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Sensitivity of broiler performance to reused litter exposure and amino acids supplementation.

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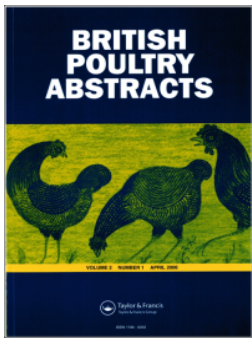
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2020 Abstracts

Oral communications, invited talks and posters accepted for presentation at the WPSA UK Branch Meeting, De Vere East Midlands Conference Centre, Nottingham (31st March and 1st April 2020). Due to the global COVID-19 pandemic in 2020 the Branch meeting was cancelled. These summaries have been edited for clarity and style by the WPSA UK Programme Committee but have not been fully peer-reviewed.

Al-Homidan, I., Ebeid, T., Mostafa, M., Abou-Emera, O., Al-Jamaan, R., & Fathi, M. Effect of probiotic and organic acids supplementation on growth performance and immune response of broiler chicks.

Arthur, C. P., Mansbridge, S. C., Rose, S. P., Kühn, I., & Pirgozliev, V. The effect of graded levels of myo-inositol on apparent ileal mineral digestibility and plasma mineral concentration of broiler chickens at day 21 of age.

Ball, E. E., Smyth, V., Sloss, J., Richmond, A., Donaldson, C., & Nelson, M. Developing a model to test the efficacy of alternatives to antibiotics in broiler diets.

Chowdhury, M. R., Ahmed, J., & Khan, M. M. H. Dietary supplementation of green and black tea waste powder on growth performance and serum biochemical metabolites of heat-stressed broiler chicken.

Dimitrova, K., Gonzalez-Ortiz, G., Mansbridge, S. C., Rose, S. P., Bedford, M. R., & Pirgozliev, V. The impact of dietary mixture of xylanase and xylo-oligosaccharides on energy availability and caecal fermentation when fed to broilers.

Gillespie, P. H., Azhar, R. M., Rose, S. P., & Pirgozliev, V. Does growing site affect the digestible energy of wheat cultivars?

Hillier, A. R., Williams, L. K., Wealleans, A. L., Thijs, L., Wilkinson, T. S., & Humphrey, T. J. The use of organic compounds to reduce *Campylobacter* growth *in vitro* and limit extra-intestinal spread within broiler chickens.

Houdijk, J. G. M., & Walker, R. Does inclusion of faba bean in starter diets allow for greater use of faba beans in grower and finisher diets for broilers?

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Hussein, M. A., Khattak, F., VerVelde, L., Athanasiadou, S., & Houdijk, J. G. M. Sensitivity of broiler performance to reused litter exposure and amino acids supplementation.

Karkelanov, N., Chobanova, S., Dimitrova, K., Rose, S. P., & Pirgozliev, V. Feeding sunflower meal as main protein source to broiler chickens.

Karkelanov, N., Chobanova, S., Dimitrova, K., Whiting, I. M., Rose, S. P., & Pirgozliev, V. Lower non-starch polysaccharides content in sunflower meal gives improved feeding value for broilers.

Kemp, A., Burton, E., & Scholey, D. Identifying which bones indicate skeletal integrity in laying hens.

Kendal, T., Watts, E. S., Rose, S. P., Bedford, M. R., & Pirgozliev, V. Effect of two rapeseed meals with and without exogenous phytase and/or xylanase on dietary phosphorus retention, excreta concentration of inositol and inositol phosphate isomers when fed to broiler chickens.

Liang, J., Dunn, I., McCormack, H., Wilson, P., Fleming, R., Schmutz, M., . . . Maidin, M. Developing a scoring system for Keel bone damage in egg-laying Hens radiographs.

Martin, J. E., Sandilands, V., Brocklehurst, S., Sparrey, J., Baker, L., Sandercock, D. A., & McKeegan, D. E. F. Manual cervical dislocation in poultry: Is it a welfare concern?

Mulvenna, C. C., McCormack, U., MCKillen, J., Bedford, M., & Ball, E. The effect of diets varying in nutrient availability with and without the addition of phytase on broiler performance and bone strength parameters.

Obadire, F. O., Oluwatosin, O. O., Oso, A. O., Adeyemi, O. A., Jegede, V. A., Osofowora, A. O., . . . Pirgozliev, V. Energy metabolisability and ileal digesta viscosity of growing turkeys fed diets containing malted sorghum sprouts supplemented with enzyme or yeast.

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Oyedemi, O. O., Oluwatosin, O. O., Jegede, A. V., Fafiolu, A. O., Pirgozliev, V., & Folorunso, O. Selected egg quality traits of laying chickens fed natural and biodegradable egg yolk colouring raw materials.

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Pirgozliev, V., Enchev, S., Whiting, I. M., Rose, S. P., Kljak, K., Johnson, A. E., . . . Atanasov, A. G. Feeding stevia (*Stevia rebaudiana*) leaves to broiler chickens at two different temperatures.

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Raine, B., Martin, J. E., Mitchell, M., Futro, A., & D'eath, R. Infrared thermography provides an accurate assessment of feather condition in broiler chickens.

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Simic, A., Gonzalez-Ortiz, G., Mansbridge, S. C., Rose, S. P., Bedford, M. R., & Pirgozliev, V. Feeding a combination of xylanase and arabino-xylo-oligosaccharides to young broiler chickens.

Watts, E. S. The effect of batch-to-batch variation and manufacturing plant on apparent amino acid digestibility of commercial rapeseed meal in broilers, with and without protease.

Whiting, I. M., Pirgozliev, V., & Bedford, M. R. Susceptibility of different wheat varieties to enzymatic hydrolysis.

Wilshaw, K. M., Desbruslais, A., Scholey, D., & Burton, E. Determination of optimum post-mortem blood sampling and storage methods for blood antioxidative capacity testing in broilers.

Wilson, M., Scholey, D., Burton, E., & Kemp, A. Determining the effects of housing system on skeletal integrity at both early and mid-lay.

Woods, S., Whiting, I. M., Rose, S. P., & Pirgozliev, V. The effect of saturated and non-saturated fat on growth performance, dietary apparent metabolisable energy and nutrient retention when fed to broiler chickens at two different temperatures.

Effect of two rapeseed meals with and without exogenous phytase and/or xylanase on dietary phosphorus retention, excreta concentration of inositol and inositol phosphate isomers when fed to broiler chickens

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Application

The processing method of rapeseed meal affects its response to dietary phytase.

Introduction

The heat damage incurred during processing of rapeseed meal (RSM) can reduce its feed value in broilers. Research by Watts, Rose, Amerah, and Pirgozliev (2020) demonstrated that by reducing the exposure of RSM to heat during processing its nutritional value in broilers was increased. The aim of this study was to compare in broilers, the nutritive values of RSM produced under standard UK industry conditions (ST) that included 60 minutes desolventizing/toasting (DT), vs RSM that was cold-pressed hexane-extracted (CPhe) and subjected to 80 minutes DT. The efficiency of phytase and xylanase was also assessed.

Material and methods

The experiment was approved by the Harper Adams University Research Ethics Committee. Ninety-six male Ross 308 chicks were reared from 7 to 21 days of age in 48 pens (2 birds in each). Eight diets in total were fed; a basal diet (Control; C) with adequate levels of CP and ME, but lower in available P compared to breeder's recommendations (2.5 vs 4.7 g/kg diet). The C was then split into 2 parts and each of the two RSM samples was incorporated into a diet in mash form at 200 g/kg and fed as they were or supplemented either with phytase (1500 FTU/kg; Quantum BlueTM, AB Vista, UK; QB), xylanase (16,000 BXU/kg; Econase[®], AB Vista, UK; XYL), or with both QB and XYL. Each diet was fed to 6 pens following randomisation. Dietary phosphorus retention coefficient (PR), and the concentration in excreta of inositol (INS) and inositol phosphate isomers

Table 1. Effect of RSM type (ST or M), Phytase (QB) and Xylanase (XYL) on dietary phosphorus retention coefficient (PR), excreta concentration of inositol (INS) and inositol phosphate isomers (IP) when fed to chickens from 7–21 d of age

RSM	Enzyme (units/kg diet)	PR	INS (nmol/g)	IP6 (nmol/g)	IP5 (nmol/g)	IP4 (nmol/g)	IP3 (nmol/g)	IP2 (nmol/g)
ST		0.519	6985	26,075	4587	3811	1359	1515
CPhe		0.557	7768	26,259	4001	3153	1215	1282
QB	0	0.539	4337	39,835	5354	2265	840	1337
	1500	0.537	10,416	12,500	3243	4699	1734	1460
XYL	0	0.543	7858	25,351	4099	3152	1200	1345
	16,000	0.533	6895	26,984	4488	3811	1375	1451
SEM		0.0100	325.9	731.4	124.1	170.6	60.0	44.6
RSM x QB								
ST	0	0.504 ^a	3974	40,256	5932 ^a	2403	861	1471
ST+QB	1500	0.534 ^{ab}	9996	11,895	3241 ^b	5219	1858	1558
CPhe	0	0.574 ^b	4700	39,414	4757 ^c	2127	819	1203
CPhe+QB	1500	0.540 ^{ab}	10,836	13,105	3244 ^b	4178	1610	1361
SEM		0.0141	460.9	1034.4	175.5	241.3	84.8	63.0
Probabilities								
RSM		0.011	0.098	0.860	0.002	0.010	0.097	<0.001
QB		0.879	<0.001	<0.001	<0.001	<0.001	<0.001	0.060
XYL		0.516	0.044	0.123	0.033	0.010	0.047	0.101
RSM x QB		0.032	0.901	0.328	0.002	0.122	0.233	0.580
RSM x XYL		0.914	0.511	0.652	0.662	0.346	0.359	0.706

^{abc}superscript letters denote differences ($P < 0.05$)

(IP) were determined. The data were analysed with ANOVA and in all instances, differences were reported as significant at $P < 0.05$.

Results

The CPhe had higher PR and released less IP5 ($P < 0.05$), compared to ST, although the differences disappear ($P > 0.05$) after QB supplementation. The ST fed birds had a higher IP4 ($P = 0.001$) and IP2 ($P < 0.001$) excreta concentrations. Feeding QB reduced IP6 and increased INS concentration in excreta ($P < 0.001$). Dietary QB also increased the excreta concentration of IP4 and IP3 ($P < 0.001$). Dietary XYL reduced INS but increased IP5,

IP4 and IP3 in excreta ($P < 0.05$). There were no ($P > 0.05$) QB x XYL or RSM x XYL interactions.

Conclusion

Although birds fed the two RSM samples did not have significantly different excretion of INS and IP6, there was an interaction with added phytase that improved PR and IP5 concentration in the standard produced RSM.

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The effect of graded levels of myo-inositol on apparent ileal mineral digestibility and plasma mineral concentration of broiler chickens at day 21 of age

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Application

The interactions with myo-inositol (Ins) and dietary minerals may affect bioavailability of minerals in feed and alter blood plasma mineral concentrations which are important in the formulation of feeds which contain high doses of phytase.

Introduction

The anti-nutritional properties of phytate can result in the chelating of minerals, producing indigestible complexes and reducing the bioavailability of Ca, Mg, Fe, Zn, Cu, Co and Mn. Phytase is routinely used in the poultry industry to primarily release bound phosphorus from phytate, which in turn reduces the chelating of these minerals. Super-dosing phytase in poultry studies is well documented, whereby there is an increase of free Ins into the ileal digesta of broiler chickens. However, the interaction of free Ins in the ileal digesta has on the mineral digestibility is less well known. In addition, if this interaction can then affect the tightly

regulated concentrations of minerals found in the blood plasma of broiler chickens.

Material and methods

A basal mash diet was split and supplemented with one of four levels of Ins, at 0 g/kg, 1.5 g/kg, 3 g/kg or 30 g/kg. Each diet was fed to 10 pens from 0–21 days, with 4 birds per a pen resulting in one hundred and sixty Ross 308 chicks being used in this study. Approval from Harper Adams University Research Ethics Committee. At day 21 one bird per pen was stunned and killed and blood collected in heparinised tubes, remaining birds were also killed and ileal digesta removed. Mineral analysis of blood plasma and digesta was carried out on a NexION® 2000 IPC-MS (PerkinElmer Inc.) according to Cope, Mackenzie, Wilde, and Sinclair (2009). Data was analysed using a one-way analysis of variance in GenStat® (18th edition) with a protected Fisher's least significant difference test to separate means.

