. Effect of Actigen™ on broiler live weight at days 10, 21, and 42 days.



Results

- Performance was above average for the breed using either treatment.
- Birds fed Actigen™ were an average 110 g heavier (P<0.05), with an ADG 2.4 higher, compared with controls after 42 d.
- Actigen™ added to broiler diet improved FCR by 0.09 at 1500 g live weight.

Conclusion

Actigen™ supplementation was associated with significantly improved performance in broilers at 42 days and an improvement in FCR (at 1500 g liveweight) of 0.09.



Figure 2. Effect of Actigen™ on broiler ADG after 42 days.

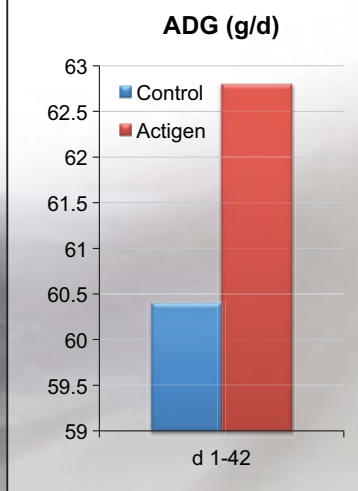
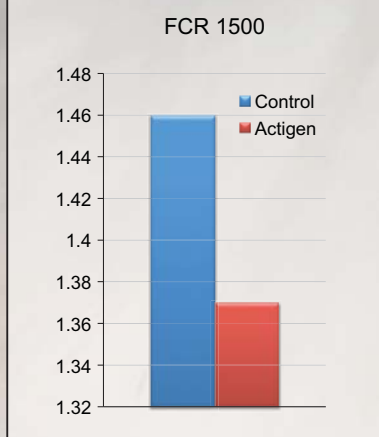


Figure 3. Effect of Actigen™ on feed conversion ratio (FCR) at 1500 g live weight.



Sel-Plex[®] organic selenium improves reproductive performance of turkeys – practical application

SARTOWSKA KATARZYNA¹, JANKOWSKI JAN², KOZŁOWSKI KRZYSZTOF², SPRING PETER³

¹Alltech Inc., Dunboyne, Co. Meath, Ireland; ²University of Warmia and Mazury, Olsztyn, Poland; ³Swiss College of Agriculture, Zollikofen, Switzerland

Objective

The objectives of the trial were to confirm the need for selenium (Se) supplementation in reproducing turkeys and to investigate the effectiveness of organic selenium.

Materials and methods

Experimental design and animals

- Two treatment groups
- 4 breeding houses of 1850 turkey hens and 150 toms each (30 weeks old after Big6)
- 24-week trial

Treatments

- SS – feed supplemented with 0.3 ppm Se as sodium selenite
- SP – feed supplemented with 0.3 ppm Se as Sel-Plex[®] (Alltech Inc.)

Measurements

- Reproductive performance was monitored during the entire production cycle – 24 weeks.
- Basic semen quality was evaluated at the end of the production cycle on 10 toms per treatment

Data analysis

- ANOVA

Results

- Laying performance was numerically improved through the entire production cycle with Sel-Plex[®]; the largest difference was seen in the last 7 weeks of production (Figure 1).
- Number of settable eggs produced per hen was 99.25 vs. 103.24 for sodium selenite and Sel-Plex[®] treatments, respectively (four more eggs from the Sel-Plex[®] group) (Table 1).
- Hatchability did not differ between houses with 94.2% of fertile eggs, 83.8% chicks hatched from fertile eggs (average of 4 houses).
- Calculations show an advantage of over 3 more healthy chicks produced per Sel-Plex[®]-fed hen.
 - 4 more eggs x 0.94 of fertile eggs x 0.84 hatch from fertile eggs = 3.16 more healthy chicks/hen.
- Semen quality evaluation revealed a tendency to higher ejaculate volume in Sel-Plex[®]- fed male treatment (P<0.12) (Table 2).

Conclusion

Supplementation with Sel-Plex[®] improved turkey reproductive performance in both males and females compared with sodium selenite, resulting in four more eggs per hen.

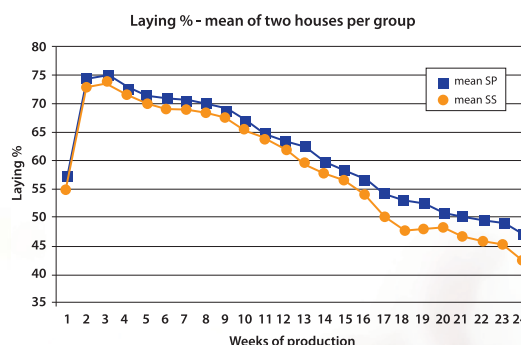
Table 1. Laying performance of hens – mean of two houses per treatment

	Selenite	Sel-Plex [®]	Difference
Overall eggs, number / hen	101.86	105.60	+3.73
Settable eggs, number / hen	99.25	103.24	+3.99

Table 2. Key semen quality parameters

	Selenite	Sel-Plex [®]	SE	P value
Ejaculate volume, mL	0.50	0.63	0.05	0.12
Sperm concentration, x10 ⁹ / mL	8.10	7.30	0.42	0.20
Sperm count, x10 ⁹ /ejaculate	4.00	4.50	0.43	0.42

Figure 1. The effect of selenium supplementation on laying performance.



Sel-Plex[®] organic Se improves health status and early growth performance of turkey progeny

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¹Alltech Biotechnology Center, Dunboyne, Ireland; ²Institute of Animal Reproduction and Food Research, Polish Academy of Sciences, Olsztyn, Poland;

³University of Warmia and Mazury, Olsztyn, Poland

Objective

To evaluate effects of selenium (Se) form in diets fed turkey breeders on the quality and performance of their progeny.

Materials and methods

Experimental design and animals

- 48 cages of 5 birds (24 cages x 5 birds per trt)
- 240 female turkey chicks (BUT Big6) from hens (48 wks of age) fed either inorganic or organic Se (120 chicks each)

Treatments: Hen nutrition

- SS – 0.3 ppm Se from sodium selenite
- SP – 0.3 ppm Se from Sel-Plex[®] (Alltech Inc.)

Chick diet

- All chicks were fed a commercial starter diet containing 0.3 ppm Se from selenite.

Measurements

- Basic performance parameters until 5 wks of age.
- Antioxidant status of egg (TBARS)
- Antioxidant status of 1-d-, and 28-d-old chicks (liver and thigh muscle TBARS, blood GPx and SOD).

Data analysis

- Data were analyzed using ANOVA.

Results

- Progeny from Sel-Plex[®]-fed breeders showed a tendency (at d 35, P=0.12) for higher body weight; which increased with bird age (Figure 1).
- Feed intake, daily weight gain, FCR, and mortality were unaffected by treatment. (data not shown).
- The antioxidant status of eggs and progeny were greater (P<0.01) from hens fed Sel-Plex[®] than from hens fed sodium selenite. (Table 1).

Conclusion

- Chicks from Sel-Plex[®] fed breeders, showed improved antioxidant status and a tendency to improved early performance.
- This suggests that organic selenium – Sel-Plex[®] is a more effective Se source for turkey breeding flocks.



Figure 1. Growth performance of turkey progeny from parent flocks fed different forms of selenium.

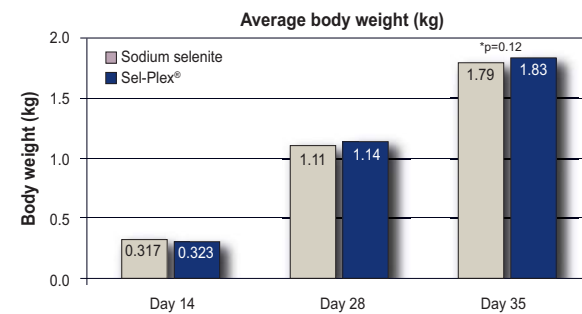


Table 1. Antioxidant status of egg and of progeny from hens fed different forms of Se.

	Selenite	Sel-Plex [®]	SE	P- value
Yolk -TBARS (n=10), nmol/g	53.103	31.125	2.474	0.000*
One-day-old chicks (n=10)				
Liver TBARS, nmol/g	80.188	79.370	11.010	0.959
Muscle TBARS, nmol/g	65.907	38.418	5.048	0.001*
Blood GPx, U/mL	5.920	7.537	0.306	0.002*
Blood SOD, U/mL	79.230	89.300	2.947	0.027*
28-day-old chicks (n=7)				
Liver TBARS, nmol/g	23.357	26.579	3.556	0.534
Muscle TBARS, nmol/g	69.034	64.507	9.960	0.753
Blood GPx, U/mL	6.631	6.329	0.581	0.719
Blood SOD, U/mL	87.293	100.021	3.799	0.035*



Distillers dried grains with solubles and Allzyme® SSF: Effects on performance and egg quality of brown egg layers through 60 weeks of egg production

P. Rossi,* A. J. Pescatore, A. H. Cantor, J. L. Pierce, T. Ao, L. M. Macalintal, M. J. Ford, W. D. King, and H. D. Gillespie
Alltech/University of Kentucky Nutrition Research Alliance, Lexington, KY

Introduction

The poultry feed industry has greatly increased its use of exogenous enzymes. Adding the appropriate exogenous enzymes to the feed can improve utilization of nutrients from the feed, thereby decreasing feed cost, improving bird performance, and decreasing the environmental impact of manure application to land. The availability of DDGS for poultry diets continues to increase. There is also a desire by poultry producers to increase the inclusion levels of DDGS into poultry diets.

Objective

To evaluate effects of feeding diets containing 15 or 23% distillers dried grains with solubles (DDGS) with or without a naturally occurring enzyme complex (Allzyme® SSF, Alltech, Inc.) on egg production parameters in brown shell hens during 60 wk of production.

Material and methods

Animals:

- 420 hens Hy-Line Brown (17 weeks of age)
- 7 replicates/treatment (12 hens/rep)
- Housed 2 hens/cage, 512 cm²/hen

Egg quality:

- 60 weeks of production
- 6 eggs/replicate every 4 weeks
- Haugh Unit – Quantum Chromodynamics Super System (TSS QCD System).
- Yolk color - Colorflex colorimeter (HunterLab).

Statistical Analysis:

- ANOVA (SAS); means separated by Fisher's protected LSD
- P ≤ 0.05 required for significance

Dietary treatments

1. Positive control (corn-soybean meal)
2. 15% DDGS
3. 15% DDGS + Allzyme® SSF (150 g/ton)
4. 23% DDGS
5. 23% DDGS + Allzyme® SSF (150 g/ton)

Observations

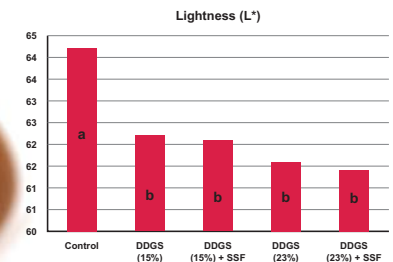
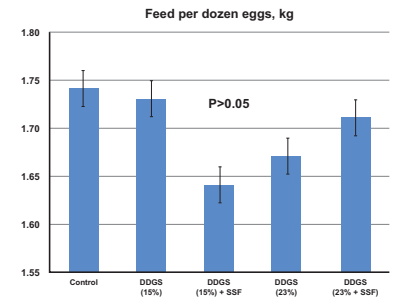
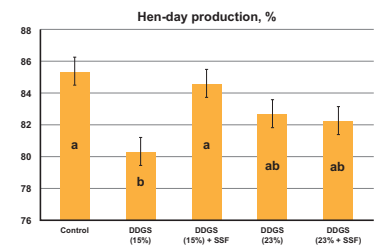
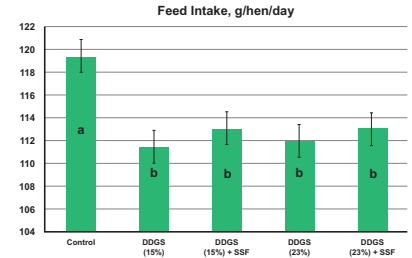
- Feed intake decreased (P<0.01) with DDGS.
- Adding Allzyme® SSF to 15% DDGS diets increased HDP.
- Adding Allzyme® SSF to DDGS diets partially alleviated the reduction in shell weight, percent shell, specific gravity and shell breaking strength.
- Haugh unit values were increased by DDGS.
- Yolk color was impacted by the addition of DDGS to the diet.
- Hens fed 15 or 23% DDGS, with or without Allzyme® SSF, had decreased yolk lightness (L*), while feeding 23% DDGS increased yolk redness (a*) and yellowness (b*) values, compared to 15% DDGS and control diet.

