

WPSA UK 33rd Poultry Science Symposium
Pre and probiotics; nutritional, veterinary and production perspectives

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Abstract No. 26827:

Mitigating clinical necrotic enteritis with probiotic *Bacillus licheniformis*

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Introduction: Necrotic enteritis (NE) usually leads to poor welfare, reduced profitability and major losses on farm. To help manage the gastro-intestinal challenge, multiple feed additives have been developed, including probiotics.

Aims and Objectives: A commercially available *B. licheniformis*, B-Act[®] was trialled to help mitigate induced NE. The probiotic product is based on a single strain of *B. licheniformis* DSM 28710, already known to support technical performance parameters such as higher final body weights and improved feed conversion ratio's (FCR) in poultry.

Material and Methods: A 42-day induced NE trial was conducted in broilers, with both health and production performance parameters recorded. All animals received the same amount of *C. perfringens* on day 19, 20 and 21 (1 ml/bird, $1.0 \times 10^{8-9}$ CFU *C. perfringens*/ml). The used *C. perfringens* strain was known to have caused NE in a commercial operation. Three treatment groups were evaluated: a negative control (basal diet), a probiotic group (1.6×10^{12} CFU *B. licheniformis* DSM 28710/tonne of feed, start until finish) and an antibiotic group. Animals in this latter group were treated with oxytetracycline hydrochloride (OXT) according to label recommendations, for three consecutive days after the NE challenge.

Results: Even under the induced NE challenge, weight gains of the probiotic and OXT groups were similar to each other and significantly higher compared to the control at the end of the study (2.06 and 2.03 kg vs. 1.8 kg respectively; $P < 0.05$). Weight gain of the probiotic group was already significantly higher compared to the control on day 21 ($P < 0.05$), indicating a potential benefit of the probiotic even before clinical establishment of NE. FCR values followed a similar pattern, with a significantly lower overall FCR for the probiotic and OXT groups compared to the control (1.744 and 1.666 vs. 1.922 respectively, d0-42; $P < 0.05$). From a health perspective, NE lesion scores and NE mortality were both significantly improved in the probiotic and OXT groups compared to the control (0.18 and 0.08 vs. 0.68 respectively; 2.5 and 0.6% vs. 7.8% respectively; $P < 0.05$).

Conclusion(s): This trial demonstrates that feeding probiotic *B. licheniformis* DSM 28710 significantly improved health and growth performance parameters of broilers under a NE challenge. The results achieved with prophylactically administered *B. licheniformis* DSM 28710 were comparable to those realised with the therapeutic antibiotic treatment. Continuous administration of the probiotic thus has the potential to be a useful and practical tool to mitigate NE in commercial broilers, even more so as benefits were already recorded before the NE challenge took hold properly.

Abstract No 26840:

Supplementation of microbial muramidase can improve egg production in high performance laying hens

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Introduction: Exogenous muramidase enzyme (Balancius®) produced by *Trichoderma reesei* has been proven to hydrolyse peptidoglycans (PGNs) from bacterial cell wall debris. Hydrolysis of PGNs with Balancius has great potential to reduce the negative effect of PGNs in the gut of poultry, thus improving gastro-intestinal functionality and performance.

Aims and Objectives: Assess the efficacy of Balancius® to improve growth and egg production performance of laying hens.

Material and Methods: A total of 945 Lohmann brown hens aged 22 weeks old were allocated to 1 of the 3 dietary treatments where Balancius® was added at 0 (T1), 150 (T2) and 300 (T3) g/tonne to a wheat, maize, barley and soyabean meal-based diet to achieve a muramidase content of 0, 150 and 300 mg/kg feed, respectively. Each treatment was replicated 21 times with 15 hens/cage in a randomised complete block design. An enriched colony cage system having 0.24 m² floor area was used. Dietary treatments were fed for 84 consecutive days (12 weeks) in mash form. Both feed and water were available *ad libitum*. Body weight gain (BWG), egg production (EP), daily feed intake (DFI), egg mass (EM), egg weight (EW), and feed conversion ratio (FCR) were determined. The data were analysed using Analysis of Variance (GenStat 19).