Table 1. Effects dietary supplementation of myo-inositol (Ins) on apparent broiler chicken ileal mineral digestibility (%) at day 21 of age

Ins (g/kg)	Treatment							
	Ca	P	Mg	Fe	Na	Co	Mn	Mo
0	53.2	61.7	31.5	28.7	-37.5 ^b	48.6 ^a	54.5 ^a	72.9 ^a
1.5	59.1	64.2	34.4	32.0	-7.2 ^a	40.7 ^b	50.5 ^a	69.7 ^{ab}
3	57.1	62.3	28.1	28.0	-7.9 ^a	26.7 ^d	19.9 ^b	63.5 ^c
30	55.0	65.3	34.4	28.1	4.6 ^a	33.6 ^c	27.1 ^b	66.8 ^{bc}
SEM	1.8	1.0	2.0	1.5	5.6	1.8	2.7	1.3
P-value	0.111	0.080	0.103	0.200	<0.001	<0.001	<0.001	<0.001
CV%	9.9	5.4	19.7	16.2	147.1	15.2	23.1	6.0

^{abc}superscript letters denote differences ($P < 0.05$)

Table 2. Blood plasma mineral concentrations of broiler chickens

Ins (g/kg)	Treatment						
	Ca mmol/L	P mmol/L	Mg mmol/L	Fe μ mol/L	Na mmol/L	Cu μ mol/L	Zn μ mol/l
0	2.9	5.6	0.99	19	150	1.7	23
1.5	2.9	5.4	0.98	19	149	1.7	26
3	2.9	5.5	0.98	22	150	2.0	24
30	2.9	5.3	0.98	19	148	2.8	22
SEM	0.62	0.23	0.038	1.1	2.2	0.55	1.1
P-value	0.996	0.903	0.998	0.174	0.961	0.485	0.174
CV%	6.8	13.0	12.3	17.8	4.7	85.3	14.3

Results

Ins did not affect plasma mineral concentrations of broiler chickens at day 21 ($P > 0.05$) (Table 1). In addition, the mineral digestibility of Ca, P, Mg or Fe was not affected ($P > 0.05$) (Table 2). However, the digestibility of Co, Mn and Mo was significantly reduced by Ins at 3 g/kg and 30 g/kg compared to the un-supplemented diet ($P < 0.05$).

Conclusion

Supplementation of Ins does not effect mineral and trace mineral concentrations in blood plasma at 21 days of age or Ca, P, Mg and Fe digestibility. *myo*-Inositol reduces the

digestibility of Co, Mn and Mo possibly due to the interactions Ins has with these minerals, warranting further research. Increased Na digestibility with greater inclusion of Ins in diets may be explained by Ins transporter proteins (SMIT 1 and SMIT 2) requiring two sodium ions for every Ins molecule transported across the bush border membrane (Walk, Bedford, & Olukosi, 2018).

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Effect of probiotic and organic acids supplementation on growth performance and immune response of broiler chicks

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Application

Using natural feed additives rather than antibiotics can improve final body weight and cellular immunity.

Introduction

Recently, use of probiotic and organic acids as natural growth promoters has gained increasing interest because of the global trend of restriction in use of antibiotics and synthetic substances. Studies on the beneficial impact on poultry performance and immune response have indicated that probiotic supplementation can have positive effects (Ebeid, Fathi, Al-Homidan, Ibrahim, & Al-Sagan, 2019; Fathi, Ebeid, Al-Homidan, Soliman, & Abou-Emera, 2017). Moreover, organic acids had several positive impacts on growth performance, immune response and controlling all

enteric bacteria, both pathogenic and non-pathogenic (Rodjan et al., 2018). The objective of the present study was to evaluate the effect of inclusion of probiotic and organic acids supplementation at different levels on broiler performance and immune response of broiler chickens.

Materials and methods

A total of 1200 day-old mixed sex broiler chicks (Ross 308) were grown over a period of 42 days. Chicks were individually weighed and randomly divided into 6 different dietary treatments. The broiler chicks at one-day of age were housed in pens and divided into 6 (2 probiotic levels (0, 0.2%) \times 3 organic acid levels (0, 0.03, 0.06%)) experimental treatments with 10 replicates of 20 birds (6 \times 10 \times 20 bird). Body weight and feed consumption were determined weekly. Sixty broilers

Table 1. Effect of probiotic and organic acids supplementation on growth performance and immune response of broilers

Item	Probiotic (P, %)		Organic acid (OA, %)			SEM	P-value		
	0	0.2	0	0.03	0.06		P	OA	P × OA
Body weight at 6 wk (g)	2417.8 ^b	2458.7 ^a	2413.2	2448.0	2454.0	14.50	0.01	0.09	0.57
Feed intake (g/bird)	4421.8	4400.8	4373.5	4423.4	4437.1	28.92	0.73	0.66	0.77
Feed conversion ratio	1.89	1.84	1.86	1.87	1.86	0.02	0.14	0.97	0.89
Mortality (%)	2.83	2.33	1.50	3.50	2.80	0.48	0.61	0.24	0.32
Toe web swelling (mm)									
24 h	0.53 ^b	0.65 ^a	0.50 ^b	0.64 ^a	0.63 ^a	0.22	0.01	0.01	0.03
48 h	0.35 ^b	0.42 ^a	0.34 ^c	0.43 ^a	0.39 ^b	0.01	0.04	0.08	0.26
72 h	0.26	0.33	0.26	0.34	0.28	0.02	0.09	0.21	0.60

^{ab} means in the same row having different superscripts differ significantly ($P < 0.05$) across factors

(10 from each treatment group) aged 4 weeks were assigned. After using PHA-P, which was injected into one toe web between the 2 and 3 digits of the left foot to assess cell-mediated immunity. Web thickness was measured with a constant tension caliper pre-injection and at 24, 48 and 72 h post-injection. The web swelling was calculated as the difference between web thickness before and after injection. The care and handling of birds are in accordance with the regulations of animal care committee of Qassim University. Data was subjected to a two-way analysis of variance using SAS software.

Results

The results are summarized in Table 1.

Conclusion

Supplementation of probiotic had a positive significant effect on final body weight. Both probiotic and organic acids improved cell-mediated immunity in broilers in the first 48 h after immune challenge.

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Identifying which bones indicate skeletal integrity in laying hens

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Application

Researchers and practitioners assessing skeletal integrity in laying hens via bone morphology should be consistent when selecting samples, noting a group of bones may be more informative than a singular bone.

Introduction

Due to a high demand for sources of animal protein, the output of a laying hen has increased over recent decades (Whitehead & Fleming, 2000). As a result, the impact of egg production on hen welfare has come under more scrutiny, with pressure to maintain skeletal health alongside high egg yields (Silversides, Singh, Cheng, & Korver, 2012). The aim of this study was to investigate which bones were optimal to assess skeletal integrity in end of lay hens.

Materials and methods

Fifteen Lohmann Brown free-range hens aged 76 weeks, and fifteen Lohmann Brown caged hens aged 79 weeks were collected from two different farms of a large UK egg producer. Birds were culled on collection. Each bird was weighed, and the sample bones were dissected out; keel, humerus, ulna, radius, femur and tibia. Both left and right

bones were taken. Bones were de-fleshed and length, width, weight, height (keel only) and breaking strength were recorded. Length was measured at the greatest distance between each end of the bone and width was measured at the midpoint. Weight was measured using a 4 d.p scale. Height was measured at the greatest distance between the crest and base of the keel. A texture analyser (TA.XT; Stable Micro systems, Guildford) with a 3-point bend rig attachment (HDP/3PB; Stable Micro Systems, Guildford) was used to break the bones at the midpoint and record breaking strength. A Gaussian Generalised Linear Mixed Model (GLMM) was used to investigate housing system, bone type and orientation on bone parameters as a measure of skeletal integrity.

Results

Bone type was a positive predictor of strength (after correcting for size). All bones showed lower strength than the femur, except the humerus. Bone orientation (left or right) did not predict bone strength.

Conclusion

Bone size (measured as bone length) and housing system were reliable predictors of breaking strength. Bone length

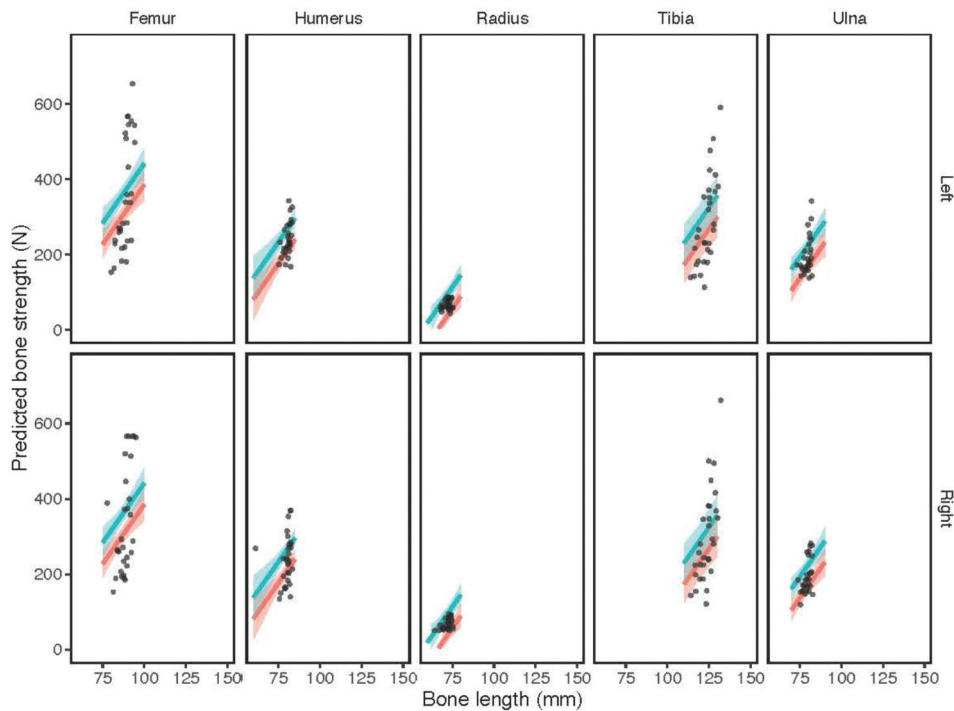


Figure 1. Model predictions for bone type and orientation as a function of size. Red are caged birds and blue free range.

was used in this investigation as an indicator of bird size. In this study bone orientation did not predict bone strength. It has previously been reported that skeletal integrity can be affected by an asymmetry in hen growth (Martinez-Cummer, Hurtig, and Leeson (2006)). A recommendation from this study is to use the humerus, tibia and the keel bone as a set to assess skeletal integrity, as they are easy to remove and provide a good insight into bone formation, as they provide a medullary bone type, structural bone type and calcium reservoir, all of which are effected by egg formation.

Acknowledgments

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Manual cervical dislocation in poultry: is it a welfare concern?

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Application

The current study represents the first to capture a detailed characterisation of high quality continuous intra-cranial electroencephalogram (iEEG) signal for non-anaesthetised broilers, laying hens and turkeys killed by manual cervical dislocation.

Introduction

On-farm despatching of poultry is a necessity for stock-management and to end suffering of sick and injured birds. The methods deployed to despatch birds in Europe, must adhere to the EU legislation, Council Regulation (EC) no.1099/2009 on the protection of animals at the time of killing, which heavily restricts the use of methods dependent

on bird weight and number of birds. The primary change has been the restriction on cervical dislocation methods (both manual and mechanical), supported by research which suggested elongated periods to loss of consciousness, operator fatigue and inconsistency in application (Gregory & Wotton, 1990). However, no previous research had evaluated genuine manual cervical dislocation (MCD). The aim of this study was to perform a detailed assessment of MCD for on-farm despatching of poultry, using key welfare measures (iEEG recording, reflex, behaviour and gross pathology assessment) and evaluate its performance across broilers, layer hens and turkeys and allow a robust assessment of its efficacy and welfare implications in non-anaesthetised birds, which has been previously documented (Martin et al., 2019).

Materials and methods

This work was performed under Home Office (UK) authority and approved by SRUC's Animal Welfare and Ethical Review Body (AU AE 16-2011). A total of 360 mixed sex birds, from three bird types/species (broilers (Ross 308), layers (Hy-Line) and turkeys (Kelly Bronze) were assessed at three stages of development (chick, early juvenile, and late juvenile/slaughter weight/end-of-lay). The four killing methods assessed were (1) cervical dislocation (manual = MCD + mechanical = HST), (2) CASH Poultry Killer (CPK), (3) Armadillo and (4) Rabbit Zinger. Birds were allocated to a killing method based on a Latin square design and bird type/age groups and killing methods were balanced for order of test (9 bird groups x 4 methods x 10 birds/group/method). A subset of 160 birds from all three bird types in the later production stages were allocated for iEEG electrode implantation surgery under general anaesthesia, with a 7-day post-surgery recovery. The efficacy of the devices was determined in three ways: (1) spectral analysis of EEG responses; (2) durations of reflexes post treatment application; and (3) postmortem examination. All data were summarised in Microsoft Excel (2010) spreadsheets and analysed using Genstat (14th Edition, UK) via Generalized Linear Mixed Models (GLMMs). We are only presenting data pertaining to MCD performance.