Composition of diets

Ingredient, %	Positive Control	15% DDGS	15% DDGS + SSF	23% DDGS	23% DDGS + SSF
Corn	56.51	51.10	51.10	46.29	46.29
Soybean meal 48% CP	28.00	20.50	20.50	16.90	16.90
DDGS	0.00	15.00	15.00	23.00	23.00
Corn oil	3.30	1.85	1.85	2.20	2.20
Oyster shell	3.00	3.00	3.00	3.00	3.00
Limestone (38% Ca)	7.30	7.60	7.60	7.62	7.62
Dicalcium phosphate	1.00	0.00	0.00	0.00	0.00
Salt, iodized	0.47	0.35	0.35	0.30	0.30
Vitamin-mineral mix	0.25	0.25	0.25	0.25	0.25
DL-Methionine	0.17	0.15	0.15	0.14	0.14
Lysine HCl	0.00	0.20	0.20	0.30	0.30
Total	100.00	100.00	100.00	100.00	100.00
Calculated analysis					
ME, Mcal/kg	2.88	2.80	2.80	2.80	2.80
CP, %	18.48	18.48	18.48	18.50	18.50
Ca, %	4.22	4.10	4.10	4.10	4.10
P, avail., %	0.29	0.17	0.17	0.20	0.20
TSAA, %	0.76	0.76	0.76	0.76	0.76

Nutrient values for DDGS: 2.71 Mcal/kg ME, 26.1% CP, 0.03% Ca, 0.55% Avail. P, 0.97% TSAA

Treatments	Shell breaking strength (N)	Specific gravity (g/cm ³)	Shell weight (g)	Percent Shell	Haugh Units
1. Control	3.1 ^a	1.080 ^a	5.8 ^a	9.1 ^a	75.0 ^c
2. DDGS (15%)	2.8 ^d	1.076 ^c	5.2 ^c	8.1 ^d	80.4 ^a
3. DDGS (15%) + SSF	3.0 ^b	1.078 ^b	5.5 ^b	8.6 ^b	78.1 ^{ab}
4. DDGS (23%)	2.8 ^{cd}	1.076 ^c	5.2 ^c	8.2 ^{cd}	77.9 ^b
5. DDGS (23%) + SSF	2.9 ^{bc}	1.077 ^{bc}	5.4 ^b	8.5 ^{bc}	79.6 ^{ab}
P value	<0.01	<0.01	<0.01	<0.01	<0.01
CV, %	16.2	0.5	11.9	11.1	11.2



Conclusions

These results indicate that negative effect of feeding high levels of distillers dried grain with solubles can be partially overcome by including Allzyme® SSF in the diets.

Distillers dried grains with solubles in post-peak diets for laying hens: Response to Allzyme® SSF

P. Rossi,* A. J. Pescatore, A. H. Cantor, J. L. Pierce, T. Ao, L. M. Macalintal, M. J. Ford, W. D. King, and H. D. Gillespie
Alltech/University of Kentucky Nutrition Research Alliance, Lexington, KY

Introduction

DDGS has become more available and economical as a feed ingredient for poultry diets. However, previous research showed nutritional limitations for its use at high levels throughout an entire production cycle. Exogenous enzymes have been shown to increase nutritional value of poultry diets and may permit higher inclusion rates of DDGS. The present study examined the use of high levels of DDGS with or without Allzyme® SSF in post-peak laying hen diets.

Objective

To evaluate effects of feeding diets containing 15 or 23% distillers dried grains with solubles (DDGS) with or without a naturally occurring enzyme complex (Allzyme® SSF, Alltech, Inc.) on performance and egg quality of laying hens during weeks 44 to 60 of the production cycle.

Material and methods

Animals:

- Exp. 1: 420 hens Hy-Line W36 (62 to 78 weeks of age)
- Exp. 2: 420 hens Hy-Line Brown (62 to 78 weeks of age)
- 7 replicates/treatment (12 hens/ rep)
- Housed 2 hens/cage, 512 cm²/hen

Egg quality:

- 16 weeks of production
- 6 eggs/replicate every 4 weeks
- Haugh Unit – Quantum Chromodynamics Super System (TSS QCD System)
- Yolk color - Colorflex colorimeter (HunterLab)

Statistical Analysis:

- ANOVA (SAS); means separated by Fisher's protected LSD
- P<0.05 required for significance

Dietary treatments

- 1- Positive control (corn-soybean meal)
- 2- 15% DDGS
- 3- 15% DDGS + Allzyme® SSF (150 g/ton)
- 4- 23% DDGS
- 5- 23% DDGS + Allzyme® SSF (150 g/ton)

Observations

- Compared with the control diet, feeding 15 or 23% DDGS with or without Allzyme® SSF reduced percent shell for brown hens (Exp. 2).
- Specific gravity was reduced by 15% DDGS without Allzyme® SSF and by 23% DDGS with or without Allzyme® SSF (Exp. 2).
- In Experiment 2 addition of Allzyme® SSF to DDGS diets improved shell quality to the level of the control diet.
- Eggs from hens fed 15 and 23% DDGS with or without Allzyme® SSF had lower yolk lightness (L*) compared with eggs from the control treatment.
- Eggs from hens fed 23% DDGS had higher yolk redness (a*) and yellowness (b*) values compared with those from the 15% DDGS and control treatments, indicating a darker yolk color.

Conclusions

The current studies suggest that DDGS can be included in the post-peak production diets up to 23% with minimal effects on performance or egg quality. In brown hens the addition of Allzyme® SSF corrected shell problems due to DDGS.

Results

Production performance parameters

Treatments	Feed Intake (g/hen/day)	Hen-day production (%)	Feed per dozen eggs (kg)	Feed Intake (g/hen/day)	Hen-day production (%)	Feed per dozen eggs (kg)
1. Control	110	73.6	1.79	114	69.5	1.98
2. DDGS (15%)	110	73.1	1.81	115	70.5	1.96
3. DDGS (15%) + SSF	110	75.5	1.74	113	71.4	1.91
4. DDGS (23%)	110	75.9	1.74	115	70.7	1.97
5. DDGS (23%) + SSF	109	74.2	1.77	116	71.1	1.96
P value	ns	ns	ns	ns	ns	ns
CV, %	3.9	6.7	7.5	4.7	9.2	10.3

Composition of diets

Ingredient, %	Positive Control	15% DDGS	15% DDGS + SSF	23% DDGS	23% DDGS + SSF
Corn	56.51	51.10	51.10	46.29	46.29
Soybean meal, 48% CP	28.00	20.50	20.50	16.90	16.90
DDGS	0.00	15.00	15.00	23.00	23.00
Corn oil	3.30	1.85	1.85	2.20	2.20
Oyster shell	3.00	3.00	3.00	3.00	3.00
Limestone (38% Ca)	7.30	7.60	7.60	7.62	7.62
Dicalcium phosphate	1.00	0.00	0.00	0.00	0.00
Salt, iodized	0.47	0.35	0.35	0.30	0.30
Vitamin-Mineral mix	0.25	0.25	0.25	0.25	0.25
DL-Methionine	0.17	0.15	0.15	0.14	0.14
Lysine HCl	0.00	0.20	0.20	0.30	0.30
Total	100.00	100.00	100.00	100.00	100.00
Calculated analysis					
ME, Mcal/kg	2.88	2.80	2.80	2.80	2.80
CP, %	18.48	18.48	18.48	18.50	18.50
Ca, %	4.22	4.10	4.10	4.10	4.10
P avail., %	0.29	0.17	0.17	0.20	0.20
TSAA, %	0.76	0.76	0.76	0.76	0.76

Nutrient values for DDGS: 2.71 Mcal/kg ME, 26.1% CP, 0.03% Ca, 0.55% Avail. P, 0.97% TSAA

Experiment 1. Egg quality results in white laying hens

Treatments	Egg weight, g	Shell weight, g	Percent shell	Yolk weight, g	Percent yolk	Albumen weight, g	Percent albumen	Haugh Unit	Shell breaking strength, N	Specific gravity g/cm ³	L*	a*	b*
1. Control	66.9	5.9	8.8	19.2	28.7	41.8	62.4	73.1	3.01	1.078	65.4 ^a	11.9 ^c	65.9 ^c
2. DDGS (15%)	66.9	5.8	8.7	19.4	29.0	41.7	62.3	75.0	2.99	1.078	63.9 ^b	14.2 ^b	68.1 ^b
3. DDGS (15%) + SSF	67.5	5.8	8.6	19.4	28.7	42.4	62.8	74.9	2.88	1.077	64.0 ^b	14.3 ^b	68.1 ^b
4. DDGS (23%)	67.5	5.9	8.7	19.5	28.9	42.2	62.4	73.2	2.90	1.076	63.3 ^c	15.4 ^a	69.7 ^a
5. DDGS (23%) + SSF	67.3	5.9	8.7	19.4	28.8	42.0	62.4	73.7	2.94	1.077	62.9 ^c	15.2 ^a	68.6 ^{ab}
P value	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	<0.01	<0.01	<0.01
CV, %	3.2	4.6	4.3	5.2	4.6	4.3	2.2	4.8	11.2	0.3	1.9	10.0	4.6

Experiment 2. Egg quality results in brown laying hens

Treatments	Egg weight, g	Shell weight, g	Percent shell	Yolk weight, g	Percent yolk	Albumen weight, g	Percent albumen	Haugh Unit	Shell breaking strength, N	Specific gravity g/cm ³	L*	a*	b*
1. Control	68.0	5.9	8.7 ^a	17.8	26.2	44.3	65.1	71.5	2.87	1.077 ^a	64.8 ^a	12.4 ^d	64.6 ^c
2. DDGS (15%)	67.3	5.5	8.2 ^b	17.3	25.7	44.5	66.1	73.4	2.66	1.075 ^b	63.7 ^b	14.0 ^c	66.0 ^{bc}
3. DDGS (15%) + SSF	67.0	5.6	8.4 ^{ab}	17.1	25.5	44.3	66.1	72.0	2.74	1.076 ^{ab}	63.7 ^b	14.3 ^{bc}	67.0 ^{ab}
4. DDGS (23%)	67.6	5.5	8.2 ^b	17.5	25.9	44.6	66.0	70.6	2.68	1.075 ^b	63.0 ^c	15.2 ^a	66.9 ^{ab}
5. DDGS (23%) + SSF	67.1	5.6	8.4 ^{ab}	17.3	25.8	44.1	65.8	72.1	2.82	1.075 ^b	63.3 ^{bc}	15.2 ^{ab}	68.1 ^a
P value	ns	ns	<0.05	ns	ns	ns	ns	ns	ns	<0.01	<0.01	<0.01	<0.01
CV, %	3.61	10.05	9.42	6.47	5.92	4.76	2.57	7.5	15.71	0.37	1.7	12.77	5.55



Comparative effects on broiler performance and litter quality: Actigen™ and BMD

GREG F. MATHIS, SOUTHERN POULTRY RESEARCH, INC.; ATHENS, GA, USA

Introduction and Objectives

Finding non-antibiotic means of promoting gut health in intensively-reared birds is an active research area as the industry moves away from antibiotic use. This experiment compared performance responses to Actigen™, a yeast cell wall carbohydrate derivative (Alltech Inc) and bacitracin methylene disalicylate (BMD) and associated impact on litter quality.

Birds, diets and housing

- 1-day-old mixed sex Cobb MX X Cobb 500 chicks obtained from Pilgrim's Pride hatchery, Commerce, GA
- 1950 birds, 50 birds in each of 39 floor pens
- 25 birds/feeder, 0.93 ft/bird; 24-h lighting, fans and side curtains for ventilation
- Corn-soy pelleted basal diet with coccidiostat in starter and grower
- 3 dietary treatments replicated in 13 blocks, randomized within blocks of 3 pens:
 - Control
 - Control + Actigen™ (0.8 kg/T d 1-7, 0.4 kg/T d 7-21, 0.2 kg/T d 21-42)
 - Control + BMD (50 g/T d 1-21, 25 g/T d 21-42)

Measurements and analysis

- Bird weights by pen were recorded on days 0, 21, and 35. Bird weights by pen by sex were recorded on day 42.
- Non-consumed feed was weighed days 21, 35 and 42.
- Litter condition was graded day 42 on a 0 (dry, friable) to 5 (wet, soggy throughout) scale. A score of 2 indicated mostly acceptable litter quality but with some areas of wet shavings or capped material; while a score of 3 denoted poor quality litter and a large proportion of wet area and capping.
- Data were subjected to ANOVA with means separated by LSD.

Results

- Live weights at day 21 were similar with efficiency improved by Actigen™ and BMD (P<0.05) (Table 1). At days 35 and 42 both live weights and FCR values were improved (P<0.05) for treated birds (Tables 1 and 2).
- Litter quality at day 42 was significantly better in pens housing birds given Actigen™ or BMD while control pen litter was in the poor quality range (Figure 1).