Results: The in-feed recovery of muramidase in T1, T2 & T3 was 0, 128 and 303 mg /kg feed, respectively. During 0 to 84 days on trial, birds fed Balancius® at 300 g/tonne of feed had significantly higher BWG (+8%), EP (+1.6%), EM (+2%) and FCR (-1.8%) compared to the T1 control (P <0.05). DFI and EW had no significant differences (P >0.05) when compared between treatments, T2 was similar to T1 & T3. The overall mortality was low (0.52%) and was not treatment associated.

Conclusion: The study suggests that supplementing hen's diet with Balancius® at 300 g/tonne of feed can improve growth and egg production performance, likely due to efficient gut functionality.

Abstract: 26855:

Competitive exclusion as a tool to reduce *Salmonella* carriage: multi-analysis

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Introduction: Competitive exclusion (CE) was described early 1970's as a tool to control *Salmonella* outbreaks in poultry farms.

Aims and Objectives: The objective of this multi-analysis is to combine all published data on the effect that one commercial CE product (Aviguard[®]) (AV) has on *Salmonella*-carriage in chickens

Material and Methods: Twelve publications were identified (reporting results of 31 experiments) comparing *Salmonella* carriage in broilers treated or not with AV. In all studies, animals were challenged with *Salmonella spp* in early life (<10 days – dose ranging from 4 to 9 log CFU/bird). Additional parameters *Salmonella* positive animals (n=7), Infection Factor (IF) and Protection Factor (PF) (n=4) were added to the database when available. Log CFU counts were analyzed using a Mixed Model (SPSS 24.0). The number of *Salmonella* positive birds was analysed using Logistic Regression, IF and PF data using non-parametric tests.

Results: Data showed a significantly lower *Salmonella* carriage in animals treated with AV. For challenged control (CC) an average of 5.5 log CFU/g cecal content was reported. This is 3 log unit higher than the AV treated animals (2.5 log CFU/g cecal content, P<0.01). The odds for CC animals to be *Salmonella* positive are 5.5 higher than for AV birds (P<0.05). IF was numerically lower for the AV-birds (IF-CC: 3.4 vs IF-AV: 1.3) and PF was 14 times higher (PF-CC=1 vs PF-AV: 14.5).

Conclusion: Results confirm that the studied CE product is effective in reducing carriage of *Salmonella* and can be a valuable tool for the poultry industry

Abstract No 26856

Field evaluation of a microbial solutions treatment program on broiler farms

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Introduction: Probiotics have been documented to have positive effects on poultry performance. More recently the benefits of the application of a protective biofilm after disinfection of broiler houses has been studied.

Aims and Objectives: The objective was to evaluate under field conditions the effect a treatment program, combining both microbial solutions, on broiler performance and welfare.

Material and Methods: The trial was carried out in 6 standard broiler sheds (1 farm, 3 consecutive crops, Ross 308). Half the houses were left to the farms' standard practices (standard disinfection, no probiotics) (CTR), in the other 3 houses the treatment program, combining the positive biofilm (mix of selected Bacilli and lactic acid bacteria (Lalfilm Pro)) and probiotic (*Pediococcus acidilactici* via drinking water (Bactocell)), was applied (PROG). Each batch lasted for 35 days. Growth performance and mortality were recorded (per house and per crop). Carcass scoring was performed at the slaughterhouse. Litter samples were taken at day 14 and 35 for dry matter analysis. Data were analyzed using a General Linear Model (SPSS 25).

Result: Average live body weight was significantly improved for the birds from PROG (+4.6%, $P < 0.05$). Dry matter of the litter of PROG houses was significantly higher compared to litter from CTR houses (CTR: 62.59% vs PROG: 67.31%). The improved dry matter content was associated with a significantly lower percentage of birds showing signs of pododermatitis (CTR: 10.6% vs PROG: 2.6%, $p < 0.05$).

Conclusion(s): The treatment program resulted in improved performances of the birds, as well as a better litter quality, resulting in improved animal welfare.

Abstract No 26860:

Identifying differences in tibial ash, morphometric parameters, and mineral profile in 21-day old broiler chicks of varying bodyweights.