Results

Overall percentage kill success for each killing method varied, with MCD being the most successful method (100%). For successful kills, the mean (\pm SD) latency to the first interval time (epoch) for $F50 < 6.8$ Hz was shortest for

MCD (3.6 ± 0.2 s) compared to other methods ($P = 0.044$). The pattern of iEEG responses for MCD from baseline through to 30 s post method application demonstrates a level of unconsciousness immediately after method application and this was maintained until brain death (Figure 1). MCD demonstrated a higher performance than HST with the greatest percentage of dislocation levels occurring at C0-C1 compared to HST (80% and 0%, respectively). The severing of either one or both carotid arteries was only observed in 34.7% of birds that underwent MCD and was never observed for birds that underwent HST (0%).

Conclusion

Only MCD and CPK should be recommended for use on farm for depatching poultry. Birds killed with MCD were unconscious within a few seconds of application and ongoing brain activity suggests that in the majority of birds, this persists until loss of brain function and is dependent on both number of carotid arteries severed and the dislocation level.

Acknowledgments

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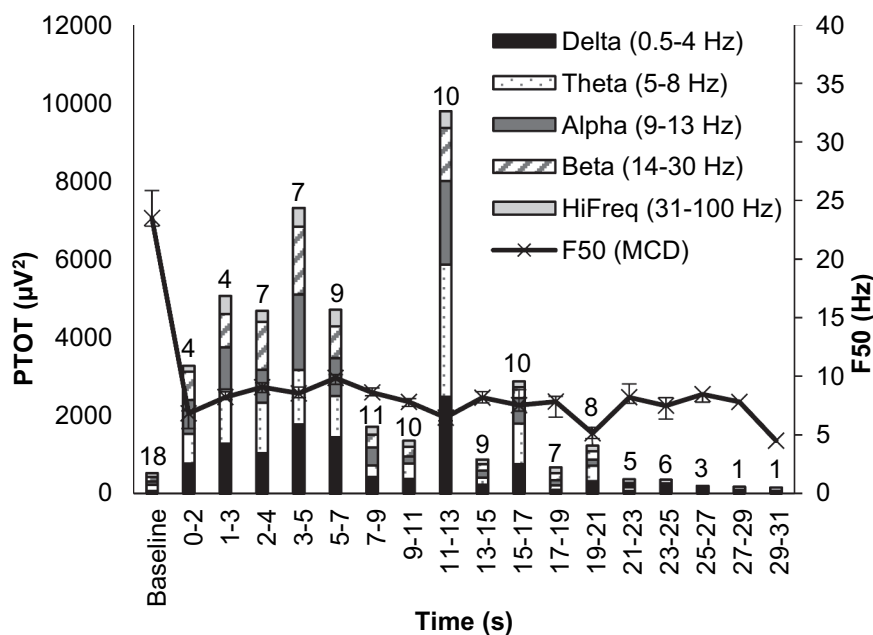


Figure 1. Time series of mean (\pm SEM) total power (PTOT) and median frequency (F50) from baseline, application of manual cervical dislocation, 0s), and every two seconds until 30s post-application. The numbers above the each 2s bar represent the N at each sample point.

The effect of diets varying in nutrient availability with and without the addition of phytase on broiler performance and bone strength parameters

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Application

Cereal based diets result in the best overall performance and final 42d weight. A diet deficient in major nutrient supply with a superdose of phytase can result in similar performance to a diet containing by-products, formulated to bird requirements.

Introduction

Phosphorus (P) losses in the excreta of poultry is a major environmental concern and there is considerable pressure to reduce excretion while maintaining production performance and bird health and welfare. Broiler chickens are unable to hydrolyse phytate-P present in dietary ingredients and traditionally P has been supplied in inorganic form. Exogenous phytase addition to diets can reduce P excretion through improving P utilisation and reducing the need for inorganic P supplementation (Abdel-Megeed & Tahir, 2015). However, the effect of phytase at different levels on diets of differing nutrient specification has not been well established. This study examined the effect of phytase addition (commercial and superdose levels) in diets varying in nutrient supply on growth performance and bone strength parameters of broilers.

Materials and methods

Male Ross 308 chicks (n = 320) similar in weight (40.15g ± 0.28 g) were randomly assigned to pens (10 birds/pen) and provided with one of four experimental treatment diets; positive control 1 (PC1); a cereal based diet + phytase (500 FTU, 0.01%); positive control 2 (PC2), a by-product based diet + phytase (500 FTU, 0.01%); negative control (NC), a by-product based diet without phytase; and NC with reduced nutrient specification plus phytase superdose (NC+, 1500 FTU, 0.03%). Each diet was provided ad libitum to 8 pens with 10 birds (8 reps/treatment). From day 0 to 28 chicks were offered starter formulations of diets (PC1 and PC2: ME = 12.6 MJ/kg, CP = 21.5%, P = 0.74%, Ca = 0.96%; NC and NC+: ME = 12.2 MJ/kg, CP = 20.9%, P = 0.52%, Ca = 0.72%). After which, finisher formulations were provided ad libitum until day 42 (PC1 and PC2: ME = 13.4 MJ/kg, CP = 20%, P = 0.66%, Ca = 0.85%; NC and NC+: ME = 13.1 MJ/kg, CP = 19.4%, P = 0.45%, Ca = 0.61%). Birds and feed were weighed on a weekly basis to determine feed intake (FI), live weight gain (LWG) and feed conversion ratio (FCR) for different periods during the study. On day 42 all birds

were euthanized, two birds from each pen were selected (at average pen weight), the right tibia was removed and used for bone strength analysis (weight (g), length (mm), diameter (mm) and max load (kg)). Analyses were performed using Genstat to conduct ANOVAs on all data recorded. Bone weight and final weight (day 42) were included as a covariate for bone parameters.

Results

For the overall period, birds offered the PC1 treatment performed better in terms of growth rate and final body weight (P < 0.01) than birds offered the other treatments (Table 1). Birds offered the NC treatment had lowest overall LWG and final body weight (P < 0.001). The NC treatment resulted in the worst performance and final body weight (P < 0.001). Adding a superdose of phytase to the NC treatment improved FI, LWG and final body weight to levels equivalent to the PC2 treatment. There was no significant difference in bone strength parameters.

Conclusion

The performance of birds fed cereal based diets with phytase (PC1) was best throughout the study suggesting that by-products such as rapeseed meal and dried distillers' grains with solubles reduced performance. The addition of a phytase superdose improved the FI and LWG and body weight of birds to levels similar to by-product containing diet formulated to adequate nutrient supply (PC2). This has important economic and environmental benefits as cost of diet production can be lowered and overall total P content reduced by superdosing of diets containing by-products and formulated to lower nutrient specifications.

Acknowledgments

This study was conducted within the PEGaSUS project funded by EU ERA-NET SusAn, DEFRA and DAERA. The authors gratefully acknowledge the contribution of AB Vista in diet formulation and study design.

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Table 1. The effects of dietary treatment on broiler performance during starter, finisher and overall periods and bone parameters

Parameter	PC1	PC2	NC	NC +	SEM	P-value
FI (g/d)	4633 ^c	4327 ^b	4072 ^a	4309 ^{ab}	79.7	<.001
LWG (g/d)	3407 ^c	3071 ^b	2908 ^a	3074 ^b	53.8	<.001
FCR	1.36	1.41	1.40	1.40	0.019	0.277
Weight day 28 (g)	1910 ^c	1838 ^b	1656 ^a	1848 ^b	22.7	<.001
Weight day 42 (g)	3447 ^a	3112 ^b	2948 ^a	3114 ^b	53.8	<.001
Bone weight (g)	25.3	23.7	24.0	24.6	0.74	0.544
Bone diameter (mm)	8.83	8.60	8.88	8.80	0.204	0.710
Bone length (mm)	110.0	113.5	113.7	112.2	0.879	0.046
Max load (kg)	42.5	40.5	41.3	40.3	2.38	0.930

^{a, b, c} indicate significant difference P < 0.05

Effect of the duration of the adjustment and collection periods on the estimate of dietary metabolisable energy and nutrient retention in broiler chicken when using indigestible marker in diet

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Application

The duration of the adjustment and collection periods have no impact on the estimate of dietary available energy and nutrient availability coefficients when determined with indigestible marker in diets.

Introduction

There is effect of the duration of collection period on N-corrected dietary apparent metabolisable energy (AMEn), dry matter (MDR), nitrogen (NR) and fat (FR) retention coefficients in chickens, when determined via total collection technique (Stancec, Vrabec, Rose, & Pirgozliev, 2019). However, information on the effect of the duration of adjustment/pre-feeding period is lacking. The reported study evaluated the effect of two adjustment periods, 4 and 8 days, and two collection periods, 48 h and 96 h on AMEn, total tract DMR, NR and FR coefficients of three ad libitum fed mash diets with different energy density in broilers, when determined by using acid insoluble ash as indigestible marker.

Materials and methods

The experiment was conducted at the National Institute of Poultry Husbandry and approved by the Harper Adams University Research Ethics Committee. A basal diet containing 499.5 g/kg of wheat, 235.0 g/kg soybean and 100.0 g/kg of maize, as major ingredients, was mixed. The basal diet was then split into 3 batches and one of them was used as a control (Diet 1), a second lot had 100 g/kg of vegetable oil added (Diet 2), and the third lot had 100 g/kg of soy hulls added (Diet 3). At 19 days old, 144 birds were allocated to 48 raised floor pens (3 birds

in each) and experimental diets were fed. Each of the pens had a solid floor with floor area of 0.36 m² and were equipped with an individual feeder and drinker. At 23 days old, 144 of the remaining birds were allocated to another 48 raised floor pens and experimental diets were fed. A total of 96 pens were involved in this study, and each diet was fed to 32 pens following randomisation. At the start of the collection period when birds were 27 d age, the solid floor of each pen was replaced with a wire mesh and all excreta were collected for 48, or for 96 hours, respectively. Excreta were oven dried, milled and subjected to further analysis. Data were statistically analysed by ANOVA using a 2x2x3 factorial arrangement of treatments.

Results

The determined AMEn and FR of diet 2 were higher ($P < 0.001$) compared to the rest (Table 1). The control diet had higher DMD and NR than the rest ($P < 0.001$). There was no impact of the adaptation or duration of collection periods on any of studied variables. There were no interactions observed ($P > 0.05$).

Conclusion

The results demonstrated that duration of adjustment and collection periods do not influence the estimate of dietary metabolisable energy and nutrient retention coefficients.

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Table 1. The effects of the adaptation and duration of collection period on experimental results

Treatment factor	AMEn (MJ/kg DM)	DMR	NR	FR
Diet				
1	12.15 ^a	0.672 ^a	0.584 ^a	0.708 ^a
2	13.77 ^b	0.635 ^b	0.527 ^b	0.806 ^b
3	10.75 ^c	0.596 ^c	0.557 ^c	0.715 ^a
SEM	0.112	0.0058	0.0075	0.0114
Adaptation (d)				
4	12.24	0.636	0.556	0.743
8	12.20	0.633	0.556	0.743
Collection (d)				
2	12.22	0.632	0.550	0.735
4	12.23	0.637	0.562	0.751
SEM	0.091	0.0048	0.0062	0.0093
P-value				
Diet	<0.001	<0.001	<0.001	<0.001
Adaptation	0.759	0.728	0.990	0.981
Collection	0.977	0.384	0.169	0.232

^{a, b, c} indicate significant difference $P < 0.05$

Infrared thermography provides an accurate assessment of feather condition in broiler chickens

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Application

The current study shows that infrared thermography shows promise as an alternative way of assessing feather condition compared to traditional visual scoring scale.

Introduction

Feather loss is a major problem for broiler chickens and laying hens kept in commercial avian systems. Specific to broilers the feather loss or damage is associated with poor litter quality and ventilation and demonstrates a key welfare concern with birds being vulnerable to heat loss and skin burns and blisters (Haslam et al., 2007). The current most commonly used method of assessing feather coverage is a visual scoring scale from zero (fully feathered birds) to five (completely bald birds), however this is a subjective measure with low inter-observer reliability. Infrared thermography (IRT) measures the surface temperature of an animal and assigns an absolute temperature value to each pixel of the thermal image making it a much more objective method. IRT has been shown to successfully assess feather condition in laying hens however feather score and IRT have not yet been compared in broiler chickens.