Table 1. Live weight and FCR at days 21 and 35 (^{a,b}Means differ, P<0.05).

Treatment	Day 21		Day 35	
	FCR	LW (kg)	FCR	LW (kg)
Control	1.336 ^a	0.735 ^a	1.727 ^a	1.566 ^b
Actigen™	1.313 ^b	0.737 ^a	1.681 ^b	1.613 ^a
BMD	1.317 ^b	0.740 ^a	1.686 ^b	1.611 ^a

Table 2. Live weight and FCR at day 42 (^{a,b}Means differ, P<0.05).

Treatment	FCR	Male	Female	Avg.
Control	1.814 ^a	2.211 ^a	1.920 ^b	2.081 ^b
Actigen™	1.767 ^b	2.254 ^a	2.013 ^a	2.134 ^a
BMD	1.782 ^b	2.246 ^a	2.003 ^a	2.124 ^a

Conclusions

Actigen™ and BMD similarly improved broiler performance and efficiency compared to the negative control. Improved litter quality was associated with this response.



Figure 1. Effects of Actigen® and BMD on litter quality score (0-5) at day 42.



Mineral content of excreta and eggs from laying hens fed Bioplex[®] or inorganic minerals



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Introduction

Low levels of added trace minerals (Cu, Mn and Fe) in Bioplex[®] (Alltech Inc.) form have been shown to support performance in layers comparable to that of traditional levels of inorganic trace minerals. Mineral excretion is reduced and a reduction in pollution potential is seen; while egg levels of Cu, Mn and Zn were unaffected. Further work demonstrated that low levels of supplemental trace minerals in this form reduced egg mottling.

Objective

To compare the mineral content of excreta and eggs from laying hens fed Bioplex[®] or inorganic minerals.

Materials and methods

Experimental design

- 72 36-wk-old Lohmann layers
- 2 treatment groups
- Diets were fed *ad libitum* for 46 days.

Treatments

- Inorganic – Traditional levels of inorganic mineral supplements (Mn 100 ppm; Zn 60 ppm; Fe 25 ppm; Cu 5 ppm)
- Bioplex[®] – Bioplex[®] mineral supplements (Mn 12.6 ppm; Zn 14 ppm; Fe 3.6 ppm; Cu 0.6 ppm)

Measurements

- All eggs were collected for the last 2 d; egg weight, shell deformation, mottling, and albumen height were measured.
- A composite of 6 eggs from d 46 was collected and shells, albumen and yolk were separated and analyzed.
- A 24-h excreta sample from each treatment group was assayed.

Table 1. Supplemental trace mineral levels in Bioplex[®] Cu, Mn Fe and Zn and Sel-Plex[®] Se-treated diets.

Mineral	Inorganic	Bioplex [®] /Sel-Plex [®]
Manganese	100	12.6
Zinc	60	14
Iron	25	3.6
Copper	5	0.6
Selenium	0.3	0.3

Table 2. Effect of Bioplex[®] minerals on egg quality and production.

Mineral	Egg weight (g)	Egg shell deformation (µm)	Albumen height (mm)	Egg production (%)
Bioplex [®]	64.9	22.2	6.7	91.9
Inorganic	62.8	21.7	6.9	93.7
SD	6.0	3.1	0.8	9.0
Significance	NS	NS	NS	NS

Table 3. Effect of Bioplex[®] minerals on egg weight, deformation and mottling score.

Mineral	Mottling score (0 – 5)	Mottled (n)	Non-mottled (n)	Egg weight M (g)	Egg weight NM (g)	Egg shell deformation M (µm)	Egg shell deformation NM (µm)
Bioplex [®]	0.90	16	15	64.8	64.3	21.3	20.4
Inorganic	1.29	18	13	65.2	65.0	24.1	21.3
SD	1.2			4.4	4.3	6.4	1.8
Significance	NS			NS	NS	NS	NS

Results

- Egg production, egg weight, egg shell deformation, mottling score and albumen height were not affected by treatment (P>0.05).
- Mottling was positively correlated with shell deformation.
- Mineral content of eggs did not differ between treatments, although eggs from Bioplex[®]-fed hens had a quarter of the level of Zn compared with hens eggs fed inorganic supplemental minerals.
- Excreta from hens fed Bioplex[®] supplemented diets compared with that from hens fed inorganic mineral diets had less Zn and Mn (P<0.01). Excreta Cu levels did not differ between diets (P>0.05).

Conclusions

- Supplementing layer diet with lower levels of Bioplex[®] trace minerals is associated with lower concentrations of Zn and Mn in excreta and does not adversely affect egg quality or production.
- In future studies, it would be interesting to study the effect of Bioplex[®] on shell quality, including cracked eggs under commercial conditions.

Table 4. Correlation coefficients

Mineral	Mottling score vs Egg weight	Mottling score vs Egg shell deformation	Egg weight vs Egg shell deformation
Bioplex [®]	-0.1460 (NS)	0.4939 (P>0.01)	0.0057 (NS)
Inorganic	-0.1427 (NS)	0.6375 (P>0.01)	-0.0907 (NS)

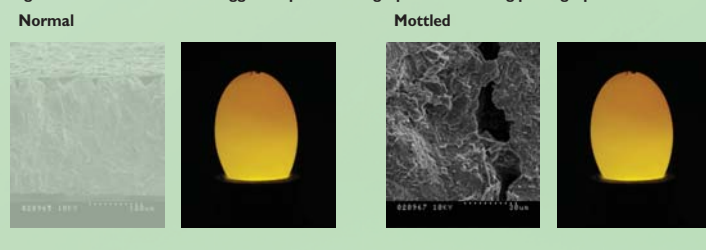
Table 5. Mineral composition of egg components (mg/g).

Mineral	Zn			Mn			Cu		
	Shell	Albumen	Yolk	Shell	Albumen	Yolk	Shell	Albumen	Yolk
Bioplex [®]	0.07	ND	0.020	ND	ND	ND	ND	ND	0.003
Inorganic	0.17	ND	0.019	ND	ND	ND	ND	ND	0.003
SD	0.15		0.002						0.001
Sig.	NS		NS						NS

Table 6. Excreta mineral composition (ppm of DM).

Mineral	Zn	Mn	Cu
Bioplex [®]	115.2	132.2	26.2
Inorganic	334.3	389.4	33.3
SD	17.7	33.7	5.3
Sig.	P>0.01	P>0.01	NS

Figure 1. Normal vs mottled egg shell: photomicrographs and candling photographs.



Actigen™ and Zn bacitracin: Comparative effects on performance, intestinal integrity and immunity of broilers



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Introduction

The poultry industry must reduce antibiotic use while maintaining efficient production of safe poultry meat and egg products. Probiotic feed ingredients, by means of promoting gut health, aid the animal in defense against disease.

Objective

To investigate the effects of supplementing Actigen™ (Alltech Inc.) on broiler performance, intestinal integrity, and immunity.

Materials and methods

Experimental design

- 3,135 male 1-d-old broiler chicks (Arbor Acres Plus x Ross)
- Completely randomized block design
- 5 treatment groups, 11 replicates per trt
- 55 pens, 57 birds per pen
- Pens equipped with nipple drinkers, tube feeders and standard gas space heaters
- Birds vaccinated against Newcastle disease (LaSota virus type) at 18 d of age

Treatments

- T1: Negative control (corn-soy diet) without Zn bacitracin
- T2: Positive control (corn-soy diet) with Zn bacitracin
- T3: Actigen™ – Diet T1 with Actigen™ (1–21 d, 400 g/t; 22–42 d, 400 g/t)
- T4: Actigen™ – Diet T1 with Actigen™ (1–21 d, 400 g/t; 22–42 d, 200 g/t)
- T5: Actigen™ – Diet T1 with Actigen™ (1–21 d, 200 g/t; 22–42 d, 200 g/t)

Measurements

- Body weight, ADG, feed intake, FCR weekly; mortality
- Intestinal lesion incidence and score (1 – 5; Table 1) (2 birds/pen, at 21, 35 and 42 d of age)
- Blood leukocyte count and heterophil:lymphocyte ratio (2 birds/pen at 21, 35 and 42 d of age).

Data analysis

- ANOVA combined with LSD test.
- Mortality data corrected by arcsine transformation.
- Chi-square test used for scores and intestinal mucosa changes (P<0.05).

Results

- Body weight (Table 2) differed (P<0.05) between treatments at 7, 14, 21 and 28 days of age only.
- FCR tended to be improved by all treatments (Figure 1, P>0.05).
- Mortality did not differ (P>0.05) between treatments but tended to be lower in birds given Actigen™.
- Percentages of intestinal lesions were similar among treatments (Table 3).
- Leukocyte counts and heterophil:lymphocyte ratio (Table 4) were in normal ranges and were unaffected by treatment.

Conclusions

- Supplementation of broiler chicks with Actigen™ increased body weight during the first 28 d (P<0.05), compared with diets containing Zn bacitracin.
- FCR tended to be improved by all treatments (P>0.05).
- Mortality was numerically lower in birds given Actigen™, compared with negative and positive controls.

Table 1. Score classification and description of changes in the intestinal mucosa.

Scores	Description
0	Intestinal mucosa with normal appearance.
1	Slightly congested, slight hyperemia and inflammation, normal mucus secretion.
2	Hyperemia severe but variable; increased mucus secretion.
3	Severe hyperemia throughout; intestinal folds inflamed with slight epithelial shedding; increased mucus with cloudy appearance and slightly bloody.
4	Mucosa totally hemorrhagic dispersed areas with shedding of the epithelia, intestinal folds severely hemorrhagic, abundant mucus, cloudy with a variable degree of bleeding and pus.
5	Mucosa totally hemorrhagic, areas of ulceration clearly defined, intestinal folds shedding or ulcerated, marked inflammation of the tissues.

Table 2. Effect of Actigen™ on broiler body weight (g/bird).

Treatments	Age (days)					
	7	14	21	28	35	42
T1	156.6 ^a	426.7 ^a	875.4 ^a	1366.4 ^a	1935.7	2468.8
T2	147.0 ^b	399.4 ^b	823.6 ^b	1314.5 ^b	1910.8	2467.6
T3	155.9 ^a	418.6 ^{ab}	867.8 ^a	1373.0 ^a	1962.0	2478.2
T4	157.1 ^a	428.6 ^a	873.0 ^a	1373.3 ^a	1941.1	2468.0
T5	154.6 ^a	419.0 ^{ab}	866.1 ^a	1375.3 ^a	1933.8	2450.8
CV	4.09	4.98	3.13	3.19	2.44	2.50

^{ab}Means differ (P<0.05)

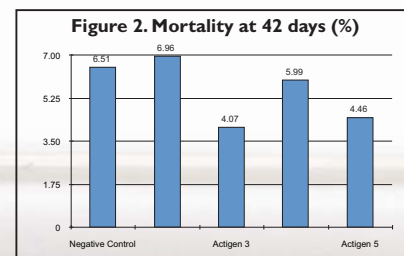
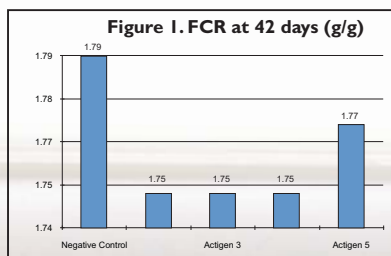
Table 3. Effect of Actigen™ on small intestinal lesion incidence and scores at 42 d of age.

Treatment	General lesions (%)	Lesions (%)		
		Score 1	Score 2	Score 3
T1	68.2	45.5	22.7	0.0
T2	77.3	68.2	13.6	0.0
T3	77.3	40.9	36.4	0.0
T4	54.6	36.4	18.2	0.0
T5	77.3	50.0	22.7	4.6

^{ab}Means differ (P<0.05)

Table 4. Effect of Actigen™ on leukocytes, heterophil:lymphocyte ratio (H:L) and counts at 42 days.

	Leukocytes	Heterophils	Lymphocytes	H:L
T1	23,559	13.6	84.6	0.16
T2	21,163	12.4	85.6	0.15
T3	21,181	13.8	84.3	0.16
T4	23,884	12.7	85.0	0.15
T5	22,409	12.0	85.6	0.14
CV	21.88	29.0	4.47	



Effect of Sel-Plex[®] and vitamin E supplementation to broilers on meat quality characteristics of raw and marinated breast fillets

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Introduction

Supplementation with Sel-Plex[®] (Alltech Inc.) in broiler diets has positive effects on meat quality characteristics of broiler breast fillets, specifically in reducing drip loss and improving oxidative stability. Marinating has become a widespread technique that is utilized in the poultry industry to improve meat tenderness and reduce the drip loss. However, information is lacking on the effects of dietary supplementation of antioxidants on marinated meat products. A study was conducted to evaluate the effects of Sel-Plex[®] and vitamin E (vit. E) supplementation on meat quality characteristics of both raw and marinated breast fillets.