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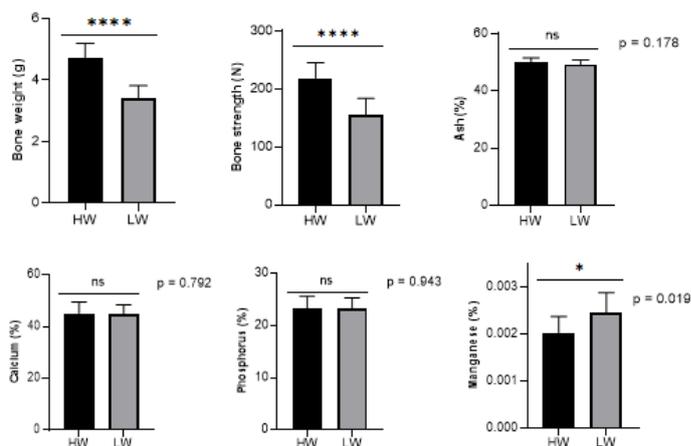
Introduction: In poultry, lack of uniformity constitutes challenges from different viewpoints including growth, health, and welfare. Significant variation in growth was observed in a recent broiler study, even with the same genetic line and management. This variation in growth comprises underweight chicks which fail to reach production targets in the early life. Understanding differences in the biological characteristics between the high and low weight chicks, may provide useful insight into precise strategies to improve uniformity, and growth of the underweight population.

Aims and Objectives: This study aimed at evaluating the differences in tibial ash, morphometric parameters, and mineral profile of high and low weight chicks at 21 days of age. The objective was to obtain insight into drivers of variation in growth and to provide precise solution to the underweight.

Material and Methods: Day old chicks (male Ross 308) were raised in a deep litter pen and fed commercial diet. Ethical approval was granted by Nottingham Trent and University of Nottingham Ethics Committees. Tibial bones were extracted from the high weight HW (n=11; Average BW 1020g) and low weight LW (n=11; Average BW 746g) broilers. Tibial weights (g) were determined using a precision balance, and bone breaking strength was analysed with texture analyser (TA. XT plus 100). Tibias were oven-dried at 105°C for 24hrs and ashed at 600°C overnight to obtain ash content. Mineral profile was analyzed by ICPMS, and statistics was done using the student t-test of GraphPad prism (9.0), significance was determined at (p<0.05).

Results: The bone measurements and mineral profile are shown in fig 1. The HW group exhibited significantly greater bone weight and strength, compared to the LW weight group. No significant differences were observed in bone ash, calcium, and phosphorus contents in both groups. However, manganese content was significantly higher in LW relative to HW group.

Figure 1: Tibial bone measurements, ash, and mineral profile of HW and LW broiler chicks at D21



Conclusion: The HW group had greater bone weight and strength, this may indicate production of bone with optimal dimension for load support. No observed differences were apparent in ash or the two important bone minerals (Ca and P). The significance of the higher Mn content observed in the LW group is still unclear and requires further investigation.

Abstract No 26876:

Gut health markers of the efficacy of probiotic solution *Bacillus subtilis* DSM 29784 in broiler chickens

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Introduction: Biomarkers to evaluate gut health are also difficult to determine with precise accuracy but they are necessary to predict animal performance and to test dietary strategies.

Aims and Objective: The aim of the present work was to evaluate the effect of the probiotic solution *Bacillus subtilis* DSM 29784 (Bs29784) on modulating these markers.

Material and Methods: In a first approach, birds were challenged with a diet high in NSP (20% rye) and a selected group received Bs29784 on top. Gut scoring were performed on all birds as described in Teirlynck et al (2011). Then, in a NE challenge, Bs29784 group was compared to control for histomorphological parameters reviewed in Kraieski et al (2017). In another performance trial on healthy animals, microbiota in samples from cecal content was assessed in both control and Bs29784 groups. Finally, in a pilot study, key metabolites were measured in the intestine of birds receiving Bs29784.

Results: Macroscopical scoring method showed negative correlation with villus length and positive correlation with T-cell infiltration. The Bacterial Enteritis Score was 2.0 for the control group and decreased to 1.0 with Bs29784. On 42 d-old broilers, Bs29784 significantly increased villi length relative to control from 0.66 to 0.79 μm in the ileum, and from 1.25 to 1.46 μm in the cecum. Bs29784 group showed higher alpha-diversity than control birds with Chao and Shannon index. We also observed higher prevalence of *Ruminococcaceae* with the supplementation of Bs29784, especially for *F. prausnitzii* (+13.4%) that is correlated with villus length and CD3 area. We also observed that *E. coli* at 21 d and *C. perfringens* at 28 d were significantly reduced in the Bs29784 group. Finally, 13 day-old chicks showed that Bs29784 increased ($P < 0.01$) the intestinal content of nicotinic acid in ileum and jejunum, revealing a mode of action through which it can improve gut health.