Materials and methods

This work was approved by SRUC's Animal Welfare and Ethical Review Body. A FLIR SC620 infra-red thermal imaging camera was used for each image taken. Thermal images of broiler chickens from four different houses across two consecutive crops were taken on one farm. A maximum of ten birds were imaged each week from 5–6 weeks of age per house, resulting in a total sample of 90 images meeting screening quality. Birds

were random selected across 6 sections within each house, with the identified birds placed in a clear space on the litter for image capture. Handling was kept to a minimum to prevent unnecessary stress and gloves were worn to minimum heat transfer. A Kestrel 4000 Weather Meter was used to monitor the environmental temperature and relative humidity and were inputted into the camera settings for each image. A visual feather score for each bird was given (Kretzschmar-mccluskey, Fisher, & Van Tuijl, 2014). The data was imported into ThermaCAM Researcher Professional 2.10 for analysis. From the output the maximum temperature, the temperature range (minimum to maximum) and the standard deviation were recorded in order to look at the variation of temperature across the body of the bird. Average temperature was also recorded and the difference between this value and the ambient temperature for each bird was calculated to correct for environmental temperature changes (DT) (Figure 1). General Linear Models were carried out to determine whether any of the temperature parameters (average temperature, proportion of pixels over 33.5°C, temperature range, maximum temperature or standard deviation of temperature) could be used to distinguish between birds of different feather scores. Ambient temperature was included as a covariate. A one-way analysis of variance was carried out to look at whether DT could distinguish between feather scores.

Results

The mean DT was significantly different for each feather score category as shown in Figure 2 ($P < 0.001$). A Tukey's post-hoc test revealed that this measure can significantly distinguish between each combination of feather score category (1–0 ($P = 0.043$), 2–0

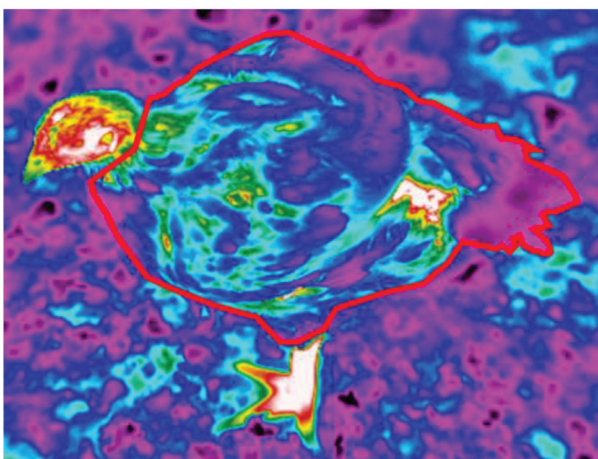


Figure 1. An example of the area (outlined in red) considered as the body of the broiler chicken for thermal image analysis.

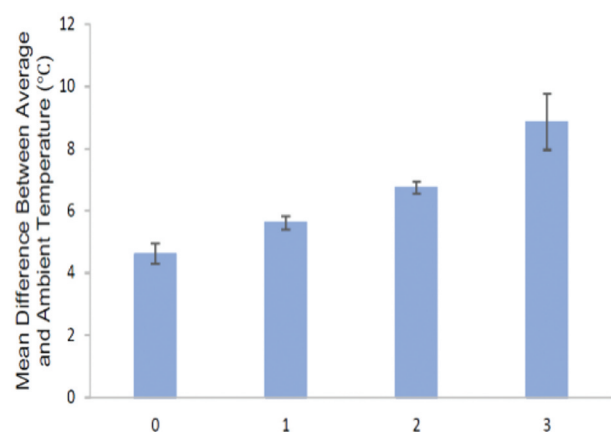


Figure 2. The mean (\pm SEM) temperature difference between the average body temperature and the ambient temperature.

($P < 0.001$), 3-0 ($P < 0.001$), 2-1 ($P = 0.001$), 3-1 ($P < 0.001$), 3-2 ($P = 0.027$).

Conclusion

There was a strong correlation between feather score and DT, as well as between feather score and the proportion of pixels in the thermogram above a threshold temperature of 33.5°C (represents areas of bald skin). The results suggest infrared thermography could be a more objective and reliable alternative used to assess feather condition in broiler chickens.

Acknowledgments

This study was conducted as part of an MSc dissertation in Applied Animal Behaviour and Animal Welfare at the University of Edinburgh.

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Selected egg quality traits of laying chickens fed natural and biodegradable egg yolk colouring raw materials

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Application

Procurement of synthetic dyes used as egg yolk colourants comes at a high cost in Nigeria. Natural sources of egg yolk pigmentation are biodegradable and have been found to be effective at minimum cost.

Introduction

Rossi et al. (2015) asserted that the use of natural yolk colourants in many countries by way of research and usage of pigments of plant origin especially xanthophyll was because of their safety to human health and greater bioavailability. This study evaluated the pigmentation strengths of natural biodegradable egg yolk colouring raw materials in the diets of laying chickens.

Materials and methods

Four potential egg yolk colourants – leaves of *Chromolaena odorata*, *Ocimum gratissimum* were air dried at 27–31 °C for an average of nine days and freeze dried at –8 °C for three days. A total of 420 24-week-old Bovans Nera laying chickens were randomly allotted to seven treatments with each treatment having 60 chickens, using Completely Randomised Design and the experiment lasted for ten weeks. Seven experimental diets were formulated where maize was the main energy source. Egg yolk colourants (EYC) – 15 mg/kg for red and 25 mg/kg for yellow; white maize (WHM) and yellow maize (YLM) diets were the control diets. Air-dried and freeze-dried *C. odorata* (COA and COF) at 25 mg/kg and 21 mg/kg respectively, as well

as air-dried and freeze-dried *O. gratissimum* (OGA and OGF) at 28 mg/kg and 25 mg/kg respectively, were the test diets. Xanthophyll contents of the potential egg yolk colourants were quantified after the leaves were subjected to different drying methods as mentioned above. Data collected included egg weight and yolk colour. Yolk colours were determined by using DSM Yolk Colour Fan. Haugh units were determined based on the formula according to Haugh (1937). Data were subjected to Analysis of Variance for Completely Randomised Design (CRD) using STAR 2.0.1 (v. 2014) as stipulated by Steel and Torrie (1980) and means were separated with Duncan's Multiple Range Test at $P < 0.05$.

Results

Egg yolks from EYC were found to be the richest ($P < 0.05$) in xanthophyll. There was no significant difference ($P > 0.05$) between the yolk colour of eggs from yellow maize diet and the yolk colours from air-dried and freeze-dried *Chromolaena odorata* as shown in Table 1. The equation in the yolk colours for YLM, COA and COF was first noticeable at week 5 of the experiment and this persisted till when the experiment lasted at week 10.

Conclusion

Air-dried and freeze-dried *Chromolaena odorata* included in the diets had a significant effect of deep yolk colouration on the eggs. Therefore, where yellow maize is not available, air-dried and freeze-dried *Chromolaena odorata* can be included

Table 1. Egg quality traits of laying birds fed natural biodegradable egg yolk colouring raw materials

Parameters	EYC	YLM	WHM	COA	COF	OGA	OGF	SEM
Egg Weight (g)	57.48 ^{ab}	55.00 ^{ab}	54.38 ^b	58.47 ^{ab}	59.74 ^a	57.62 ^{ab}	57.26 ^{ab}	1.45
Yolk Colour	14.44 ^a	6.38 ^b	1.22 ^c	3.30 ^b	4.33 ^b	1.44 ^c	1.33 ^c	1.75
Haugh Unit	78.22	74.18	73.88	77.78	82.31	80.68	74.16	5.68

^{a, b, c} indicate significant difference $P < 0.05$

EYC – white maize + egg yolk colourants, YLM – yellow maize, WHM – white maize, COA – white maize + air-dried *Chromolaena odorata*, COF – white maize + freeze-dried *C. odorata*, OGA – white maize + air-dried *Ocimum gratissimum*, OGF – white maize + freeze-dried *O. gratissimum*

in layer chickens' diets because of their high potentials of imparting deep egg yolk colour.

Acknowledgments

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The use of organic compounds to reduce *Campylobacter* growth *in vitro* and limit extra-intestinal spread within broiler chickens

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Application

Campylobacter is a major problem for poultry producers, posing a human health threat and triggering immune responses and energy diversion in the bird. Antibiotics are ineffective control measures; organic compounds with anti-inflammatory potential as feed additives offer potential solutions, but the mode of action is unclear. The data presented in this study provide a foundation for further assessment of anti-inflammatory compounds to control the spread and translocation of *Campylobacter* in poultry.

broiler flocks is of high importance. *Campylobacter* is a frequent member of the intestinal microbiota of chickens and can be pathogenic or commensal in nature (strain dependant). Strains shown to be pathogenic in chickens stimulate an inflammatory response within gut mucosa and compromise the tight junctions in the intestinal epithelium. However, understanding of the mechanisms involved is poor. This study trialled four compounds with known anti-inflammatory properties against *C. jejuni* and *C. coli* isolates to determine the effect on growth of the bacteria. The compounds were applied to avian and human epithelial cell lines to determine if they limit the invasion potential of *Campylobacter*.

Introduction

Contaminated broiler chicken tissues are the primary vehicle for *Campylobacter* infection in humans and thus controlling the spread of *Campylobacter jejuni* and *Campylobacter coli* within

Materials and methods

Growth curves of a collection of *Campylobacter* strains isolated from the liver, ileum or caeca of naturally infected

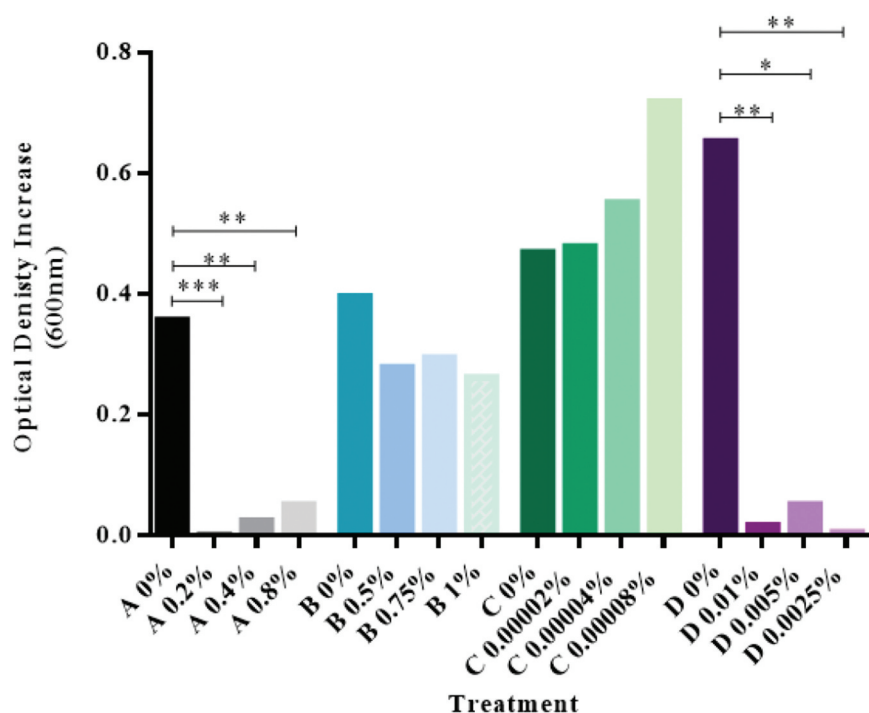


Figure 1. Average increase in optical density (600 nm) of five *Campylobacter* strains (M1, 11,168, G7, C13 & L29) at 24 hours with the addition of varying concentrations of anti-inflammatory compounds. Product A 0.2% shows the most significant reduction in average growth across all treatments ($P > 0.001$). Both product A and D show a significant reduction in *Campylobacter* growth across all treatments. (A = short chain fatty acid, B = medium chain fatty acid, C = mineral compound, D = probiotic strain).

broiler chickens (N = 27) were carried out. These strains were grown over 24 and 48 hours in the presence of four organic compounds at varying concentrations; a one-way ANOVA was conducted to analyse the dataset. Caco-2 and 8E11 epithelial cells lines (human and avian respectively) were cultured using standard protocols. A gentamicin invasion assay (GIA) was conducted using a standard protocol to determine the invasiveness of the *Campylobacter* isolates into the 8E11 and Caco-2 cells. The cells were exposed to the anti-inflammatory compounds and an alamar blue assay undertaken to determine the viability of the cells in each treatment. Following this the cells were treated with the anti-inflammatory compounds at varying concentrations and a second GIA conducted; an ANCOVA was employed to determine the statistical significance of the results from the invasion assays.

Results

Two of the anti-inflammatory compounds significantly reduce the growth of *Campylobacter in vitro*, as shown in

Figure 1. Liver isolates are more invasive than those from the ileum and caeca when applied to both human and avian epithelial cell lines. The addition of anti-inflammatory compounds to 8E11 and Caco-2 cell lines determined the range of concentrations that were used during the second phase of gentamicin invasion assays.