Materials and methods

Experimental design

- 576 Cobb 500TM broilers
- 4 treatment groups
- 48 pens of 12 birds/pen (12 replicates per treatment)

Treatments

- Control – corn-soybean meal diet (no Se or Vit. E added)
- SP – Control + 0.3 ppm Se (Sel-Plex[®])
- VE – Control + vit E (all-*rac*- α -tocopherol acetate) at 30 IU/kg
- VE+SP – Control + vit. E at 30 IU/kg + 0.3 ppm Se (Sel-Plex[®])

Measurements

- Broilers were harvested and breast fillets (pectoralis major) were collected at 49 d of age for the raw and 56 d of age for the marinated portions of the experiment.
- Breast fillets were marinated in a solution containing 3.2% sodium pyrophosphate and 4% NaCl (pH: 9.74) for 13 h.
- Meat quality characteristics important to the consumer were evaluated:
 - Water holding capacity via drip loss of the breast fillets at 3 and 7 d of refrigerated storage
 - Oxidative stability via thiobarbituric acid reactive substances (TBARS), which react with byproducts from lipid oxidation, in breast fillets after 7 d of refrigerated storage
 - Color stability over 7 d refrigerated storage by objective measures of CIE values of lightness (L*), redness (a*), and yellowness (b*)
 - Cooking loss (moisture loss) and tenderness of the cooked product

Results

Raw breast fillets

- Sel-Plex[®] supplementation significantly reduced drip loss of fillets at 3 d over the control and vit. E treatments (Figure 1). At 7 d, drip loss for the Sel-Plex[®] treatment was lower ($P < 0.05$) than that of the vit. E treatment.
- Vit. E + Sel-Plex[®] supplementation considerably reduced TBARS values of fillets after 7 d of storage over the control, thereby extending product shelf life by improving oxidative stability (Figure 2).
- Treatments did not affect color stability, cooking loss, or tenderness values of raw fillets.

Marinated breast fillets

- Use of a basic marinade (pH>7) may have increased the susceptibility of the fillets to lipid oxidation since the 7 d TBARS values were considerably higher than those observed for the raw fillets.
- Overall, antioxidant supplementation reduced ($P < 0.05$) the 7 d TBARS values over the control in the marinated fillets, with the greatest reduction observed for the vit. E + Sel-Plex[®] treatment (Figure 3).
- Treatments did not affect color stability, cooking loss, or tenderness values of marinated fillets.

Conclusions

Increased susceptibility for lipid oxidation in marinated chicken products seems curbed by supplementing broiler diets with both Sel-Plex[®] and vitamin E, thus extending product shelf life.

Figure 1. 3 and 7 d (respectively for each diet) drip loss values for the raw breast fillets (n = 1 breast /pen). Means without a common letter differ $P < 0.05$.

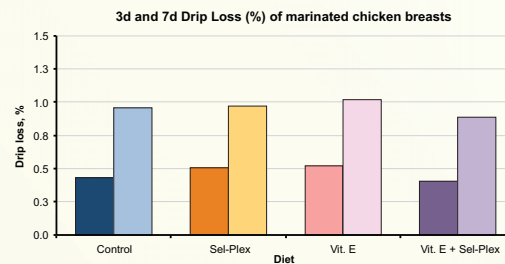


Figure 2. 7 d TBARS results for the raw breast fillets (n = 1 breast /pen). TBARS values are expressed as mg MDA/kg meat. Means without a common letter differ $P < 0.05$.

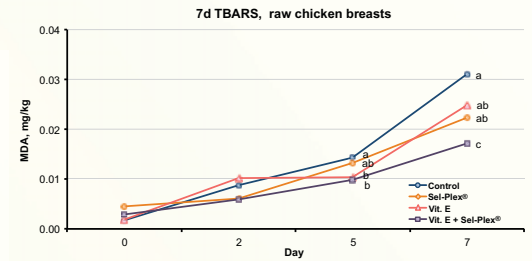
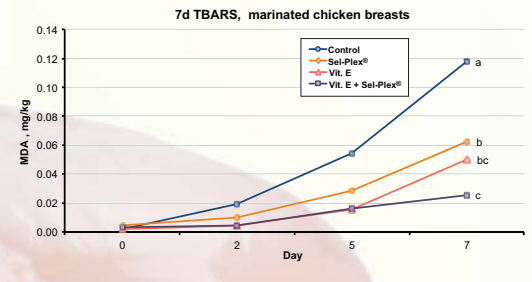


Figure 3. 7 d TBARS results for the marinated breast fillets (n = 1 breast /pen). TBARS values are expressed as mg MDA/kg meat. Means without a common letter differ $P < 0.05$.



Effects of Actigen™ and an AGP program on broiler performance under heat stress and health stress conditions



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Objective

To evaluate the performance and carcass characteristics of heat-stressed broilers reared on re-used litter when fed diets containing Actigen™ (Alltech Inc.) and/or an antibiotic growth promoter combination under heat stress and health stress conditions.

Materials and methods

Experimental design

- 960 Cobb 500 male and female 1-d-old broiler chicks
- 5 treatments with 8 replicates (pens) of 24 birds each
- 48 pens (1.60 m x 1.40 m)
- Continuous light program; temperature regulation (summer)
- Litter in pens was previously used by 2 consecutive flocks
- Ambient temperature range:
- Birds vaccinated against Marek's disease and Avian Pox
- Corn-soy-based diets formulated as per Rostagno et al. (2005).
- Treatments were isoenergetic and isonutritive.
- Feed and water available *ad libitum*

Treatments

- Hal – Halquinol (Hal, 30 g/t) (1–21 & 22–42 d)
- Avi – Hal (30 g/t) + avilamycin (Avi, 100 g/t) (1–21 & 22–42 d)
- Hal + Actigen™ 400 – Hal (30 g/t) + Actigen™ (400 g/t of feed) (1–21 & 22–42 d)
- Hal + Actigen™ 400/200 – Hal (30 g/t) + Actigen™ [400 g/t of feed (1–21 d) & 200 g/t of feed (22–42 d)]
- Hal + Actigen® 200 – Hal (30 g/t) + Actigen® (200 g/t of feed) (1–21 & 22–42 d)

Measurements

- Weight gain, feed intake, feed conversion, mortality rate, production efficiency index (PEI)
- Yield of carcass and premium cuts (breast, thigh, and drumstick), carcass and premium cuts calculated as a ratio of live weight

Data analysis

- ANOVA combined with Student-Newman-Keuls (P<0.05).

Results

- Hal plus Actigen™ at 400 g/t increased (P<0.05) weight gain compared with Hal alone in birds 1 to 21 d of age (Table 1).
- For d 1 to 42, Hal plus Actigen™ at 400 g/t (d 1 to 21) and 200 g/t (d 22 to 42) increased (P<0.05) weight gain compared with Hal alone to a level comparable to Hal and Avi combined.
- Feed intake, PEI, and mortality did not differ (P>0.05) between treatments; however, compared with Hal alone, FCR improved (P<0.05) d 22 to 42 in birds fed Hal plus Actigen™ at 400 g/t from d 1 to 21 followed by 400 or 200 g/t d 22 to 42.
- Carcass and premium cut yields did not differ between treatments (P>0.05).

Conclusions

- In heat-stressed, health-challenged broilers, Actigen™ was able to improve broiler performance to a greater extent than Hal alone and to an extent equivalent to Hal and Avi combined.
- Actigen™ added to Hal was able to significantly improve FCR d 22 to 42.
- Actigen™ added to Hal had no adverse effects on feed intake, feed efficiency production efficiency, carcass or premium cut yields, or mortality.

Table 1. Effect of Actigen® on average weight gain, feed intake and feed conversion from 1 to 21 d of age.

Variable	Treatment					CV (%)
	Hal	Hal+ Avi	Hal+ Actigen™ (400 g/t)	Hal+ Actigen™ (400/200 g/t)	Hal+ Actigen™ (200 g/t)	
Initial weight, g	42.3	42.2	42.4	42.3	42.2	0.73
Weight gain, g	735 ^b	771 ^{ab}	780 ^a	785 ^a	758 ^{ab}	4.26
Feed intake, g	1,096 ^b	1,132 ^a	1,121 ^a	1,153 ^a	1,121 ^a	7.67
FCR	1.50	1.47	1.44	1.47	1.48	8.20

^{a,b} Means differ P<0.05; Hal – Halquinol; Avi - Avilamycin.

Table 2. Effect of Actigen™ on average weight gain, feed intake and feed conversion for broilers from 22 to 42 d of age.

Variable	Treatment					CV (%)
	Hal	Hal+ Avi	Hal+ Actigen™ (400 g/t)	Hal+ Actigen™ (400/200 g/t)	Hal+ Actigen™ (200 g/t)	
Initial weight, g	776.8	824.8	822.7	826.9	803.2	3.73
Weight gain, g	1,593	1,727	1,694	1,725	1,638	7.31
Feed intake, g	2,963	3,084	2,989	2,972	2,930	5.51
FCR	1.86 ^a	1.79 ^{ab}	1.77 ^a	1.73 ^a	1.80 ^{ab}	3.74

^{a,b} Means differ P<0.05; Hal – Halquinol; Avi - Avilamycin.

Table 3. Effect of Actigen® on average weight gain, feed intake, feed conversion, and mortality rate for broilers from 1 to 42 d of age.

Variable	Treatment					CV (%)
	Hal	Hal+ Avi	Hal+ Actigen® (400 g/t)	HAL+ Actigen® (400/200 g/t)	HAL+ Actigen® (200 g/t)	
Initial weight, g	42.3	42.2	42.4	42.3	42.2	0.73
Weight gain, g	2,328 ^b	2,509 ^a	2,474 ^{ab}	2,509 ^a	2,399 ^b	5.52
Feed intake, g	4,047	4,235	4,104	4,171	4,071	5.19
Feed conversion	1.74	1.69	1.66	1.66	1.70	4.26
Mortality, %	13.9	9.7	12.5	11.5	17.4	94.43
PEI	282	325	318	325	283	15.70

^{a,b} Means differ P<0.05; Hal – Halquinol; Avi - Avilamycin.



Effect of selenium source on testes weight, selenium content and gene expression profiles in Single Comb White Leghorn roosters

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34

Se
78.96

Introduction and Objectives

Aside from the two selenoproteins, GPX4 and SEPP1, little is known about the role of Se in the testes. The objective of this study was to evaluate the effect of Se source on testes weight, Se content and gene expression profiles in mature roosters.

Materials and Methods

Animals:

- 17 wk-old Single Comb White Leghorn roosters (n=7/trt)
- Three treatments: basal diet (0.09 mg Se/d, Table 1) plus:
 - Control: no supplemental Se (C)
 - SS: 0.3 ppm inorganic Se, sodium selenite (SS)
 - SP: 0.3 ppm organic Se, Sel-Plex[®] (SP)

Sample collection:

At 40 wks of age, 7 birds were randomly selected from each treatment and testes removed and weighed. Samples (~1g) were flash frozen in liquid nitrogen.

Selenium analysis:

- Analyzed using atomic fluorescence spectrophotometry

RNA isolation and microarray

- RNA was isolated using Qiagen RNeasy kit; microarray profiling using Affymetrix Chicken genome array
- GeneSpring GX 10.0 used to qualify and normalize microarray data and Ingenuity Pathway Analysis used for biofunction analysis of differentially expressed genes.

Real-time PCR

- Power SYBR[®] Green PCR Master Mix (Applied Biosystems) using an ABI 7500 Real-Time PCR System
- Data normalized to the housekeeping gene ring finger protein 4 (Rnf4) and to control samples (Diet 1)
- Relative quantification (RQ) values were calculated using the delta delta Ct method

Table 1. Composition of basal diet.

Ingredient	% of diet
Ground corn	53.825
Soybean meal (48% CP)	29.45
Ground limestone	10.5
Corn oil	4.10
Dicalcium phosphate	0.985
Iodized salt	0.475
TMX-91-1 mineral mix*	0.25
DSM vitamin mix*	0.20
DL-methionine	0.215
Calculated composition (as fed)	
AME, kcal/kg	2.896
Protein, %	18.9
Calcium, %	4.26
Available P, %	0.298
Methionine, %	0.519
Methionine + cysteine %	0.819
Lysine, %	1.042

*Provided per kg of diet: 7937 IU vit A, 2205 ICU vit D3, 26 IU vit E, 1.6 mg vit K, 1.6 mg thiamin, 3.3 mg riboflavin, 8.8 mg pantothenic acid, 35 mg niacin, 3.2 mg pyridoxine, 0.088 mg biotin, 1.1 mg folic acid, 0.016 mg B12, 397 mg choline.