Conclusion: Gut health biomarkers could be used for both research and practical purposes, and can be used to evaluate the efficacy of Bs29784 in broilers.

Abstract 26887:

The link between broiler flock heterogeneity and cecal microbiome composition

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Introduction: Despite low genetic variation of broilers and deployment of considerate management practices, there still exists considerable body weight (BW) heterogeneity within broiler flocks which adversely affects the commercial value.

Aims and objectives: The purpose of this study was to investigate the role of the cecal microbiome in weight differences between animals. Understanding how the gut microbiome may contribute to flock heterogeneity helps to pave the road for identifying methods to improve flock uniformity and performance.

Material and methods: Two hundred eighteen male broiler chicks were housed in the same pen, reared for 37 days, and at study end the 25 birds with highest BW (Big) and the 25 birds with lowest BW (Small) were selected for microbiome analysis. Cecal contents were analyzed by a hybrid metagenomic sequencing approach combining long and short read sequencing.

Results: We found that Big birds displayed higher microbial alpha diversity, higher microbiome uniformity (i.e. lower beta diversity within the group of Big birds), higher levels of SCFA-producing and health-associated bacterial taxa such as Lachnospiraceae, Faecalibacterium, Butyricoccus and Christensenellales, and lower levels of Akkermansia muciniphila and Escherichia coli as compared to Small birds.

Conclusions: Cecal microbiome characteristics could be linked to the size of broiler chickens. Differences in alpha diversity, beta diversity and taxa abundances all seem to be directly associated with growth differences observed in an otherwise similar broiler flock.

Abstract No 27029:

Probiotics strains are more resistant than *Enterococcus cecorum* to the antimicrobial action of botanicals

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Introduction: Botanicals are well-known for their antimicrobial action to inhibit intestinal pathogens in livestock. However, it is not clear whether they could affect also the beneficial microflora or supplemented probiotics.

Aims and Objectives: This study aimed to investigate *in vitro* the inhibitory and bactericidal effect of various botanicals against four probiotic strains (*L. acidophilus*, *L. paracasei*, *L. rhamnosus* and *E. faecium*) in comparison with *E. cecorum* a chicken intestinal pathogen.

Material and Methods: *Lactobacillus acidophilus* (LA), *Enterococcus faecium* (EF), *Lactobacillus rhamnosus* (LR), *Lactobacillus paracasei* (LP) and *Enterococcus cecorum* (EC) were used for this study. Vanillin, thymol, carvacrol, eugenol, α -pinene, limonene and cinnamaldehyde were tested using a microdilution method from 30 to 0.24 mM (2-fold dilutions) for 24 h. Minimal inhibitory concentration (MIC) was considered the lowest dose giving null absorbance. Samples with null absorbance were seeded on agar to assess minimal bactericidal concentrations (MBC) after 24 h.

Results: MIC values against the four probiotics were found for vanillin, α -pinene, limonene (30 mM), eugenol (15 mM), thymol, carvacrol (7.5 mM) and cinnamaldehyde (3.75 mM).

Against EC, MIC values were found for α -pinene, limonene (30 mM), vanillin, eugenol, cinnamaldehyde (3.75 mM), thymol and carvacrol (1.87 mM).

Vanillin, thymol, carvacrol and eugenol killed all the bacteria, with MBC values at least 2 times higher for probiotics than for EC. Cinnamaldehyde was bactericidal only on LA, EF and EC.

Conclusion(s): Probiotics are generally more resistant to botanicals compared to *E. cecorum*, suggesting that selected botanicals at the right concentrations could be used to positively modulate chicken microflora, controlling pathogens without interfering with beneficial bacteria.

Abstract No 27032:

Influence of manganese amino acid complex in layer diets on shell organic matrix composition, eggshell ultrastructure and mechanical property

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Introduction: Manganese is vital in the metabolism of glycosaminoglycans (GAGs) and uronic acid (UA), key components of the eggshell organic matrix.

Aim and Objective: To investigate the effect of Mn sources on eggshell quality.