Conclusion

The anti-inflammatory compounds used in this study are shown to decrease the growth rate of *Campylobacter* strains *in vitro*. However, the process is unclear and requires further research. Identifying the way in which these compounds are reducing both *Campylobacter* growth and invasion underpin the next stage of this research. Transwell experiments will be employed to determine the TEER (transepithelial electrical resistance) of healthy cell lines, cell lines exposed to *Campylobacter*, cell lines exposed to anti-inflammatory compounds and cell lines exposed to a combination of both *Campylobacter* and anti-inflammatories to determine the integrity of the monolayer under various treatments.

Developing a Scoring System for Keel Bone Damage in Egg-laying Hens Radiographs

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Application

The development of reliable phenotypes to measure keel bone damage is critical for the implementation of genetic selection strategies to reduce incidence.

Introduction

Egg-laying in hens is associated with osteoporosis – particularly if increased demand for calcium is not matched by sufficient dietary calcium. This increases susceptibility to bone fractures and keel bone damage especially with a move to alternative systems. Current methods of scoring keel bone damage require technical expertise (palpation), lack accuracy, and except for palpation cannot be performed over time. Radiographic analysis of keel bones, potentially in the future in living hens, provides greater accuracy and allows for measurement over time. There is currently only one scoring system developed to assess keel bone damage radiographically (Rufener, Baur, Stratmann, & Toscano, 2018). However, it is subjective, requires standardized training and does not account for specific fracture-level characteristics. This study aims to develop a new radiographic keel scoring system which assesses specific keel bone fracture-level characteristics and can be used as a proxy measure for bone health. Keel bone x-rays of related pedigree egg-laying hens were scored, heritability was calculated and comparison with existing traits were made.

Materials and methods

The new radiographic scoring system consists of two elements: a fracture assessment template and an assessment

guide to aid fracture assessment. The assessment system is designed to assess specific fracture-level characteristics of the keel bone and looks at 8 different characteristics: overall proportion of bone affected, location of fracture, number of fractures, type of fractures, direction of fractures, healing state, overall fracture deformity, and deviation each with a score 1–4. These characteristics were chosen based on their relevance to fracture severity. Depending on the severity of each assessed characteristic, the keel bone is given a numerical Fracture Score (8–32) and Grade (1–6), with higher graded keel bones having more severe fractures. Radiographic scoring was performed on 942 keel bone radiographic images from related hens (Line 44) using the new scoring system. Heritability was calculated using variance components analysis whilst the comparison of traits was carried out by unbalanced ANOVA of keel bone grade scores in GenStat. All studies were on pedigree hens to allow genetic parameters to be calculated, tibia and humerus breaking strength and keel density was performed as previously (Rodriguez-Navarro et al., 2018).

Results

There is a low heritability of Keel Bone Score, sire estimate of heritability was essentially zero. Predictions from the ANOVA for Grade 1 and Grade 6 Keel Bone Scores respectively for keel bone density (measured using aluminium step-wedge as reference) was 0.62 ± 0.006 mm Al equivalent and 0.69 ± 0.044 mm Al equivalent ($P = 0.053$), 183.4 ± 2.9 N and 156.2 ± 13.1 N for humerus breaking strength ($P = 0.004$), 216.2 ± 3.3 N and 170.4 ± 13.0 N for tibia breaking strength

($P < 0.001$), 1633 ± 9 g and 1470 ± 48 g for cull weight ($P < 0.001$), 17.33 ± 0.11 mm and 15.00 ± 0.70 mm for keel bone mid-depth ($P < 0.001$).

Conclusion

Heritability of Keel Bone Grades is poor, however, evidence for keel bone damage increasing keel bone density due to fracture callus may explain this. Results also show an inverse relationship between Keel Bone Grade and tibia and humerus breaking strength – higher graded fractures have lower breaking strengths. This result is encouraging as it shows Keel Bone Grade is related to skeletal health. Reduction in keel bone mid-depth with increasing keel bone grades may be due to damage resulting in keel bone

deviation and reduction in mid-depth. Low body weight can also be a factor in causing increased keel bone damage.

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Dietary supplementation of green and black tea waste powder on growth performance and serum biochemical metabolites of heat-stressed broiler chicken

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Application

Dietary inclusion of green tea waste powder can alleviate the negative effect of heat stress (HS) on growth performance and health status of broilers at 28 days of age.

effect of green and black tea waste powder on growth performance and serum biochemical metabolites in HS broiler chickens.

Introduction

Heat stress can induce reactive oxygen species (ROS) and causes oxidative stress that affects the growth performance and health of poultry species (Hu, He, Arowolo, Wu, & He, 2019). Reduction in growth performance directly related to lower feed intake or impaired nutrient absorption and metabolism (Luo et al., 2018). In addition, oxidative stress may decrease immune functions of broiler chicken and increase the susceptibility to systematic infection by pathogens, leading to a high mortality rate in chickens (Hu et al., 2019). Recent studies have shown that polyphenols such as essential oils and plant extracts can improve growth and immune status in HS broilers. Epigallocatechin gallate is the primary component of tea waste, and it was hypothesised that green and black tea has strong antioxidant properties which will offset the adverse effects of oxidative stress in broiler chicken. Therefore, the present study was aimed to investigate the

Materials and methods

Day-old male Cobb-500 broilers were pre-fed for 10 days, and a total of 240 healthy chicks that were similar in body weight (286 ± 22 g) randomly divided into 5 groups, each group had 8 replicates including 6 birds per replicate and lasted 28 days. Among 5, one group was housed in normal temperature at 28°C with 55% humidity (thermoneutral group, TN), whereas other 4 groups were maintained in controlled temperature with similar humidity at 35°C from 0600 to 1800 h followed by 28°C from 1800 to 0600 h (groups, HS). Birds of TN group were fed with commercial broiler diet provided by Nourish poultry and hatchery Ltd. In contrast, HS birds were received basal diet without (HS-C) or with supplementation of green tea waste powder at 500 mg (HS-G), black tea waste powder at 500 mg (HS-B) or mixture of green (250 mg) and black (250 mg) tea waste powder (HS-M) per kg of diets. All chickens had free access to ad libitum fresh feed and drinking water for 24 h. At the end of the experiment (28 day) after 12 h fasting, live weight gain and feed intake of chickens were recorded for each

Table 1. Effects on tea waste powder supplementation on performance and serum biochemical metabolites of heat stressed (HS) broilers

Parameter	Treatment					SEM	P-value	
	TN	HS-C	HS-G	HS-B	HS-M		TN vs. HS-C	Effects of tea waste under HS
Feed conversion ratio	1.61	2.14 ^a	1.57 ^d	1.71 ^c	1.87 ^b	0.029	<0.001	<0.001
Glucose (mmol/l)	11.4	9.87 ^d	14.3 ^a	12.7 ^b	11.8 ^c	0.37	0.106	<0.001
Total protein (g/l)	37.9	26.9 ^c	46.9 ^a	38.9 ^b	34.6 ^b	1.45	<0.001	<0.001
Cholesterol (mmol/l)	10.4	4.60 ^a	2.33 ^b	3.49 ^{ab}	3.50 ^{ab}	0.280	<0.001	00.006
IgG (g/l)	2.80	1.35 ^d	2.97 ^a	1.92 ^b	1.81 ^c	0.041	<0.001	<0.001
CK (u/l)	3382	8192 ^a	2530 ^c	3623 ^b	4046 ^b	164.7	<0.001	<0.001
AST (u/l)	221	333 ^a	131 ^c	318 ^a	277 ^b	6.0	<0.001	<0.001
LDH (u/l)	2748	4965 ^a	2718 ^c	3888 ^b	4048 ^b	48.3	<0.001	<0.001
Feed conversion ratio	1.61	2.14 ^a	1.57 ^d	1.71 ^c	1.87 ^b	0.029	<0.001	<0.001

^{a, b, c} indicate significant difference $P < 0.05$

replicate and feed conversion ratio (FCR) was calculated. Blood samples were randomly collected from one chicken in each replicate and centrifuged at 1600 g for 10 min at 4°C to separate serum and stored at -20°C for later analyses of serum biochemical parameters. Performance and serum metabolites data under HS were analysed by using Genstat (18th edition) as one-way ANOVA followed by Tukey's test. A comparison between TN and HS-C was also performed as a single degree of freedom to evaluate the effect of HS on basal diet without supplementation.

Results

Heat stress reduced FCR, serum levels of total protein, immunoglobulin G (IgG) and serum cholesterols, and increased the activities of serum creatinine kinase (CK), aspartate aminotransferase (AST) and lactate dehydrogenase (LDH) compared to TN group ($P < 0.05$) (Table 1). Under HS, dietary inclusion of green tea waste powder increased FCR, IgG, the serum concentration of glucose and total protein; and normalised blood cholesterol, CK, AST, and LDH enzyme activities.

Conclusion

The changes in serum metabolites content and enzyme activity might have been due to HS induced oxidative damage. A comparison between dietary inclusion of green and black tea waste powder revealed that the former has a stronger effect on nutrient metabolism and enzymatic activities in broiler chicken by alleviating oxidative stress.

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Effects of threonine, arginine and glutamine supplementation on caecal microbiome profiles and litter characteristics of broilers exposed to reused litter

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Application

Under conditions in which exposure to reused litter improved bird performance, amino acids supplementation modified selected gut health indices in support of its detrimental effect on performance.

Introduction

Threonine, arginine and glutamine (TAG) supplementation has been associated with improved performance during coccidial challenge (Gottardo et al., 2016). In contrast, we observed reduced performance upon TAG supplementation in the presence of reused litter (Hussein, Khattak, Vervelde, Athanasiadou, & Houdijk, 2020), whilst raising on reused litter improved performance. Here, we hypothesise that caecal and litter based gut health indices taken from that study are sensitive to TAG supplementation and reused litter exposure.

Materials and methods

A total of 320 Ross 308 day-old male broilers were allocated to 32 pens (10 birds per pen). Using a 2 × 2 factorial arrangement, birds were fed either control diets (C) or C + TAG and were placed on either fresh (clean) or one-month-old litter (reused), recycled from a previous flock and positive for avian pathogenic E.coli. The TAG treatment consisted of feeding threonine and arginine at 25% above required, and 1% glutamine. Birds were fed ad libitum a starter mash (0–11d) and a grower mash (12–

21d) rations, formulated to meet Ross 308 requirements based on the ideal protein concept. This study was approved by SRUC's Animal Experiment Committee. Body weight gain, feed intake and mortality corrected feed conversion ratio were assessed and reported elsewhere (Hussein et al., 2020). On d 21, 2 birds per pen were euthanized and their pooled caecal contents assessed for microbial profiling using the guanine+cytosine methodology (% G + C profiling) and short-chain fatty acids (SCFA) concentration and composition. Litter samples were collected on d21 and analysed for pH and moisture content. Data were analysed via analysis of variance to assess the effects of diet, litter and their interactions. Significant interactions were compared using Tukey-adjusted multiple comparisons. Results were considered significant at the level of $P < 0.05$.

Results

The concentration of total SCFA did not differ between treatments ($P = 0.515$) and averaged 96.83 ± 8.15 mM. Table 1 shows that the percentage of propionic acid was increased for TAG supplemented birds placed on clean litter but reduced for birds on reused litter. Also, it shows that TAG supplementation for birds placed on clean litter increased the percentage of branched-chain SCFA to a greater extent than for birds on reused litter. TAG supplementation lowered the dominant caecal G + C peak (48 to 54% G + C region) for birds on clean litter only ($P < 0.05$). Reused litter pens had higher pH and lower moisture levels

Table 1. Effects of AA supplementation on gut health indices of broilers, 21 d after placing on clean or reused litter

Treatments		SCFA (%)					G + C (%)	Litter analysis	
Litter	Diet	Acetic acid	Propionic acid	Butyric acid	Lactic acid	Branched SCFA	48–54	pH	Moisture (%)
Clean	C	72.45	3.98	13.34	10.07	0.101	4.24	6.67	30.00
	TAG	71.32	5.67	12.88	9.79	0.315	3.90	6.57	27.36
Reused	C	73.99	6.35	10.45	9.08	0.117	4.16	7.35	25.10
	TAG	72.26	3.65	11.79	12.14	0.155	4.10	7.40	22.78
SED		2.06	1.05	1.55	1.80	0.052	0.06	0.07	1.67
P value									
Diet		0.337	0.508	0.690	0.286	0.003	<0.001	0.707	0.041
Litter		0.404	0.819	0.083	0.597	0.064	0.138	<0.001	<0.001
DietxLitter		0.838	0.008	0.418	0.204	0.025	<0.001	0.172	0.894

than clean litter pens, whilst TAG supplementation reduced litter moisture.