*Provided per kg of diet: 6 mg Cu 0.53 mg I, 120 mg Fe, 83 mg Mn, 60 mg Zn and 5 mg Co. No Se added; basal diet contained 0.094 ppm Se.

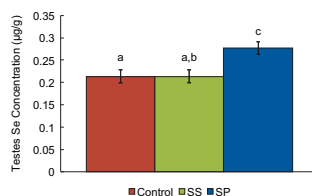
Statistical Analysis

- Proc Mixed (SAS) was the fixed effect and rooster was the random variable; Student's t-test to separate RQ means.
- Microarray data subjected to a t-test using GeneSpring GX 10.0 and statistical differences declared at P<0.01

Results: Summary

- Testes weights did not differ between treatments.
- Testes Se content was greater in SP than in control or SS roosters (P<0.01); but did not differ between control and SS roosters.
- 155 genes were differentially expressed in SS testes and 442 were differentially expressed in SP testes. 48 genes were commonly regulated in SS and SP testes
- The main biofunctions and pathways affected by selenite were related to cell cycle regulation, organ development and function, and cellular organization and function (Table 2).
- The main biofunctions and pathways affected by Sel-Plex[®] included cell signaling, cell structure and development and tissue structure and development (Table 3).

Figure 1. Effect of Se source on testes Se (µg/g).



Conclusion

Our preliminary gene expression findings suggest that in addition to playing a major role in the sperm morphology, Se is important for the regulation of expression of genes involved in the maintenance of testicular structure. In addition, Se supplementation in the form of SP leads to a greater enrichment of genes in these functional categories than SS. Using these initial findings, it is of great interest to investigate further the role of specific selenoproteins in these pathways.

Figure 2. Relative quantification (RQ) of mRNA levels of glutathione peroxidase 4 (GPX4) and selenoprotein P1 (SEPP1) in the testes of roosters supplemented with Se.

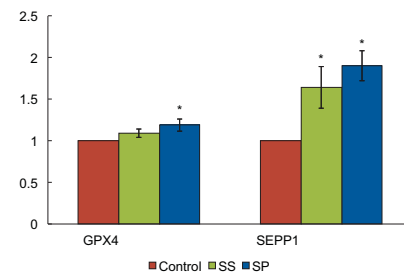


Table 2. Top canonical pathways and biofunction-associated genes altered by selenite supplementation

Top Canonical Pathways	Ratio
Induction of apoptosis by HIV	3/65 (0.46)
Cell cycle: G2/M DNA damage checkpoint regulation	2/44 (0.045)
p53 signaling	3/92 (0.033)
Type I Diabetes Mellitus signaling	3/119 (0.025)
TREM1 signaling	2/69 (0.029)

Top Biological Functions	No. of altered genes
Molecular and Cellular Functions	
Cell death	12
Cellular assembly and organization	8
Cellular Function and Maintenance	10
Cellular development	11
Gene expression	10
Physiological System Development and Function	
Hematological system development and function	10
Embryonic development	11
Organ development	4
Renal and urological development and function	2
Organ morphology	5

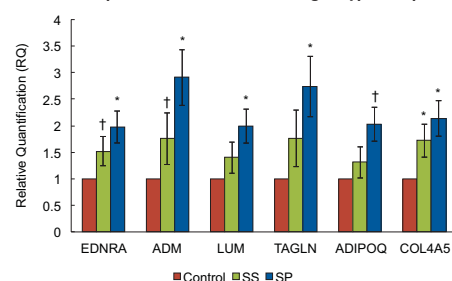
Table 3. Top canonical pathways and biofunction-associated genes altered by Sel-Plex[®] supplementation.

Top Canonical Pathways	Ratio
Hepatic fibrosis/Hepatic stellate cell activation	12/134 (0.09)
ILK signaling	15/186 (0.081)
Acute phase response signaling	12/178 (0.067)
Actin cytoskeleton signaling	14/235 (0.06)
Fcy receptor-mediated phagocytosis in macrophages and monocytes	8/101 (0.079)

Top Biological Functions

Molecular and Cellular Functions	No. of altered genes
Cellular movement	88
Cell morphology	74
Cellular Development	103
Cellular growth and proliferation	112
Cell death	105
Physiological System Development and Function	
Tissue development	90
Hematological system development and function	66
Immune cell trafficking	45
Lymphoid tissue structure and development	23
Organismal development	63

Figure 3. Validation of the changes in relative expression of select genes in the testes of control, SS or Sel-Plex[®] (SP) hens using real-time PCR. Selected genes were: EDNRA= endothelin receptor type A; ADM=adrenomedullin; LUM=lumican; TAGLN=transgelin; ADIPOQ=adiponectin 1Q; COL4A5= collagen, type IV, alpha 5.



Effect of NuPro® on performance and gut immunity of broiler chickens



A. Yitbarek¹, H.M. Echeverry¹, P. Munyaka¹, O. Abu-Dahab¹, M. Einarson³, S. Sharif⁴, W. Guenter¹, J.D. House², and J.C. Rodriguez-Lecompte^{1*}

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Introduction

- The innate immune system in broilers is the first line of defense against invading organisms.
- Toll-like receptors (TLRs) are part of the pathogen recognition receptor family and help identify pathogenic organisms to initiate immune response.
- Under TLR activation, cytokines are produced by heterophils to induce further immune response.
- Nucleotides are macromolecules that can be synthesized *de novo*, but day-old birds do not synthesize sufficient amounts.
- Dietary supplementation with nucleotides has improved immune response in infant humans.

Objectives

Investigate the effect of NuPro® (Alltech Inc.) comprised of yeast-derived protein and other macromolecules, on production parameters (feed intake, feed conversion ratio, body weight, and mortality rates), expression of TLR 2 & 4, and Interleukin 6, 10, & 12 as well as interferon- γ of the innate immune system of broiler chickens.

Material and methods

- Randomized complete block design
- Three treatments:
 - Control: Monensin 99 mg/kg (T1),
 - Monensin + growth promoter (BMD 110 mg/kg)(T2), and
 - Monensin + BMD + 2% NuPro® in the starter diet (NuPro®)(T3)

- 18 Pens, 60 male day old birds per pen, 6 replications per treatment (total # of birds 1080).
- Birds were weighed every 7 days starting at day 0.
- 5 birds from each group euthenized at 42 days
- Samples collected from: Ileum, cecal tonsils, bursa, and spleen.
- TLR 2 & 4 expression was measured in all 4 sample collection sites.
- IL 12p40, IL 6, IL 10 & IFN- γ expression was measured in all 4 sample collection sites.
- Plasma malondialdehyde measured in all treatments.
- Data statistically analyzed by GLM, SAS 9.20

Figure 1. Plasma malondialdehyde (MDA) amount, an indicator of lipid peroxide. Bars represent mean + SE (n = 5)

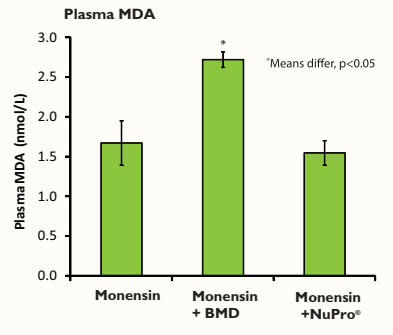
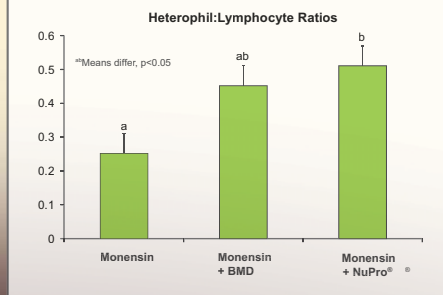


Figure 2. Ratios of Heterophils:Lymphocytes in blood



Results

Table 1. Performance and mortality in broiler chickens

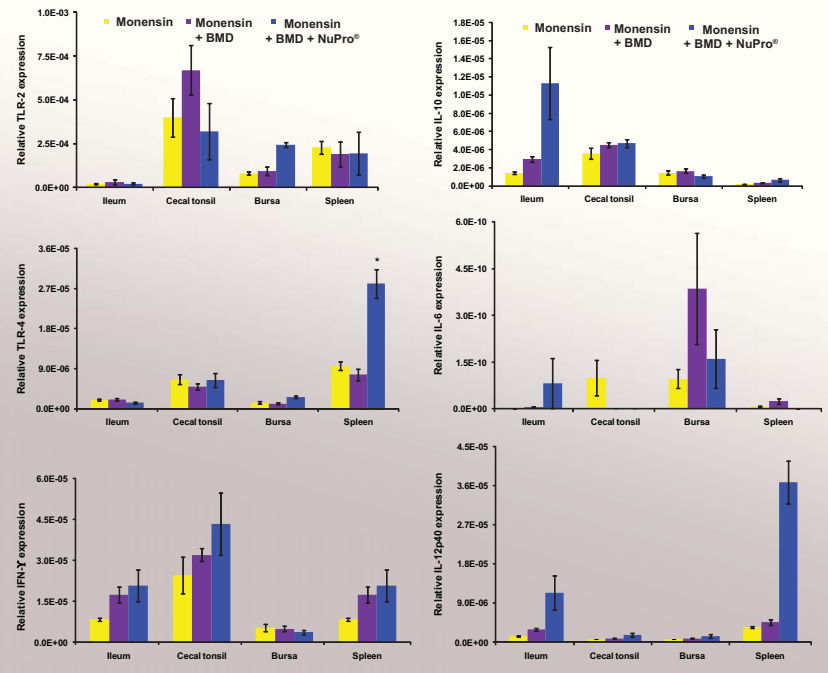
Variable	Diets											
	Starter			Grower			Finisher			Cumulative		
	Control	BMD	NuPro	Control	BMD	NuPro	Control	BMD	NuPro	Control	BMD	NuPro
Feed intake	571	571	565	831	826	821	4384 ^a	4498 ^a	4516 ^a	5787 ^a	5895 ^a	5905 ^a
Body weight	481 ^a	471 ^b	457 ^c	580	581	580	2204	2330	2328	3317 ^a	3430 ^a	3413 ^a
FCR	1.180 ^a	1.207 ^a	1.245 ^a	1.433	1.405	1.438	2.028 ^a	1.895 ^b	1.942 ^b	1.746	1.720	1.731
Mortality										5.56	6.39	3.33

^{a,b}Means differ, P<0.05

Summary

- Mortality tended to be lower in broilers fed NuPro® (T3) in comparison to T1 & T2.
- Expression of TLR 2 & 4 was unaffected
- IL 12p40 was significantly up-regulated by T3 in the spleen indicating activation of NK cells.
- Expression of IL6 was higher in T2 indicating a systemic challenge.
- Expression of INF- γ was higher in ileum of birds given NuPro® indicating Th1 differentiation.

Figure 3. Relative TLR and Cytokine expression levels in the cecal tonsil, ileum, bursa and spleen. Bars represent mean +SE (n = 60), * indicates significant relative gene expression (p < 0.05)



Acknowledgements

Alltech Inc., Manitoba Agri-Food Research and Development Initiative (ARDI), Poultry Research Unit: Harry Muc



Toll-like receptor and cytokine profile of chickens supplemented with Actigen™



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⁴Department of Pathobiology, University of Guelph, ON, Canada

Introduction

- Innate immune system in broilers is the first line of defense against invading organisms.
- Toll-like receptors (TLR) are part of pathogen recognition receptor family and help identify pathogenic organisms to initiate immune response.
- Carbohydrate receptors are important sensor molecules that can be stimulate the innate immune system. They can trigger immune responses associates with phagocytosis and cytokine production.
- Dietary supplementation with yeast-derived CHO has improved gut microbiology by neutralization of Pilli I bacteria.

Objectives

Investigate the effect of a yeast derived carbohydrate fraction (Actigen™, Alltech Inc.) on production parameters (feed intake, feed conversion ratio, body weight, & mortality rates), gut and blood immune parameters in broilers chickens.

Material and methods

- Factorial design
- Three treatments:
 - Control: Monensin, 99 mg/kg (T1),
 - Monensin + growth promoter (BMD 110 mg/kg) (T2), and
 - Monensin + Actigen™ (800 g starter, 400 g grower, 200 g finisher) (T3)
- 18 Pens, 60 male day old birds per pen.
- Every treatment was replicated 6 times. Total # of birds 1080.
- Birds were weighed every 7 days starting with day 0.
- 5 birds from each group euthanized at 42 days
- Samples collected from: Ileum, and cecal tonsils.
- TLR 2 & 4 expression was measured in all 2 sample collection sites.
- IL 12p40, IL 6, IL 10 & IFN- γ expression was measured in 2 locations.
- Heterophil/Lymphocyte ratio was measured in all treatments.
- Data statistically analyzed by GLM, SAS 9.20

Results

Table 1. Performance and mortality of broiler chickens.