Methods and Material: 324 Hy-line Brown layers were randomly assigned into 3 dietary treatments with 6 replicates each. From 23 to 46 weeks, layers were fed a Control corn-soybean diet (21,95 mg Mn/kg) or the control diet supplemented with 40 mg/kg Mn as sulphate (MnSO₄) or 40 mg/kg Mn as amino acid complexed (MnAAC). Eggshell ultrastructure analysis, GAGs and UA content were measured at 34 and 46 weeks of age. Eggshell thickness and eggshell breaking strength were evaluated at the end of every 4 weeks. Data were analysed using a one-way ANOVA at P<0.05.

Results: Egg production (23 to 46 weeks) was greatest with MnAAC supplementation followed by MnSO₄ (88.4 ≤ 90.8 ≤ 92.9 %, p=0.016 Control, MnSO₄, MnAAC, respectively). Mn supplementation increased uric acid (3.72 ≤ 4.06 < 4.67 mg/g, 46 weeks, p= 0.007 Control, MnSO₄, MnAAC respectively) and GAGs (2.11 < 2.31 < 2.40 mg/g, 46 weeks, p=0.06, Control, MnSO₄, MnAAC respectively) in the eggshell membrane at 46 weeks of age. At this time, the MnAAC fed group had eggshells with decreased mamillary cone width and improved effective and mamillary layer thickness resulting in greatest resistance to break compared with other treatments (31.7 ≤ 34.3 ≤ 36.94, p= 0.005, Control, MnSO₄, MnAAC, respectively).

Conclusion: Dietary MnAAC improved eggshell quality (breaking strength) by modifying the eggshell membrane organic matrix and the eggshell ultrastructure.

Abstract No 27049:

***Bacillus* sp. PB6 improves performance and alters the microbiome of breeding pheasants under commercial conditions in Poland**

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Introduction: The impact of probiotic supplementation on pheasant breeder performance and microbiome was assessed in pheasant breeders under commercial conditions. Across the trial probiotic supplementation increased daily laying rate, increasing and prolonging peak lay, as well as improved microbiome uniformity amongst individual birds, improved Firmicutes:Proteobacteria ratio in the caecum and significantly altered the caecal microbial composition towards a healthier state.

Aims and objective: The availability of microbiome sequencing technology is especially exciting in less studied poultry species, including pheasants. Therefore, a study was conducted on a commercial pheasant breeder farm to assess the impact of probiotic supplementation on performance and microbiome composition.

Material and Methods: A 12 week study was conducted at commercial pheasant breeding and rearing facilities in Poland. Half the birds on the farm were supplemented with *Bacillus* sp. PB6 at 2 x 10⁸ cfu/kg. Laying data and mortality was recorded daily. Six hens/treatment were sacrificed at the end of the laying period. DNA was extracted from ileal and caecal content and 16S rRNA amplicon sequencing methods were applied to all samples. Alpha and beta diversity measures were calculated, as were differences in relative abundance at the phylum and family level.

Results: Supplementation increased daily laying rate (+5.9%, P<0.05), speed to and height of peak lay, and reduced the decline from peak. Rooster mortality was substantially decreased. The caecal ratio of Firmicutes:Proteobacteria increased following supplementation (1.36 control, 2.35 PB6). Supplementation induced higher uniformity in the distribution of bacterial taxa; compared to the control, fewer individual extremes were observed. Supplementation also numerically increased relative abundances of *Lachnospiraceae* and *Ruminococcaceae* (explained by increased *Faecalibacterium*, which contains only a single species: *F. prausnitzii*, an abundant butyrate-producer), among others.

Conclusions: Application of *Bacillus* sp. PB6 to the diets of breeding pheasants improved performance, likely due to alterations in microbiome structure. The study also provided novel insight into the microbiome of laying pheasants.

Abstract No 27050:

Impact of probiotic supplementation on the microbiome of broiler chickens housed on two commercial farms

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Introduction and Aims: Availability of whole microbiome sequencing technology allows deeper assessment of microbiome shifts following probiotic supplementation and may be useful in elucidating modes of action. This study aimed to identify a probiotic-associated “microbiome fingerprint” in broilers across two commercial broiler farms.

Material and Methods: Half the birds on each farm were supplemented with *Bacillus* sp. PB6 at 1 x10⁸ CFU/l drinking water. Diet and housing differed between the farms. On day 10 and 28, 10 male birds were selected from each group for ileal and caecal content collection. DNA was extracted and 16S rRNA amplicon sequencing methods were applied to all samples. Alpha and beta-diversity were calculated, and differences in relative abundance at the phylum and family level were assessed. P<0.05 was considered significant.