Conclusion

These data support the view that birds fed control diets and exposed to reused litter had increased caecal propionic acid, which through gluconeogenesis may have contributed to the better performance that has been reported elsewhere (Hussein et al., 2020). On the other hand, AA supplementation under these unexpected effects of reused litter exposure showed a detrimental impact on caecal microbial profiling and elevated levels of branched-chain SCFA. These are indicators of surplus protein being fermented and may be a basis for the reduced performance observed upon TAG supplementation (Hussein et al., 2020).

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The effect of batch-to-batch variation and manufacturing plant on apparent amino acid digestibility of commercial rapeseed meal in broilers, with and without protease

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Application

Variability in the apparent ileal amino-acid digestibility coefficients of UK produced, commercial rapeseed meal, is low when fed to broilers.

Introduction

Rapeseed meal (RSM), a by-product of the oil crushing industry, is routinely used in as a protein source in poultry feed. There are currently two main, large-scale crushing facilities (CF) in the UK that produce RSM. The raw material they use originates from multiple European locations that can differ in environmental conditions and crop management practices. These factors as well as differences in the processing specifications unique to each facility can influence the apparent ileal amino-acid digestibility coefficients (AID) of the end product. To develop more accurate nutrient

matrices for feed formulation, robust information on this variability is vital.

Materials and methods

All procedures were approved by The Animal Experiment Committee of Harper Adams University, Shropshire, UK. Eight different commercial rapeseed meal (RSM) batches were obtained at fortnightly intervals from two primary UK crushing plants (plant A and plant B). Eight experimental diets containing titanium dioxide were mixed in mash form to contain 800 g/kg of a wheat/soybean meal based basal feed and 200 g/kg RSM. Half of each RSM diet and a basal control diet were supplemented with commercial mono-component protease (MCP) (intended activity 4000 units per kg). From day-old 432 male broiler chicks were reared according to breeders' recommendations. On 13 d age

Table 1. The effect of rapeseed meal batch, crushing facility and protease of the apparent ileal digestibility of total, dispensable and indispensable amino acids. AA: apparent ileal digestibility coefficients of amino acids; Tot: total; Disp: dispensable; Ind: indispensable; RSM: rapeseed meal; CF: crushing facility; MCP: mono-component protease

Treatments		Tot aiAAd	Dis aiAAd	Ind aiAAd
RSM batch	A2	0.649	0.625	0.666
	A3	0.646	0.619	0.664
	A4	0.660	0.631	0.681
	A5	0.605	0.574	0.659
	B1	0.672	0.647	0.689
	B2	0.645	0.615	0.666
	B3	0.608	0.578	0.630
	B4	0.599	0.552	0.631
	SEM	0.037	0.035	0.038
CF	A	0.640	0.612	0.668
	B	0.631	0.598	0.654
	SEM	0.018	0.017	0.019
MCP	YES	0.647	0.615	0.671
	NO	0.624	0.596	0.651
	SEM	0.018	0.017	0.019
P value	RSM	0.789	0.497	0.949
	CF	0.730	0.553	0.615
	MCP	0.369	0.439	0.479
	RSM*MCP	0.698	0.699	0.737
	CF*MCP	0.369	0.591	0.389

birds were randomly transferred in groups of four to raised floor pens and randomly allocated to one of 18 diets which were fed *ad libitum* along with fresh water. On the final day (21 d age) ileal digesta were collected for determination of marker by spectrometer and amino

acids (Amino Acid Analyser INGOS AAA 400). The studied variables were compared statistically on Genstat by randomized block ANOVA using pre-planned comparisons. Differences were reported as significant at $P \leq 0.05$.

Results

No significant interactions were found between the main treatment factors ($P < 0.05$). The results (Table 1) showed no significant main effect of crushing facility or RSM batch on the coefficients of AID. No significant effects of MCP were observed on total AID. There were tendencies towards an effect of MCP on the AID of dispensable and indispensable AA ($P < 0.1$). The coefficients of variation for the AID of total, indispensable and dispensable AA were 3.7, 4.2 and 5.1, respectively.

Conclusion

The AID data demonstrates that variability between batches of commercial RSM produced within and between the UKs two main crushing facilities is low. The uniformity observed suggests that both the processing specifications of the facilities and the raw materials they use are similar. On this occasion supplementation with this particular MCP had no significant effect on the AID of total AA however the AID of both dispensable and indispensable AA increased. Further research is needed to determine its effect on individual AA.

Influence of combined cow and camel rumen content on some haematological and serum biochemical indices of finisher broiler chickens

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Application

Combined Rumen Content of cow and camel can alternatively be used to replace maize if properly processed without posing any health risk with better performance of broiler chicken.

Introduction

Poultry production is one the promising sectors in developing country like Nigeria. Recently, there is a tremendous setback in this sector as a result of increasing cost of protein and energy feedstuffs especially maize which from the bulk of energy in poultry feeds with the staple food-feed competition existing between livestock and man. This scenario necessitated the exploration into cheap, unconventional feedstuffs such as cow and camel rumen content waste. Rumen content is an abattoir waste that can offer a cheap, viable protein and substantial utilizable energy to maize which has become very expensive in Nigeria Dairo, Aina, and Agafa (2005). Previous findings showed that addition of either cows (Gebrehawariat,

Animut, Urge, & Mekasha, 2016) or camel (Makinde, Abdullahi, & Mohammed, 2017) revealed better performance at 15% for poultry birds. An experiment was therefore carried out to investigate the effect of combined rumen content on blood parameters fed to broiler chickens.

Materials and methods

Diets were formulated such that camel and cow rumen content (CCRC) at ratio 50:50 were used to substitute maize in graded levels 0, 20, 25, 30 and 35% in practical broiler diets formulated according to the recommendations of NRC, 1994 for broilers finisher in the tropics. The levels of all other feed ingredient remained unchanged. Five diets were formulated such that the CCRC was partially included at levels: 0, 20, 25, 30, and 35% respectively. Thirty 56d Marshall Broiler chickens were divided into 5 groups of 6 birds each. At the end of the experiment, 6 birds per treatment were randomly selected, fasted over night from which blood samples were collected and used for haematological and serum biochemical measurements. Haematological

Table 1. Effect of combined cow and camel rumen content on some Haematological and serum biochemical parameters of finisher broiler chickens. CCRC = combine camel and cattle rumen content. PCV = packed cell volume WBC = white blood cell, ALT = alanine aminotransferase and AST = aspartate aminotransferase

Parameters	0%	20%	25%	30%	35%	P value
PCV (%)	33.75	33.70	33.10	34.75	37.25	0.12
WBC ($\times 10^3/\text{mm}^3$)	233.25	241.55	239.65	239.65	253.75	0.13
Serum Albumin (g/l)	15.50	17.50	14.50	15.50	16.50	0.11
Uric acid (mmol/l)	2.07	2.07	1.57	1.57	1.95	0.73
Creatinine(mmol/l)	45.50	44.00	41.00	39.00	42.50	0.21
AST (u/L)	323.50	279.00	280.50	290.50	291.00	0.16
ALT (u/L)	179.83	167.00	168.00	163.33	165.00	0.09

samples were collected into sample tube containing ethylene Diamine Tetra-acetic acid (EDTA). Haematological parameters analyzed were packed cell volume PCV, WBC were determined according to the methods of Bush (1991). Serum was obtained after the blood in the anticoagulant-free tubes were allowed to stand for two hours at room temperature and centrifuged at 2000 revolution per minute for 10 minutes. Serum albumin, uric acid, creatinine, alanine aminotransferase ALT and aspartate aminotransferase (AST) were also determined.

Does inclusion of faba bean in starter diets allow for greater use of faba beans in grower and finisher diets for broilers?

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Application

Faba beans may be included up to 10 to 15% in grower and finisher rations, independent on whether starter rations contain 5% of faba beans or not.

Introduction

Home-grown protein sources have great potential to reduce reliance on soya bean meal (SBM) provided that potential constraints on nutritional value and intake potential can be accounted for. We previously assessed the nutritional value of Scottish grown faba beans (FB) in broilers and observed that the inclusion of 20% FB at the expense of SBM in iso-nitrogenous diets tended to reduce performance in young birds (Olukosi, Walker, & Houdijk, 2019). It might be considered that this arose from over-exposure to FB during the starter phase. We therefore hypothesised that growth response to FB inclusion in grower and finisher rations may be sensitive to inclusion of a small amount of FB in starter rations.

Materials and methods

A total of 800 Ross 308 day-old male broilers were allocated to 80 pens (10 birds per pen) and were subject to one of 10 feeding treatments ($n = 8$ pens in a randomized block design). Two starter treatments (0 and 5% FB) were

Results

All the haematological and serum bio-chemical parameters measured were not significantly ($P > 0.05$) affected by the treatment imposed on the birds (Table 1).

Conclusion

It was concluded that CCRC at 35% inclusion levels could be used to substitute maize in the diets of finisher broiler chickens as it does not pose any health risk to the chicken based on the haematological serum and biochemical parameters measured.

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factorially combined with five grower/finisher treatments (0, 5, 10, 15 and 20% FB). The diets were iso-nitrogenous and wheat-SBM based, with FB being exchanged against SBM based on digestible lysine levels, formulated to meet Ross 308 requirements, using pure amino acids as required and allowing for small variations in wheat and oil to maintain similar levels of AME. Birds were fed ad libitum the starter (0–11d; coarse mash), grower (11–25d; pellets) and finisher (25–34d; pellets) rations. Diets did not contain coccidiostats. Bird weight gain, feed intake and mortality corrected feed conversion ratio were determined for each feed phase; data reported here is for the d0 to d34 phase only. Data were analysed via 2×5 ANOVA for starter treatments (0 vs 5% FB), grower/finisher treatments (0, 5, 10, 15 and 20% FB) and their interaction.

Results

Table 1 shows that inclusion of 5% FB in the starter ration reduced weight gain and feed intake by 3.0% each, and thus not affecting feed conversion ratio. The absence of FB in grower and finisher rations unexpectedly resulted in reduced performance. Weight gain and feed intake increased, and feed conversion reduced, as FB increased to 10%. A further rise to 15% resulted in no further improvement of performance, whilst there was small reduction in performance as FB levels increased from 15% to 20%.

Table 1. Effects of faba bean (FB) inclusion levels in starter (S) and/or grower/finisher (GF) rations on broiler performance (d0-d34)

FB in GF	FB in S					
	Weight gain (g/bird)		Feed intake (g/bird)		FCR (g/g)	
	0 (%)	5 (%)	0 (%)	5 (%)	0 (%)	5 (%)
0 (%)	1908	1881	3108	2973	1.526	1.476
5 (%)	2351	2258	3443	3359	1.362	1.390
10 (%)	2349	2307	3407	3405	1.358	1.380
15 (%)	2221	2255	3285	3335	1.377	1.381
20 (%)	2361	2140	3518	3176	1.389	1.387
<i>s.e.d.</i>	74		105		0.027	
<i>P-values</i>						
S	0.040		0.033		0.966	
GF	0.001		0.001		0.001	
S*GF	0.171		0.095		0.265	

Conclusion

This data supports the view that FB may be included up to 10–15% in grower and finisher rations to reduce dependency on SBM, independent on whether starter rations contain 5% of FB, though the latter did slightly reduce performance as a whole. The reduced performance in the absence of FB was unexpected, though it cannot be excluded that this may indicate that FB exert some gut health benefits otherwise obtained through coccidiostats, which were here omitted. Also, whether the use of starter diet as a mash rather than a crumb contributed requires further study.

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Sensitivity of broiler performance to reused litter exposure and amino acids supplementation

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Application

The ideal amino acid profile for broilers would not need to be reconsidered if a purported challenging condition does not penalize their performance.

Introduction

When broilers are exposed to immune challenges, e.g. those arising from (reused) litter, amino acid (AA) digestibility may reduce and/or AA demand may increase, e.g. for immune responses (Runge, Blackall, & Casey, 2007). Threonine (T) is the third limiting AA after lysine and methionine in conventional poultry diets and is required for mucin synthesis as part of the intestinal immune function, inhibition of apoptosis, stimulation of lymphocyte proliferation and enhancement of antibody production (Zarrin-Kavyani, Khatibjoo, Fattahnia, & Taherpour, 2018). Also, arginine (A) and glutamine (G) supplementation may have beneficial effects on performance and immune function under normal rearing conditions (Murakami, Fernandes, Hernandez, & Santos, 2012). However, effects of dietary supplementation of these AA, alone or in combination (TAG), on resilience, defined as the ability of birds to limit the pathogenic impacts on performance, are not well

understood. Therefore, we investigated the effects of T, A and G supplementation on growth performance and internal organs weight in broilers raised on clean or reused litter.