Variable	Diets											
	Starter (1-14d)			Grower (15-21d)			Finisher			Cumulative		
Feed intake	Control	BMD	Actigen	Control	BMD	Actigen	Control	BMD	Actigen	Control	BMD	Actigen
	571	571	578	831	826	832	4384 ^b	4498 ^b	4462 ^b	5787	5895	5871
Body weight	481 ^a	471 ^b	476 ^{ab}	580	581	586	2204 ^b	2330 ^a	2325 ^{ab}	3317 ^a	3430 ^{ab}	3437 ^a
FCR	1.180 ^a	1.207 ^a	1.213 ^a	1.433	1.405	1.418	2.028 ^a	1.895 ^a	1.919 ^b	1.746	1.720	1.708
Mortality										5.56	6.39	3.89

^{ab}Means differ, P<0.05



Summary

- Actigen™ supplementation affected feed intake and body weight. The mortality was the lowest in comparison to T1 (Monensin) and T2 (Monensin + BMD) with direct economic benefits.
- Expression of TLR 2 was unaffected by treatment either in the ileum or cecal tonsils. However, a downregulation of TLR4 was found, indicating a direct effect of Actigen™ on the amount of gram-negative bacteria in the GIT.
- Expression of IL-6 was higher in Actigen™ in the cecal tonsil area indicating a local activity against microorganisms.
- Expression of IL-10 was not significant between groups; however associating the numerical difference in the favor of the Actigen™ group with the upregulation of IL-6 is possible to infer a high regulation between pro- and anti-inflammatory responses.
- IL -12p40 and INF- γ expression were not significant between groups.

Acknowledgements

Alltech Inc., Manitoba Agri-Food Research and Development Initiative (ARDI), Poultry Research Unit

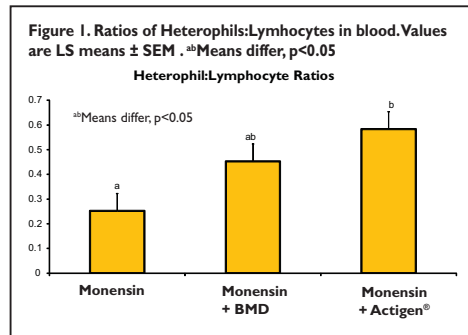


Figure 1. Ratios of Heterophils:Lymphocytes in blood. Values are LS means \pm SEM. ^{ab}Means differ, p<0.05

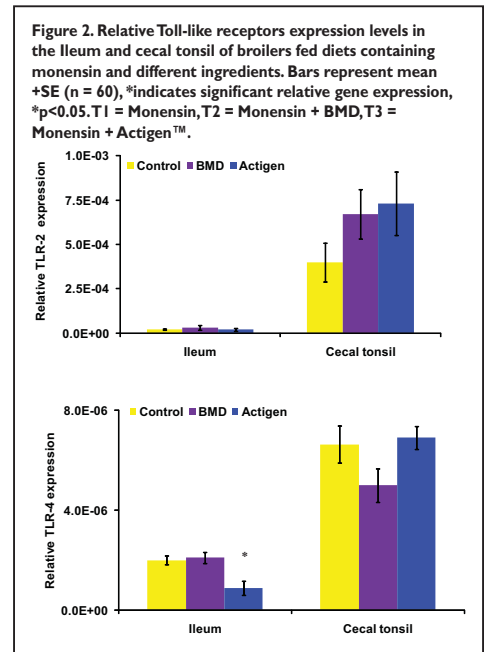
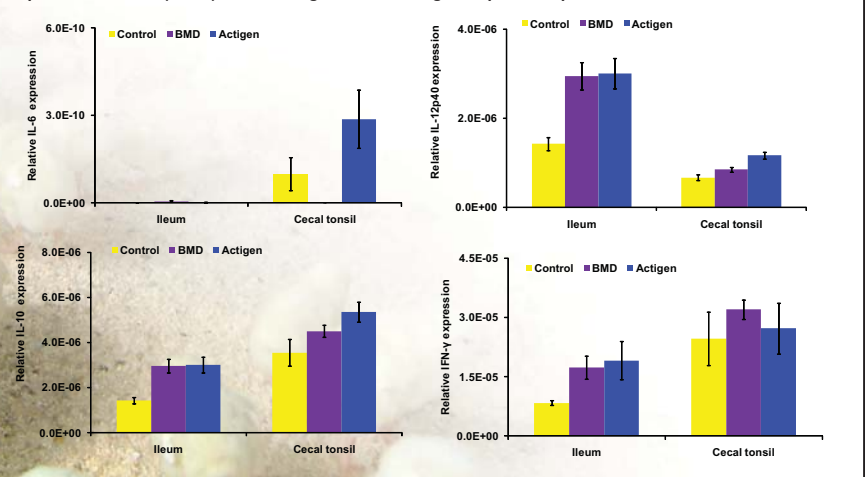


Figure 2. Relative Toll-like receptors expression levels in the ileum and cecal tonsil of broilers fed diets containing monensin and different ingredients. Bars represent mean \pm SE (n = 60). *indicates significant relative gene expression, *p<0.05. T1 = Monensin, T2 = Monensin + BMD, T3 = Monensin + Actigen™.

Figure 3. Relative cytokine (IL-6, IL-10, IL-12p40 and IFN- γ) expression levels in the ileum and cecal tonsil. Bars represent mean \pm SE (n = 60). *indicates significant relative gene expression, *p<0.05.



Temporal changes in selenium-dependent intestinal recovery in reovirus-challenged broiler chickens

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Introduction

Avian reovirus (ARV)-associated diseases range from asymptomatic infections to more serious conditions involving skeleton, muscles, and digestive organs (Benavente and Martínez-Costas, 2007, Ni *et al.*, 1995). Natural ARV gastrointestinal infection may also involve the bursa of Fabricius (Jones *et al.*, 1989). ARVs of turkey origin can affect chickens as judged by experimental evidence (Al-Afalet and Jones, 1989, Spackman *et al.*, 2005). Here we wished to extend our earlier observations on these effects using the turkey origin ARV-CU98, which is able to inhibit growth and development in broiler chickens (Macalintal, 2004). Generally, ARV is cleared from the intestinal tract within seven days post challenge (Songserm *et al.*, 2003), but when ARV-CU98 was orally inoculated into broiler chickens at day of hatch, signs of enteritis were seen at 21 days of age as demonstrated by the presence of intestinal villi that were shortened, blunted and with greater crypt depth (Read-Snyder, 2009; Read-Snyder *et al.*, 2009). Broilers fed Sel-Plex[®] in both control and infected groups had increased villus lengths, more shallow crypts, and greater height: crypt depth ratios. Although ARV had probably cleared the system at 21 days post challenge, the damage attributed to the resultant intestinal pathology remained evident for an extended time.

Objectives

The timing of intestinal recovery within dietary selenium treatments after ARV challenge remained obscure. Thus, the objectives of this investigation were to

- ascertain the effects of dietary selenium on ARV intestinal pathology
- describe intestinal recovery using morphometric measurements

Material and Methods

Chicks and Diets: Feather-sexed Ross 708 chicks were placed in heated batteries and fed isocaloric (3069 Kcal/kg ME; 16.37% CP; 2.06% fat; 1.57% fiber) Torula yeast diets with either

- No supplemental Se (<0.02 ppm)
- Sel-Plex[®] (Alltech, Inc. Nicholasville, KY; 0.3 ppm Se) or
- Sodium Selenite (0.3 ppm Se).

ARV-CU98 Challenge: At 5d post-hatch by oral gavage ($10^{4.2}$ pfu/chick)

Tissue Collection and Fixation:

- 10 mm segments of duodenum, jejunum and ileum were dissected and flushed with PBS
- 5 μ tissue sections were stained with H & E
- Histomorphometric parameters were determined by a computerized microscope-based image analyzer.

Statistical Analysis: GLM (SAS Institute, 1996); using the average of ten measurements for each parameter from each bird at each time post challenge. Fisher's LSD was used where appropriate.

Results

The influence of selenium supplementation on ileum villus height, depth of the crypts of Lieberkuhn, villus height: crypt depth ratios, and thickness of the muscularis mucosa in response to ARV challenge are shown in the following figures.

Summary and Conclusion

- Sel-Plex[®]-fed chicks were found to have an earlier initiation of intestinal morphology recovery, at day 7 following a reovirus challenge. This advantage was maintained through 16 days post challenge. Control-selenium deficient chicks at 16 days post challenge also had significantly long villi than selenite-fed broilers.
- Crypt depths were increased through 7 days post reovirus challenge, but began to show decreasing depths by 16 days post challenge. Selenite-fed broilers tended to have crypt depths greater than all other groups throughout the experimental period.
- Overall, Sel-Plex[®]-fed broilers maintained greater villus height: crypt depth ratios than all other groups. Reovirus challenge caused a significant decrease in the villus height: crypt depth ratios at 2 days post challenge. These ratios began to recover by 16 days post challenge, but not to the same level as found in the Sel-Plex[®]-fed, challenged and Sel-Plex[®] control groups.
- The muscularis mucosa was generally thinnest within selenium deficient groups. Selenium supplementation increased muscularis mucosa thickness. At 4 days post reovirus challenge, muscularis mucosa thickness increased significantly and remained thickened through 16 days post challenge.
- It was concluded that Sel-Plex[®] supplementation had increased the rate of recovery of the ileum of broiler chickens given a reovirus challenge. The reason for the more rapid recovery of intestinal morphology in Sel-Plex[®] fed broilers is not readily apparent, but it was hypothesized that improved redox status in the Sel-Plex[®]-supplemented animals might have played a significant role.
- Points of Sel-Plex[®]-related intestinal recovery post reovirus challenge, appeared to be associated with expression of polymeric immunoglobulin receptor and secretory IgA in bile and in the intestinal fluids (see accompanying posters).
- Increased rate of intestinal recovery from challenge by an enteric pathogen should aid in improved performance of the flock.

Figure 1. Villus height effects.

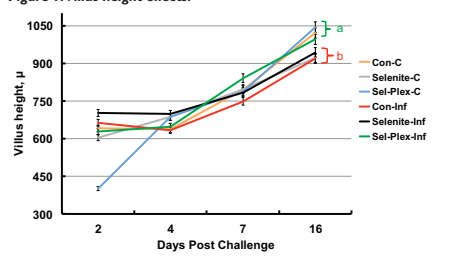


Figure 2. Crypt depth effects.

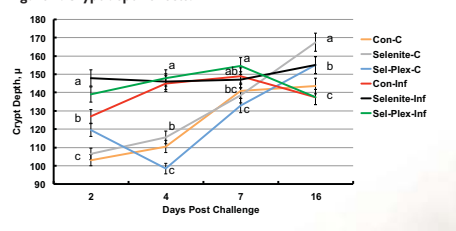


Figure 3. Villus height: crypt depth ratio responses.

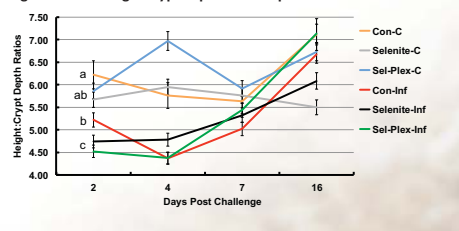
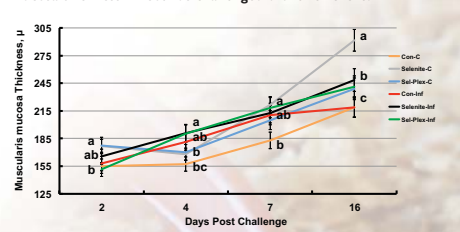


Figure 4. Influence of selenium supplementation on muscularis mucosa thickness in reovirus-challenged broiler chickens.



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Differential effects of sodium selenite and Sel-Plex® selenium yeast on the hepatic gene expression profile of laying hens

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Introduction

While both inorganic and organic Se sources are currently used in animal feeds, numerous studies have suggested the advantages of using organic Se over inorganic Se, indicated by higher bioavailability, better animal performance and fertility; yet the molecular mechanisms are unclear. The objective of the current study is to evaluate and compare the effects of inorganic (sodium selenite, SS) and organic Se (Sel-Plex®, SP, Alltech Inc.) on the hepatic gene expression profiles of laying hens.