Results: Ileal and caecal alpha-diversity was higher in supplemented birds at d10; ileal beta-diversity on both farms was also significantly different between treatments at d10 and d28. Population shifts with supplementation were consistent across farms. Supplementation increased relative ileal levels of *Lachnospiraceae* (g: *Blautia*), *Ruminococcaceae*, *Enterococcaceae*, and *Leuconostocaceae* (g: *Weisella*); relatively more species of the *Oscillospiraceae* (g: *Flavonifractor*), *Christenellaceae* and *Bacteroidaceae* were seen in supplemented caecal samples.

Conclusions: The study demonstrates the limitations of 16s analysis as, on a genus level many groups (i.e. *Weisella*) contain both beneficial as well as opportunistic pathogens. Optimising specific qPCR to target specific markers could also support which changes are beneficial, neutral or detrimental (e.g. *Candidatus savagella*, *Blautia*) and could identify the most predictable probiotic “fingerprint”.

Abstract No 27051:

Probiotic supplementation reduces mortality by 25% and antibiotic use by >40% in commercially reared pheasants

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Introduction: Probiotics have proven effective in promoting the health of various poultry species, to date there has been limited data on the effect in game birds.

Aims and Objective: This study aimed to look at the effect of probiotics on the growth and health of pheasants.

Material and Methods: The trial was conducted on a commercial rearing farm in Yorkshire. Birds were housed in pens, with access to indoor shelter and a covered range. 24,000 birds were fed control diets, and 42,000 birds were treated with probiotic *Bacillus* sp. PB6 at 2×10^8 cfu/kg feed. 12 birds/pen were weighed at hatch and at catch (at 47 ± 5 days). Cumulative mortality data was recorded per batch at week 1, week 2 and at catch. Medication use data per batch was recorded by the farmer, and data collated and averaged by treatment.

Results: At catch, control birds weighed on average 523 g with supplemented birds weighing 538 g. Relative mortality was reduced by 24.77% following supplementation (5.33% control, 4.01% PB6; $P < 0.05$). Supplemented pheasants received 3.4 courses of antibiotic treatment, with 5 courses given to control birds. Number of days treated was also reduced (23 days control, 14 days PB6, -39.13%). The reduction in antibiotics covered all used types of antibiotics, including amoxicillin, tiamulin and lincomycin. Overall, fewer and shorter **courses** of antibiotics resulted in a 51.87% decrease in total mg of active ingredient used.

Conclusions: Supplementation with probiotics may be a valuable strategy in antibiotic reduction for the game industry, improving performance and bird health.

Abstract No. 27229:

Towards understanding the *stimbiotic* effect of Xylo-oligosaccharides (XOS) based prebiotics on the caecal microbiota

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Introduction: Supplementation of broiler diets with XOS often improves performance via increases in Short Chain Fatty Acids (SCFA) in the caecum in volumes beyond that expected from the amount of XOS added.

Aims and Objectives: This study investigated the effect of feeding Ross308 broilers diets supplemented with XOS on protein expression of caecal microbiota. In the genomes of Bacteroidetes there exists derivatives of a prototypic, *Starch Utilisation System (Sus)* or "*Sus-like systems*" for degradation of complex polysaccharides. XOS acts as a signalling molecule that stimulates upregulation of the proteins of these *Sus-like* system. Hence the aim of this study is to investigate the proposed '*stimbiotic mechanism*' whereby XOS stimulates caecal microbiota to upregulate the proteins involved in the degradation of complex polysaccharides.

Materials and Methods: Bacterial cells from broilers fed a control (CON) or XOS supplemented diet were extracted from the caecal contents. The bacterial pellet was prepared for proteomic analysis using Liquid Chromatography Mass Spectrometry. Protein Pilot search and generation of a spectral library in PeakView (SWATH microapp) revealed 418 unique proteins for SWATH quantitation, of which 382 were quantified.

Results: Heatmap generation and hierarchical clustering showed that the samples clustered clearly between CON and XOS treatment groups. Twenty-nine proteins were significantly differentially expressed with 20 proteins significantly higher in the XOS group of which 9 proteins are part of the *Sus-like* system.

Conclusion: The significant increase in the *SusC-like* and other membrane proteins in microbiota of XOS fed birds indicates that XOS stimulated the Bacteroidetes to upregulate their xylan degrading (*Sus-like*) system.