Materials and methods

A total of 800 Ross 308 day-old male broilers were allocated to 80 pens (10 birds per pen). There were 5 dietary treatments (control (C), C + T (25% above required), C + A (25% above required), C + G (1% of G) and C+ TAG (at the same levels at T, A, G individually) and were placed on either fresh (clean) litter or one-month-old litter (reused) that was recycled from previous broiler study. Each diet-litter treatment combination had 8 pens in a randomized block design. Birds were fed ad libitum a starter mash (0–11d) and a grower mash (12–21d) rations, which were formulated to meet Ross 308 requirements based on the ideal protein concept. This study was approved by SRUC's Animal Experiment Committee. Body weight gain (BWG), feed intake (FI) and mortality corrected feed conversion ratio (FCR) were assessed. On d 21, 2 birds per pen were individually weighed, euthanized and empty gizzard, bursa and spleen weights were determined as a percentage of body weight. A 5 × 2 factorial analysis of variance was used to

Table 1. Effects of amino acid (AA) supplementation on growth performance (0–21d) and internal organs weight of broilers reared on clean or reused litter

Parameter	Clean litter					Reused litter					P-values			
	C	T	A	G	TAG	C	T	A	G	TAG	SED	diet	litter	diet* litter
BWG (g)	741	719	721	705	668	767	762	738	737	714	25	0.013	0.004	0.913
FI (g)	1047	977	1011	960	919	1017	993	967	1007	952	25	<.001	0.657	0.048
FCR (g/g)	1.45	1.39	1.44	1.39	1.42	1.35	1.33	1.34	1.39	1.37	0.05	0.786	0.004	0.497
Gizzard (%)	2.35	2.37	2.37	2.40	2.68	2.28	2.13	2.25	2.32	2.37	0.11	0.009	<.001	0.447
Bursa (%)	0.21	0.21	0.20	0.22	0.20	0.25	0.21	0.24	0.22	0.21	0.02	0.416	0.033	0.198
Spleen (%)	0.10	0.09	0.09	0.08	0.09	0.09	0.10	0.09	0.08	0.09	0.01	0.294	0.981	0.512

assess the effects of diet, litter and their interactions. Significant interactions were compared using Tukey's test.

Results

Table 1 shows that diet and litter treatments interacted for FI, with TAG supplementation reducing intake on clean litter only. Birds raised on reused litter had greater BWG (+4.6%) and better FCR (−4.3%) than those placed on clean litter. Litter and diet did not interact on organ weights. However, birds raised on reused litter had lower relative weights of gizzard and higher relative weights of bursa than those on clean litter. Furthermore, birds supplemented with TAG had a significant higher relative gizzard weight than those supplemented with T.

Conclusion

This study demonstrated that exposure to reused litter may benefit bird performance and improve the weights of the gizzard and immune system-related organ (bursa). Under these conditions, the impact of AA supplementation to

ideal protein formulated conditions on resilience could not be tested. An assessment of gut health indices from this study may elucidate the positive effect of reused litter and the absence of beneficial AA supplementation on bird performance.

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Feeding sunflower meal as main protein source to broiler chickens

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Application

Dietary Sunflower meal can substitute soybean meal in broiler diets.

Introduction

Price volatility in the soybean meal (SBM) market forces the use of alternate protein sources in poultry diets. Sunflower meal (SFM) is a widely available and valuable protein source that may offer a sustainable alternative to SBM. The relatively low metabolisable energy (ME) and lysine, and high non-starch polysaccharide (NSP) contents of SFM limits its inclusion in broiler feed (Rama Rao *et al.*, 2006). The objectives of the study were to evaluate the effect of substituting soya bean meal (SBM) with SFM, with some lysine adjustment, when fed to broiler chickens from 0 to 42 days of age on daily growth performance variables including feed intake (FI), weight gain (WG), feed conversion ratio (FCR) and average bird weight at 42d age.

Materials and methods

The experiment was approved by the Animal Experimental Committee of Trakia University, Stara Zagora, Bulgaria. A sample of SFM was obtained (Tivatrade, Stara Zagora, Bulgaria) that contained 456 g/kg crude protein (CP), 3.8 g/kg crude fat (CF) and 164 g/kg NSP, from which 54 g/kg soluble. A proprietary control broiler diets, including starter, grower and finisher were mixed containing SBM but not SFM. To produce the rest of the diets, SBM was replaced with 29%, 66% or 100% SFM. All four dietary treatments were balanced to contain the same ME, total crude protein, lysine, methionine, threonine and tryptophan, and the major minerals in the starter, grower and finisher phases. Day-old male Ross 308 broiler chicks were obtained from a local hatchery and housed in an environment-controlled room. Lighting and temperature settings conformed to breeder specifications (Aviagen, Edinburgh, UK). All birds received the experimental mash diets over three feeding periods, starter from 0 to 10d age, grower from 11 to 29d age and finisher from 30 to 42d age. Each diet was fed to 6 pens (5 birds each) following randomisation. Growth

Table 1. The effect of graded levels of dietary sunflower meal on overall daily feed intake (FI), weight gain (WG), feed conversion ratio (FCR) and body weight at 42d age when fed to broilers from day old

Overall inclusion level (g/kg)		FI	WG	FCR	Weight at 42d (g/b)
sunflower meal	soybean meal	(g/b/d)	(g/b/d)	(g:g)	
0	286.7	101.6	54.1	1.840	2437
83.3	206.8	103.8	56.7	1.834	2426
190.0	101.8	104.1	56.1	1.822	2465
293.3	0	105.3	53.2	1.953	2332
SEM		2.14	2.01	0.0440	87.3
P-value		0.682	0.566	0.163	0.729

performance variables were determined at day old and after that when diets were changed. A randomized complete block ANOVA was performed.

Results

All birds remained healthy and mortality was less than 5% throughout the study period. Their growth rates were lower than breeder's performance objectives, but the birds were kept in a small pen facility and fed mash diets. There were no differences observed in FI, WG and FCR when different inclusion levels of SFM were compared ($P > 0.05$). In agreement with these results, Rama Rao et al. (2006) also reported that WG was not affected by total replacement of SBM with SFM at 42 d of age. However, the present experiment was only able to detect statistically significant difference in growth rate of greater than 10%. Smaller differences may be commercially important to the poultry feed industry but experiments with much larger replication would be required to demonstrate statistical significance.

Conclusion

The results confirm that SFM can be used to substitute SBM in poultry diets balanced for metabolisable energy, total crude protein, lysine, methionine, threonine and tryptophan, and the major minerals. A note of caution is that this form of experiment was only able to detect significant differences of greater than 10% in growth rate and smaller differences may be economically important.

Acknowledgments

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Feeding stevia (*Stevia rebaudiana*) leaves to broiler chickens at two different temperatures

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Application

There were no advantages to feeding stevia to broiler even at high temperature.

Introduction

The rise in Earth temperature is an increasingly important concern for poultry producers. Heat stress (HS) is an environmental factor which leads to oxidative stress, namely the disruption of the equilibrium between antioxidants and reactive oxygen species (ROS). Stoyanova, Geuns, Hideg, and Van Den Ende (2011) reported that stevia can be considered as antioxidant, capable of scavenging ROS, thus may be improving growth performance during HS. Although stevia was involved in poultry studies already, there is a lack of information on the impact of stevia on N-corrected dietary metabolisable energy (AMEn), nitrogen (NR) retention coefficient, feed intake (FI), weight gain (WG), feed conversion ratio (FCR), the relative

size of the pancreas and blood glutathione peroxidase (GSH-Px) in broilers at high ambient temperature. The aim of the study was to compare the effect of dietary stevia on the aforementioned variables when fed to broiler chickens reared at two different temperatures – standard temperature (ST; 20°C) and higher temperature (HT; 30°C) from 7 to 21 days of age.

Materials and methods

The experiment was approved by the Harper Adams University Research Ethics Committee. Ninety-six 7d old female Ross 308 chickens were allocated to 32 raised-floor pens (3 birds in each). Sixteen pens were allocated in 2 separate rooms with ST and the rest were allocated in 2 rooms with HT, respectively. The birds were fed one of two mash wheat-based diets: a basal diet (B) formulated with 20 g/kg grass meal and containing 206 g/kg crude protein and 12.67 MJ/kg diet; B + 20 g/kg stevia (determined stevioside 7.61 mg/100 g; rebaudioside 4.81 mg/100 g).

Table 1. Effect of experimental treatments on studied variables

Treatment	FI (g/b/d)	WG (g/b/d)	FCR (g:g)	AMEn (MJ/kg DM)	NR	GSH-Px (g HB/L)	Pancreas (%)
Temperature (T°C)							
ST; 20°C	58.7	34.9	1.689	13.40	0.666	664.0	0.47
HT; 30°C	50.3	31.4	1.611	12.76	0.612	628.0	0.42
SEM	0.7	0.8	0.049	0.61	0.048	13.9	0.02
Stevia							
no	54.9	34.0	1.625	13.14	0.643	662.0	0.41
yes	54.0	32.3	1.674	13.02	0.635	631.0	0.48
SEM	0.9	0.7	0.024	0.12	0.009	22.6	0.02
T°C * Stevia							
HT no Stevia	50.2	31.2	1.623 ^a	12.73	0.614	645.0	0.36
HT plus Stevia	50.4	31.6	1.599 ^a	12.79	0.609	612.0	0.48
ST no Stevia	59.7	36.7	1.627 ^a	13.54	0.671	679.0	0.46
ST plus Stevia	57.6	33.0	1.750 ^b	13.25	0.661	649.0	0.49
SEM	1.2	1.1	0.054	0.62	0.049	17.3	0.03
Probabilities							
T°C	0.015	0.091	0.375	0.535	0.506	0.212	0.188
Stevia	0.477	0.126	0.155	0.482	0.555	0.339	0.013
T°C * Stevia	0.362	0.059	0.037	0.306	0.864	0.954	0.165

^a, ^b, ^c indicate significant differences $P < 0.05$.

The results were statistically compared with ANOVA, following a split-plot design.

Results

Dietary stevia increased the relative size of the pancreas of the birds ($P = 0.013$). There was a stevia by temperature interaction for FCR ($P = 0.037$). Feeding stevia at ST increased FCR, although there was no difference in FCR of birds fed diet with or without stevia at HT. High temperature reduced ($P = 0.015$) FI and tended ($P = 0.590$) to reduce WG of birds fed stevia.

Conclusion

Dietary stevia increased the size of the pancreas of broilers. When fed at ST, stevia increase FCR. Rearing birds at HT reduced their FI.

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Energy and nutrient utilisation and growth performance of chicken fed diets containing graded levels of supplementary sea buckthorn dried berries

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Application

Incorporating dry sea buckthorn berries can improve dietary energy and nutrient availability in broiler diets.

Introduction

Sea buckthorn (SB) (genus *Hippophae*) is a berry-bearing, hardy bush of the family *Elaeagnaceae* that is widely spread in Asia, Europe and recently introduced to Americas. Feeding berries of SB has shown some health benefits, such as preventing liver damage by toxins and healing ulcers that coupled with the abundance of bioactive components in SB, including various antioxidants. However, the impact of SB on growth performance of broilers has been inconsistent as some authors (Biswas, Bharti, Acharya, Pawar, & Singh, 2010) found an

increase in growth performance variables in response to dietary SB, others (Ben-Mahmoud, Mohamed, Bláha, Lukešová, & Kunc, 2014) did not. There is also a lack of information on the impact and optimum level of SB on dietary available energy and nutrient availability in poultry. Thus, the objective of the present study was to quantify the responses in daily feed intake (FI), weight gain (WG) and feed efficiency (FCE) resulting from feeding graded levels of SB to broilers. Responses to N-corrected dietary apparent metabolisable energy (AMEn), dry matter (DMR), nitrogen (NR) and fat (FR) retention coefficients were also determined.

Materials and methods

The experiment was approved by the Harper Adams University Research Ethics Committee. Eighty female Ross

Table 1. Effect of the experimental diets on bird growth performance, N-corrected dietary apparent metabolisable energy (AME), dry matter (DMR), nitrogen (NR) and fat (FR) retention coefficients

Treatment	Sea buckthorn (g/kg)					SEM	Probability		
	0	3	6	9	12		P	L	Q
Feed intake (g/b/d)	63.5	61.0	62.2	62.9	61.6	1.3	NS	NS	NS
Weight gain (g/b/d)	44.3	43.3	43.8	44.1	42.3	0.9	NS	NS	NS
Feed conversion efficiency (g:g)	0.699	0.711	0.705	0.704	0.688	0.009	NS	NS	NS
AMEn (MJ/kg)	11.83	12.63	12.84	12.86	12.64	0.22	0.015	0.012	0.011
DMR	0.636	0.683	0.696	0.692	0.681	0.014	0.032	0.029	0.016
NR	0.605	0.654	0.669	0.661	0.653	0.017	0.085	0.059	0.037
FR	0.751	0.807	0.800	0.804	0.810	0.014	0.040	0.018	0.111

308 chicks were reared from 7 to 21 days of age in 40 pens (2 birds in each). Five diets in total were fed; a basal diet (Control; C) with 211 g/kg CP, 12.69 MJ/kg ME, 9.7 g/kg Ca and 4.8 g/kg available P. Even though SB had a high oil content no further adjustments were made to the basal diet. The C was then split into 5 parts and dried SB berries were incorporated into a diet in mash form at 0, 3, 6, 9 and 12 g/kg, resulting in five diets. The SB berries were collected from Troyan region of Balkan mountain, Bulgaria, cold pressed and the residues of berries and seeds were air dried and preserved in a freezer for about 3 months. A kilogram of SB berries contained 23.62 MJ GE, 233 g fat, 180 g CP and 903 g DM. Each diet was fed to 8 pens following randomisation. The data were analysed with ANOVA. Orthogonal polynomials were used to compare treatment differences for linear (L) and quadratic (Q) relationships with increasing SB level.