Materials and methods

Bird handling and diets

198 Cobb×Cobb broiler breeder hens were housed in floor pens in groups of 11 hens. From hatching until 6 wks of age, all hens were fed the same starter diet. At 6 weeks, hens were switched to a basal developer diet and randomly assigned to one of the three treatments: basal diet (control, C), basal diet + 0.3 ppm organic Se (SP) or basal diet + 0.3 ppm inorganic Se (SS). At 23 weeks of age, hens were photostimulated (16 h light : 8 h dark) and switched to a basal layer diet until sample collection (Table 1). At 49 weeks of age, seven hens from each treatment were randomly selected and euthanized by argon asphyxiation followed by cervical dislocation. The liver samples were collected and snap frozen in liquid nitrogen, and then stored at -80°C.

Microarray gene expression profiling

Gene expression data were obtained for each of the birds using the Chicken Genome Array (Affymetrix) which is comprised of 32,773 probe sets corresponding to over 28,000 chicken genes. Total RNA isolation, cRNA preparation and hybridization to the gene-chip followed the standard protocols suggested by Affymetrix.

Microarray data and pathway analysis

To identify genes differentially regulated and the bio-physiological themes they represent, statistical and bioinformatics procedures including data normalization, differential tests and signaling pathway analyses were conducted. Genes with a $P < 0.05$ and corresponding signal intensity fold change (FC) > 1.2 were deemed as changed. Multiple bioinformatics tools including GeneSpring (Agilent), Ingenuity Pathways Analysis (IPA, Ingenuity Systems) and LifeGenDB (LifeGen Technologies) were used in this study.

Table 1. Top hepatic gene networks associated with dietary Se supplementation

ID	Networks	Score*
Gene function networks related to SP		
1	Cell Morphology, Infection Mechanism, Behavior	42
2	Cell Cycle, Cellular Movement, Cell Morphology	42
3	Cell Signaling, Cell Morphology, Hematological System Development and Function	38
4	Cellular Compromise, Cell Cycle, Cell Morphology	38
5	Gene Expression, RNA Damage and Repair, Lipid Metabolism	35
Gene function networks related to SS		
1	Hematological System Development and Function,	41
2	Dermatological Diseases and Conditions, Genetic Disorder, Lipid Metabolism	39
3	Cell-To-Cell Signaling and Interaction, Connective Tissue Development and Function, Cellular	34
4	Endocrine System Disorders, Gastrointestinal Disease, Inflammatory Disease	30
5	Connective Tissue Disorders, Genetic Disorder, Dermatological Diseases and Conditions	29

The most significant functions for each network are listed. Network score was based on a p-value calculation, which calculates the likelihood that the network eligible molecules that are part of a network are found therein by random chance.

Conclusions

The differences in liver gene expression profiles, especially on genes involved in energy production and cellular stress, may partially explain the reported biological differences related to SP and SS.

Figure 1. Venn diagram of genes differentially regulated by SP or SS in the liver of hens when compared to control birds.

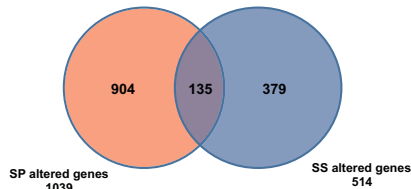
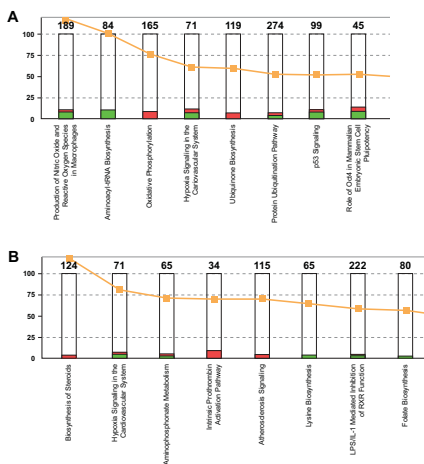


Figure 2. Significant canonical pathways that are involved with SP (A) or SS (B) regulated genes. Green bar = downregulated, red bar = upregulated.



Summarized results

- Compared with control, 1039 transcripts were differentially regulated by SP (508 down, 531 up, $P < 0.05$, $FC > 1.2$); 514 transcripts were changed by SS (207 down, 307 up respectively). 135 transcripts were commonly changed by both SP and SS (Figure 1).
- Functional analysis of genes differentially expressed in liver indicated that SP supplementation may affect gene networks associate with multiple biological functions such as cell morphology, cell cycle, gene expression, RNA damage and repair, lipid metabolism and infection mechanism. SS regulated genes are related to immune response, inflammatory disease, lipid metabolism and system development and function (Table 1).
- Pathway analysis further suggested that genes relate to energy production processes in mitochondria such as oxidative phosphorylation and ubiquitome biosynthesis pathway were increased by SP (Figure 2A). But no such affects were detected in SS-fed chickens (Figure 2B).
- Results also indicated that genes associated with production of nitric oxide and reactive oxygen species in macrophages was suppressed by SP. The two genes CCNK and GADD45B, which are involved in P53 signaling were also down-regulated by SP (Figure 3). These changes may suggest a lower cellular stress condition (hypoxia, oxidative and DNA damage etc) in SP-fed chickens.
- SS specifically up-regulated the expression of aryl hydrocarbon receptor (AHR), aldo-keto reductase (AKR1D1), microsomal glutathione S-transferase 2 (MGST2) (Figure 4), three genes whose defects potentially relate to increased risk of several hepatic system diseases. Besides, multiple up-regulated genes associated with atherosclerosis signaling and intrinsic prothrombin activation pathway in SS-fed chickens may suggest a link between supplementation of high level inorganic Se and the risk of chronic inflammatory response in the liver.

Figure 3. Effects of Se supplementation on hepatic gene expression of laying hens.

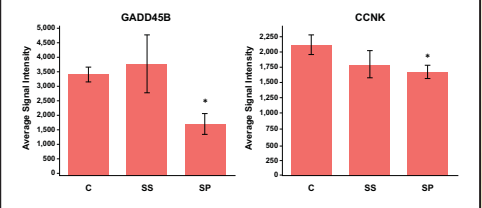
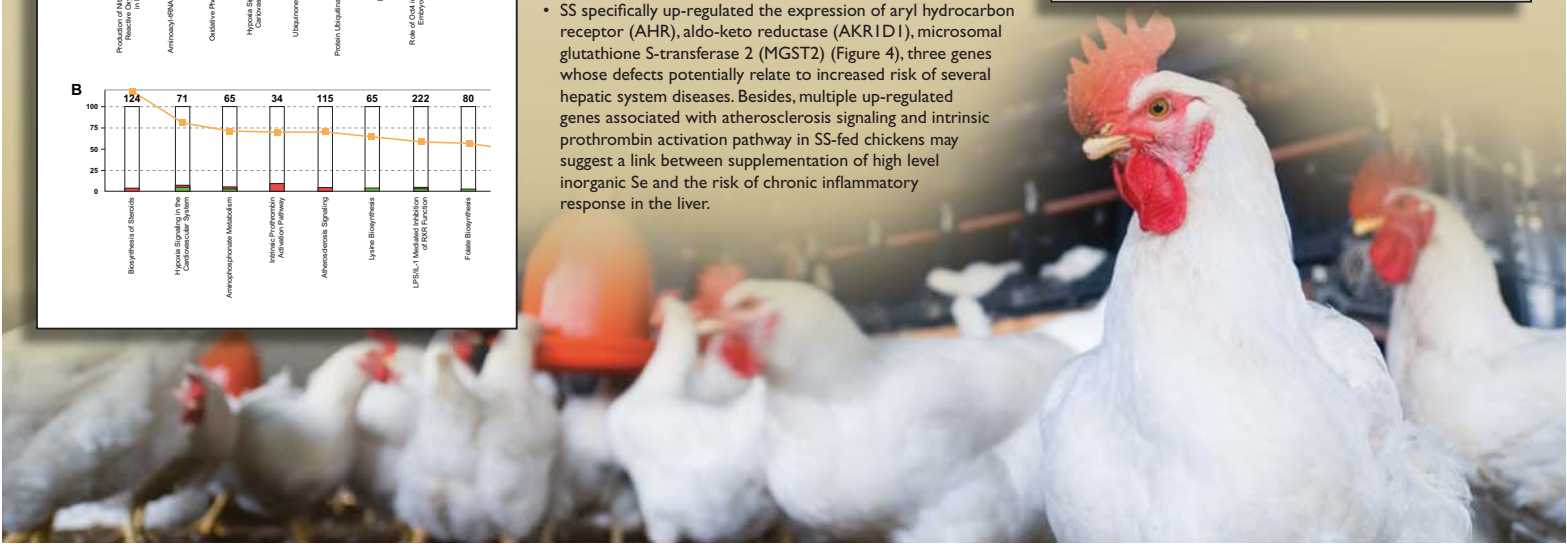
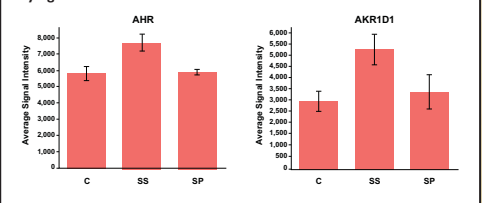


Figure 4. Effects of Se supplementation on hepatic gene expression of laying hens.



Gene expression study reveals the association of dietary supplementation of Actigen™ and the regulation of pathogen-influenced signaling pathways in broiler chickens



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Introduction

Previous studies suggested that dietary supplementation of Actigen™ (Alltech, Inc.), a product derived from yeast cell wall mannan oligosaccharides, has a beneficial effect in promoting intestinal health and performance of animals. The objective of this study was to investigate the effects of Actigen™ on intestinal gene expression profiles of broilers to gain insight into the mechanisms related to its activities.

Materials and methods

- Bird handling and diets:** 1-day old chicks were allocated to 2 groups and randomly assigned to one of the following diets: corn-soy based control diet (Control) or Control diet plus 400 g/ton Actigen™. After 3 weeks of feeding, seven chicks from each diet group were killed and jejunum samples were collected and snap frozen in liquid nitrogen, and then stored at -80°C.
- Microarray gene expression profiling:** Gene expression data were obtained for each of the birds using the Chicken Genome Array (Affymetrix) which is comprised of 32,773 probe sets corresponding to over 28,000 chicken genes. Total RNA isolation, cRNA preparation and hybridization to the gene-chip followed the standard protocols suggested by Affymetrix.
- Microarray data and pathway analysis:** To identify genes differentially regulated and the bio-physiological themes they represent, a series of statistical and bioinformatics procedures were run on the data, including differential tests and signaling pathway analyses. To minimize the possibility of misleading findings, probe sets with very low signal intensity and labeled as 'Absent' by the Affymetrix algorithm across samples were excluded from further analysis. Only genes with a $P < 0.01$ and corresponding signal intensity fold change (FC) > 1.2 were deemed as changed.

Results

- Compared with control, 928 genes were significantly changed by Actigen™ (456 down, 472 up, $P < 0.01$, FC > 1.2), in which the expression of TMIGD1, a gene encoding for a transmembrane and immunoglobulin domain was increased by 800%.

- Functional analysis on genes differentially expressed indicated that Actigen™ activity is likely involved with biological processes such as carboxylic acid metabolic process, vitamin metabolic processes, lipid metabolism and several others (Figure 1).
- Pathway analysis further suggested that cellular fatty acid metabolism, cofactor and vitamin metabolism, carbohydrate metabolism and energy production related genes were down-regulated by Actigen™ (Figure 2A); while analysis focused on up-regulated genes suggests a strong connection between Actigen™ and signaling pathways that are directly involved in cellular immune response, inflammatory response and antimicrobial response such as toll-like receptor signaling, interferon signaling and retinoic acid inducible protein-1 (RIG1) receptor mediated innate immunity (Figure 2B).
- Significant up-regulation of genes such as toll-like receptor 3 (TLR3), myxovirus resistance 1 (MX1), interferon regulatory factor 7 (IRF7), suppressor of cytokine signaling 1 (SOCS1) and down-regulation of ADP-ribosyltransferase (CHAT2) further indicate that Actigen™ has modulatory effects on the intestinal immune system (Figure 3, 4).

Conclusions

Alterations in intestinal gene expression profiles, especially on genes involved in cellular immune and antimicrobial response confirmed that dietary supplementation of Actigen™ may enhance the antibacterial capability of broilers.

Figure 1. Gene ontology (GO) analysis of genes mapped for 928 differentially expressed genes by Actigen™ supplementation in broiler diet. The GO terms were based on the ToppGene database (<http://toppgene.cchmc.org/>).