Abstract No 27227:**Effect of waste stream substrate on the growth rate of larvae: a natural source of antimicrobial peptides for poultry gut health**A. Nourhan¹, E. Burton¹, M. Naeem^{1*}, D. Scholey¹, A. Alkhtib¹¹School of Animal, Rural and Environmental Sciences, Nottingham Trent University, Brackenhurst Campus, Nottingham NG25 0QF, UK.*Corresponding author: naeem.naeem@ntu.ac.uk

Introduction: Insect meal production on sea waste substrates may be an environmentally viable rearing substrate. Insect larvae meal (ILM) is known to contain antimicrobial peptides (AMPs) that could enhance poultry gut health. The viability of ILM as a source of AMPs is depending on establishing economically and environmentally production.

Aims and Objectives: The study examined the effect of differing waste stream substrates on the growth rate of larvae as a gut health enhancer for poultry.

Material and Methods: Mealworm larvae boxes (n=8) were reared on 3 waste stream substrates; food waste, sausage waste and sea waste. Larvae biomass and substrate consumption were used to calculate the mean gain to substrate ratio alongside protein level.

Results: Sea waste substrate increased ($P<0.05$) mealworm biomass gain:substrate (g/g) and crude protein (%) compared to food and sausage waste streams. However, the dry matter (%) was increased ($P<0.05$) on food waste (Table 1).

Table 1: Effect of waste stream substrates on the growth rate of larvae as a gut health enhancer for poultry

Parameters	Treatment (Mean \pm SE)			P-value
	Food waste	Sausage waste	Sea waste	
D0-14				
Mealworm biomass gain (g)	16.06 (0.171) ^c	29.63 (0.509) ^a	24.73 (0.461) ^b	<0.001
Substrate consumption (g)	5.705 (0.416) ^a	5.618 (0.371) ^a	3.2 (0.667) ^b	0.003
Mealworm biomass gain:substrate (g/g)	2.927 (0.229) ^c	5.469 (0.437) ^b	9.233 (1.102) ^a	<0.001
Mortality by weight (%)	136.3 (10.73)	167.3 (13.33)	171.8 (8.29)	0.066
Mealworm dry matter (%)	58.17 (7.075) ^a	44.25 (0.492) ^b	40.57 (0.512) ^b	0.005
Mealworm crude protein (%)	57.22 (0.661) ^b	55.45 (0.253) ^c	63.52 (0.127) ^a	<0.001

^{a-c}: Means with differing superscripts within a row differ significantly ($P\leq 0.05$).

Conclusion: Sea waste appears a viable waste stream substrate to raise the larvae meal. The next step is to assess prebiotic effects.

Abstract No 27226:

Measuring the impact of dietary supplementation with citrus or cucumber extract on chicken gut microbiota using 16s rRNA sequencing

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Introduction: The need to find good alternatives to antibiotic growth promoters is a major challenge for the broiler industry. The presence of active molecules such as pectin, limonene, carotenoids and polyphenols in novel plant extracts, citrus and cucumber, makes them interesting candidates.

Aims and Objective: The main aim is to test the hypothesis that the feed of broiler chickens, supplemented with citrus or cucumber extracts, have a positive effect on gut health through modification of the gut microbiota.

Materials and Methods: A dietary trial (10birds x 4replicates x 3diets) was performed to test the hypothesis that a baseline diet (diet 1) supplemented with citrus (diet 2) or cucumber extract (diet 3) can modulate the gut microbiota. To study the microbiota profile 12 birds per diet were culled at 14 and 28 days of age. The DNA from the jejunum and caeca was extracted and amplified by a 16s specific-PCR, sequenced using Illumina Sequencer and analysed with QIIME™.

Results: Alpha and beta diversity analysis showed that diet 2 and 3 did not significantly affect the bacterial diversity or taxonomy. However, Linear discriminant analysis Effect Size (LEfSe) showed that in the jejunum, diet 2 increased the abundance of beneficial bacteria such as *Lactobacillus* and decreased *Enterococcus* and *Clostridium* at both 14 and 28 days of age. In the caeca, diet 2 increased the abundance of *Blautia* (another potentially beneficial bacterium) and decreased *Enterococcus* in each age group. Diet 3 had no major effects on the microbiota of the jejunum or caecum in either age group.

Conclusion: These results suggest that the use of citrus extract in broilers diets produces subtle beneficial effects on the microbiota which could promote gut health.