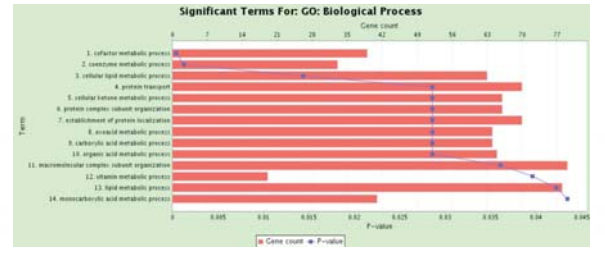


Figure 2. Significant signal pathways related with down-regulated (A) or up-regulated (B) genes by Actigen™. Green bar = down-regulated genes, red bar = up-regulated genes.

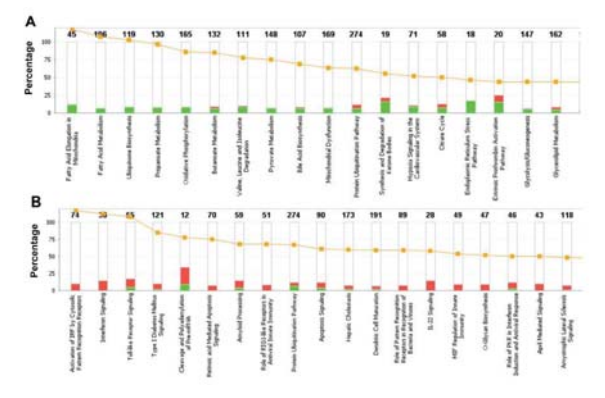


Figure 3. Genes involved in interferon signaling were significantly induced in the intestine of broilers by Actigen™. Red indicates increased expression by Actigen™.

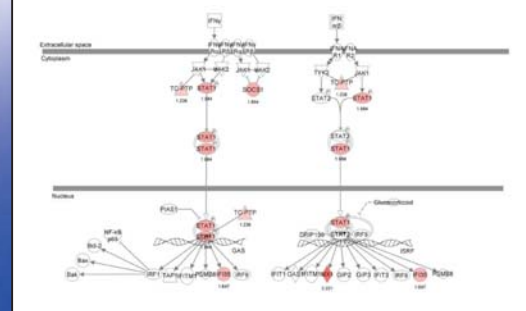
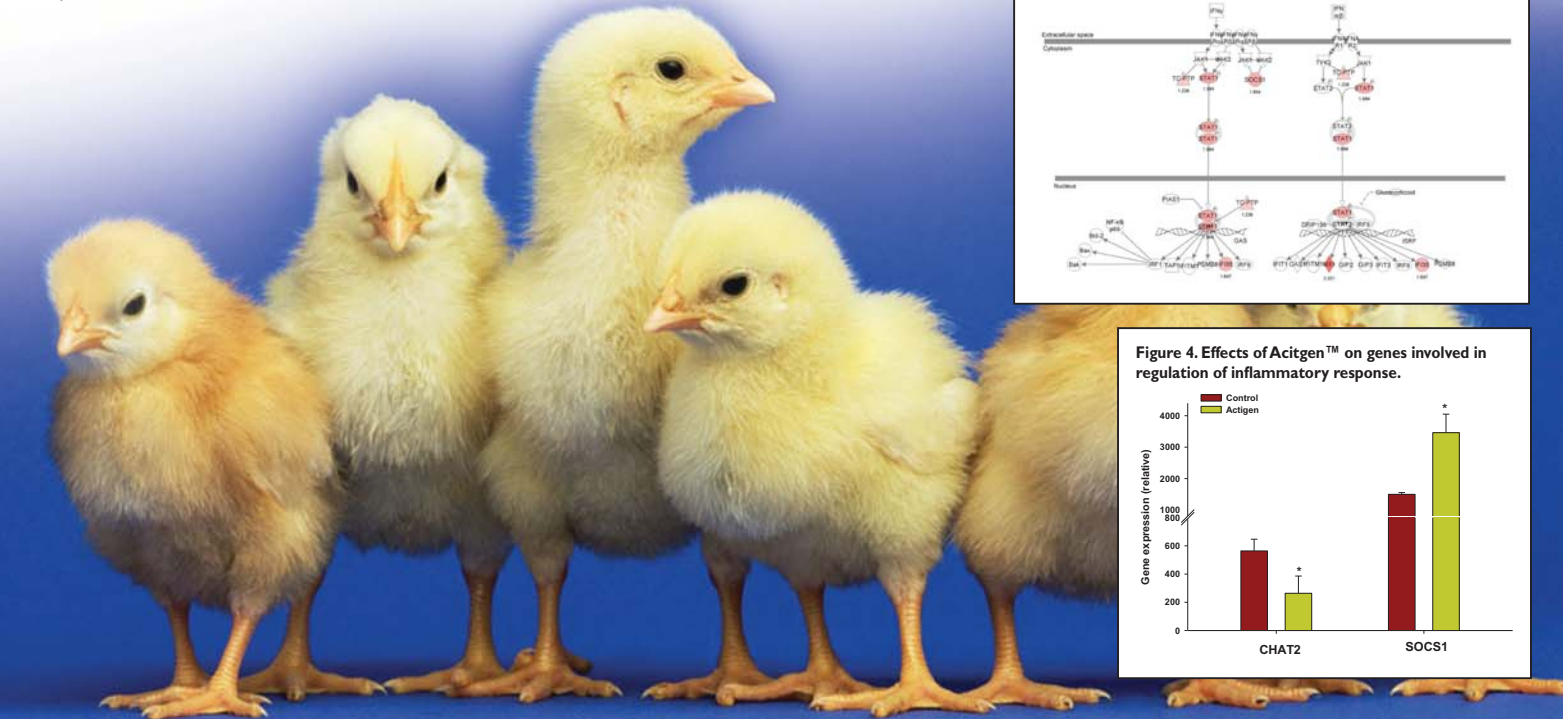
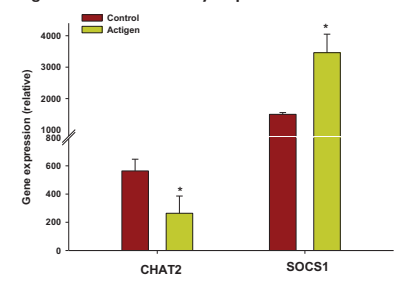


Figure 4. Effects of Actigen™ on genes involved in inflammatory response.



Evaluation of Bioplex[®] and Sel-Plex[®] minerals on broiler performance

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Objective

To evaluate the use of nutritional programs containing organic minerals (Bioplex[®], Alltech Inc.) and selenium yeast (Sel-Plex[®], Alltech Inc.) on broiler performance.

Methods

Location

- Animal Science Department of Universidade Federal de Viçosa, Brazil

Animals and experimental design

- 2,000 Cobb 500 1-d-old chicks
- Completely randomized design
- 8 treatment groups of 250 birds each
- 3-phase feeding program: Starter (1 – 21 d), grower (21 – 35 d), finisher (35 – 49 d)
- Duration of trial = 49 days

Treatments (Table 1)

- T1: Diet with no trace mineral supplementation
- T2: Diet with inorganic trace mineral supplementation
- T3: Diet with Bioplex[®], Sel-Plex[®] (Alltech Inc.) at 120% T4 level
- T4: Diet with Bioplex[®] + Sel-Plex[®] (See Table 1.)
- T5: Diet with Bioplex[®], Sel-Plex[®] at 80% T4 level
- T6: Diet with Bioplex[®], Sel-Plex[®] at 60% T4 level
- T7: Diet with Bioplex[®], Sel-Plex[®] at 40% T4 level
- T8: Diet with Bioplex[®], Sel-Plex[®] at 20% T4 level

Measurements

- Performance parameters included weight gain, feed consumption, feed conversion, viability and productive efficiency index (PEI = Average weight [kg] · (100 - mortality) / (age at slaughter) × (feed conversion) · 100).

Data analysis

- SAEG software was used for the ANOVA (Statistical and Genetic Analysis System – UFV).
- Differences between means were determined to be significant by Dunnett's test at P<0.05.

Results

- Viability was poorest for the unsupplemented (T1) diet: 8.8% (starter), 71.3% (grower) and 100% (finisher). For this reason, it was not used to contrast with the other treatments.
- Performance did not differ (P>0.05) between treatments, except for reduced feed consumption in the starter phase using Bioplex[®] and Sel-Plex[®] at 40% of the T4 level (Tables 2 and 3).

Conclusions

- Bioplex[®] and Sel-Plex[®] can be fed to broilers at 240, 220 and 200 g/ton in starter, grower and finisher phases, respectively, without impairing performance.
- These reduced concentrations represent only 14% (starter), 13% (grower), and 12% (finisher) of current-day inorganic mineral recommendations and thus suggest that Bioplex[®] and Sel-Plex[®] minerals offer increased bioavailability.

Table 2. Effect of Bioplex[®] and Sel-Plex[®] supplementation on performance of broilers (days 1 – 21).

Treatment	Weight gain (kg)	Feed consumption (kg)	FCR	Viability (%)
2	0.846	1.113 ^A	1.32	99.20
3	0.849	1.126 ^A	1.33	98.00
4	0.839	1.094 ^A	1.30	99.60
5	0.837	1.100 ^A	1.31	98.00
6	0.838	1.099 ^A	1.31	99.20
7	0.830	1.073 ^B	1.29	99.60
8	0.831	1.086 ^A	1.31	98.40
CV, %	2.50	2.87	2.14	2.48

^{A,B}Means differ P< 0.05.

Table 3. Effect of Bioplex[®] and Sel-Plex[®] supplementation on performance of broilers (days 1 – 49).

Treatment	Weight (kg)	Weight gain (kg)	Feed consumption (kg)	FCR	Viability (%)	Productive efficiency index
2	3.54	3.49	6.15	1.74	95.2	396
3	3.59	3.54	6.26	1.74	94.4	396
4	3.53	3.49	6.12	1.73	95.6	398
5	3.56	3.52	6.19	1.74	94.8	397
6	3.52	3.47	6.09	1.73	94.8	393
7	3.50	3.46	6.11	1.74	95.6	392
8	3.48	3.43	6.12	1.76	95.6	384
CV, %	2.11	2.14	2.08	1.65	5.07	5.13

References

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- Rostagno, H. S.; Albino, L. F. T.; Donzelle, J. L. et al. Composição de alimentos e exigências nutricionais de aves e suínos: tabelas brasileiras. Viçosa – MG: UFV 2005.
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Table 1. Experimental treatments.

Treatment	Supplement	Dose (kg/t of feed)	Phase	Zn (ppm)	Mn (ppm)	Fe (ppm)	Cu (ppm)	Se (ppm)	I (ppm)
T1	No minerals	0.00	All	0	0	0	0	0	0
T2	Inorganic minerals ¹	1.00	All	90.00	90.00	60.00	10.00	0.40	1.00
T3	Organic minerals ² 120% of T4	1.44	S	57.60	72.00	43.20	8.64	0.26	2.88
		1.32	G	52.80	66.00	39.60	7.92	0.24	2.64
		1.20	F	48.00	60.00	36.00	7.20	0.22	2.40
T4	Organic minerals ² 100% of T4	1.20	S	48.00	60.00	36.00	7.20	0.22	2.40
		1.10	G	44.00	55.00	33.00	6.60	0.20	2.20
		1.00	F	40.00	50.00	30.00	6.00	0.18	2.00
T5	Organic minerals ² 80% of T4	0.96	S	38.40	48.00	28.80	5.76	0.17	1.92
		0.88	G	35.20	44.00	26.40	5.28	0.16	1.76
		0.80	F	32.00	40.00	24.00	4.80	0.14	1.60
T6	Organic minerals ² 60% of T4	0.72	S	28.80	36.00	21.60	4.32	0.13	1.44
		0.66	G	26.40	33.00	19.80	3.98	0.12	1.32
		0.60	F	24.00	30.00	18.00	3.60	0.11	1.20
T7	Organic minerals ² 40% of T4	0.48	S	19.20	24.00	14.40	2.88	0.09	0.96
		0.44	G	17.60	22.00	13.20	2.64	0.08	0.88
		0.40	F	16.00	20.00	12.00	2.40	0.07	0.80
T8	Organic minerals ² 20% of T4	0.24	S	9.60	12.00	7.20	1.44	0.04	0.48
		0.22	G	8.80	11.00	6.60	1.32	0.04	0.44
		0.20	F	8.00	10.00	6.00	1.20	0.04	0.40

S: starter; G: grower; F: finisher

¹ All minerals in the inorganic form. Composition per kg of inorganic mineral mix: 90.0 mg Zn, 90.0 mg Mn, 60.0 mg Fe, 10.0 mg Cu, 400 mg Se and 1.0 mg iodine.

² All minerals were of an organic form with the exception of iodine (potassium iodate). Composition per kg of Bioplex[®] and Sel-Plex[®]: 40.0 mg Zn, 50.0 mg Mn, 30.0 mg Fe, 6.0 mg Cu, 180 mg Se and 2.0 mg of iodine