Results

Feeding SB did not change ($P > 0.05$) any of the growth performance variables. Although the determined AMEn of the C was unexpectedly low, addition of SB to diet

gave the expected increases to the AMEn, DMR and NR ($P < 0.05$). Diets containing SB increased their NR coefficient in a linear fashion ($P = 0.018$) with increased SB dietary inclusion. There were no ($P > 0.05$) deviations from linear and quadratic relationships for any studied variable.

Conclusion

Although birds fed sea buckthorn berries did not have significantly improved growth performances, there was an improvement in dietary metabolisable energy and nutrient retention coefficients following linear or curvilinear patterns.

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Does growing site affect the digestible energy of wheat cultivars?

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Application

Xylanase addition improved digestible energy of wheat-based diets for broilers but did not interact with wheat quality variables due to cultivar and growing site.

Introduction

Wheat is an important source of energy in UK poultry feeds and can be the only cereal in diet for finishing broilers. The nutritive quality of wheat is variable but the reasons for this variability are poorly understood. Although there are differences in the energy availability and protein digestibility between different wheat cultivars (Pirgozliev, Rose, & Bedford, 2010; Pirgozliev, Rose, & Kettlewell, 2006) this may be confounded by the growing site and environmental conditions during the growing season. Xylanase is used to improve the nutrient availability of wheat-based diets and there is a need to understand if the efficacy is dependent on wheat quality variables. The aim of this study was to determine the

differences in digestible energy (DE) and protein digestibility (PD) in three wheat cultivar samples, each grown in three different geographical sites in the same harvest year. In addition, we examined the effect of xylanase supplementation.

Materials and methods

The experiment was approved by the Harper Adams University Research Ethics Committee. Three current UK wheat cultivars; Lili (L), Barrel (B) and Kerrin (K) from three growing sites; Lincolnshire (Lin), Cambridgeshire (Cam) and Yorkshire (York) (nine wheat samples in total) were used in this study. Nine isonitrogenous diets were formulated all of which included the wheat sample at 670 g/kg and 330 g/kg of a balancer (Azhar et al., 2019). Differences in protein of wheat samples were adjusted by addition of wheat protein isolate. A total of 880 male Ross 308 broilers were obtained from a commercial hatchery and allocated to a single floor pen and offered a standard wheat-based broiler starter

Table 1. The effect of wheat cultivar, site of production and Xylanase enzyme supplementation on dietary digestible energy (DE) and protein digestibility (PD) coefficient when fed to broiler chickens

Variable	Wheat cultivars			Sites			SEM	Xylanase			Probabilities		
	B	K	L	Cam	Lin	York		-	+	SEM	Cultivar	Site	XYL
DE	12.67	12.90	12.84	13.03	12.55	12.82	0.167	12.54	13.06	0.14	NS	NS	0.009
PD	0.767	0.766	0.779	0.770	0.760	0.782	0.007	0.765	0.777	0.006	NS	0.078	NS

feed formulated to meet Ross 308 nutrient requirements. On the first day of the experiment (at 7 d of age) eight birds were placed in each pen (0.6 m X 0.6 m solid floor area) within a controlled environment room. Each diet was fed at random to 6 pens from 7 to 21 d age. The husbandry conditions followed industry standards. Access to the feed and the water was ad libitum. At the end of the study, at 21d age, three birds per pen were stunned and killed, and ileal digesta was obtained and pooled in a plastic tub. The digesta was then freeze-dried and milled to pass 0.75 mm screen before further analysis. Dietary DE and ND were determined as previously described (Yang et al., 2019). The DE and ND were compared statistically by ANOVA using a $3 \times 3 \times 2$ factorial arrangement of treatments. The main effects analysed were the wheat cultivar, site of production and the XYL supplementation.

Results

Feeding Xylanase improved dietary DE by 0.56 MJ ($P = 0.009$), although the site of production tended

($P = 0.078$) to influence PD of diets. There were no significant interactions between any of the studied treatment factors.

Conclusion

The study shows that there were no detectable differences between three selected cultivars grown in there different UK growing sites. The response to xylanase was expected but did not interact with any other treatment factor.

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The effect of saturated and non-saturated fat on growth performance, dietary apparent metabolisable energy and nutrient retention when fed to broiler chickens at two different temperatures

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Application

Unsaturated fat diets give improved growth and nitrogen retention but no additional benefits to birds reared under high temperatures.

Introduction

Various nutritional strategies have been suggested to overcome the negative impact of heat stress (HS) in broilers, including dietary mineral and protein levels, various antioxidants and plant extracts (Del Vesco et al., 2015; Harsini, Habibian, Moeini, & AbdolMohammadi, 2012). However, there is lack of knowledge whether the source of dietary fat, i.e. non-saturated (NSF) or saturated (SF) can influence HS resistance in broilers. Compared to SF, NSF usually provides higher metabolisable energy, nutrient digestibility and performance (Tanchaorenrat, Ravindrana, Zaefariana, & Ravindran, 2013). The aim of the study was to compare the effect of dietary SF and NSF on daily feed intake (FI), weight gain (WG), feed

conversion ratio (FCR), N-corrected apparent metabolisable energy (AMEn), dry matter (DMR), nitrogen (NR) and fat retention (FR) on broiler chickens reared at two different temperatures – standard temperature (ST; 20°C) and higher temperature (HT; 35°C) from 14 to 35 days of age.

Materials and methods

The experiment was approved by the Harper Adams University Research Ethics Committee. One-hundred-and-twenty 14d old male Ross 308 chickens were allocated to 24 raised-floor pens (5 birds in each). Twelve pens were allocated in 2 separate rooms with ST and the rest were allocated in 2 rooms with HT, respectively. The birds were fed one of two mash diets: one diet containing NSF (rapeseed oil), and another diet containing SF (Megalac®), resulting in 4 experimental treatments in total. The results were statistically compared with ANOVA (GenStat 17th edition, VSN international Ltd, UK) using a split-plot design.

Table 1. Effect of rearing temperature and dietary fat source on daily feed intake (FI), weight gain (WG), feed conversion ratio (FCR), N-corrected apparent metabolisable energy (AMEn), dry matter (DMR), nitrogen (NR) and fat (FR) retention coefficients in broilers

Variable	Temperature (T°C)			NSF	FAT		T°C	Probabilities	
	20°C	35°C	SEM		SF	SEM		FAT	T°C x FAT
FI (g/b/d)	109.0	60.0	1.7	86.0	83.0	1.7	<0.001	NS	NS
WG (g/b/d)	67.0	29.0	1.7	49.0	47.0	1.5	<0.001	NS	NS
FCR (g:g)	1.618	2.048	0.047	1.819	1.846	0.032	<0.05	NS	NS
AMEn (MJ/kg DM)	13.48	13.43	0.19	13.80	13.12	0.17	NS	<0.05	NS
DMR	0.718	0.699	0.015	0.724	0.693	0.010	NS	<0.05	NS
NR	0.661	0.509	0.022	0.604	0.565	0.013	<0.05	<0.05	NS
FR	0.747	0.763	0.009	0.825	0.686	0.013	NS	<0.001	NS

Results

Birds reared at ST had higher FI, WG and NR ($P < 0.001$), and reduced FCR ($P < 0.05$), compared to birds reared at HT. Diet containing NSF had higher AMEn, DMR, NR ($P < 0.05$) and FR ($P < 0.001$), versus SF diets respectively. There were no ($P > 0.05$) temperature x fat interactions (Table 1).

Conclusion

Rearing birds at high temperature reduced broiler performance variables and nitrogen retention, but not metabolisable energy, dry matter and fat retention coefficients. Birds fed saturated fat diets had reduced metabolisable

energy, dry matter, nitrogen and fat retention coefficients, but there were no additional benefits for the birds kept at high temperatures. This study will help poultry producers make informed choices when rearing broilers in higher temperatures and when considering sources of dietary fat to maximise bird performance.

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Lower non-starch polysaccharides content in sunflower meal gives improved feeding value for broilers

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Application

Low total, but not soluble, non-starch polysaccharides content in sunflower meal is associated with better nutrient availability for broilers.

Introduction

Sunflower meal (SFM) is a potentially attractive protein source for poultry (Rama Rao *et al.*, 2006). During the standard process of SFM production, the husk is not removed, and the meal contains high amounts of fibres that reduces its metabolisable energy and inclusion level in poultry diets. However, there are new technologies available for SFM production, which separate the husk from the meal improving its feeding quality for broilers. The aim of the study was to compare dietary N-corrected apparent metabolisable energy (AMEn), nitrogen digestibility (ND) coefficient, analysed on digesta, total tract dry matter (DMR) and fat (FR) retention, of four SFM samples, produced under different conditions, when fed to broilers.

Materials and methods

The experiment was approved by the Animal Experimental Committee of Harper Adams University. Four SFM samples produced in Bulgaria were obtained. The hulls in the first SFM sample were not removed. The hulls in the second SFM were partially removed. The hulls in the third and fourth SFM samples were removed and sample 4 was pelleted. The SFM samples were analysed for crude fat, crude protein and non-starch polysaccharide contents following standard AOAC techniques (Table 1). For diet preparation, a standard broiler starter feed was replaced weight for weight by 20% of the respective SFM products. Day-old male Ross 308 broiler chicks were obtained from a local hatchery and housed in an environment-controlled room. Lighting and temperature settings conformed to breeder specifications (Aviagen, Edinburgh, UK). All birds received a standard mash broiler starter diet from day 0 to 10 and access to feed and water was provided ad libitum. On day 10 all birds were assigned to one of 40 small floor pens (2 birds per pen). Each diet was fed to 10 pens

Table 1. The effect of sunflower meal (SFM) samples on dietary N-corrected apparent metabolisable energy (AMEn), on dry matter basis, nitrogen digestibility (ND), total tract dry matter (DMR) and fat (FR) retention coefficients when fed to broilers

Sample	Chemical composition of sunflower meal samples				Nutrient availability of experimental diets			
	CP (g/kg)	Fat (g/kg)	NSPt (g/kg)	NSPs (g/kg)	AMEn (MJ/kg)	ND	DMR	FR
1	373.6	4.2	217	51	12.71 ^a	0.729 ^{ab}	0.621 ^a	0.812 ^a
2	400.1	1.0	202	48	12.66 ^a	0.682 ^a	0.647 ^{ab}	0.843 ^{ab}
3	425.3	4.1	172	52	12.86 ^{ab}	0.805 ^c	0.666 ^b	0.850 ^{ab}
4	456.2	3.9	164	54	13.31 ^b	0.784 ^{bc}	0.671 ^b	0.853 ^b
SEM					0.15	0.019	0.008	0.010
cv%					3.6	7.8	4.0	3.8
P-value					0.017	<0.001	<0.001	0.029

^{a, b, c} indicates significant differences $P < 0.05$.

following randomisation. Dietary AMEn and nutrient availability coefficients were determined using indigestible marker (acid insoluble ash). Ileal digesta were collected for ND determination. The results were statistically compared using a randomised block ANOVA (GenStat 17th edition, VSN international Ltd, UK). Tukey's multiple range test was used to determine significant differences between diets when $P < 0.05$.

Results

Samples 3 and 4 contained more CP, and relatively low NSPt compared to samples 1 and 2 (Table 1). Diets containing sample 4 had higher AMEn compared to diets based on samples 1 and 2 ($P < 0.05$). In general, diets based on samples 3 and 4 tended to have higher AMEn and nutrient availability coefficients compared to the rest ($P < 0.1$). The overall feed intake was 734 g per bird and did not differ between treatments ($P > 0.05$). The results suggest that SFM samples

lower in total NSP, but not soluble NSP, have better feeding value for broilers.

Conclusion

Lower content of total NSP in SFM samples are associated with higher nutrient availability for broilers. There was no association with soluble NSP.

Acknowledgments

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Xanthophyll and beta carotene contents of differently processed natural, biodegradable raw materials to be used as potential egg yolk colourants in layer chicken diets

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Application

There is a high cost of procuring synthetic dyes being used as egg yolk colourants. Potential egg yolk colourants of natural sources are available, cheaper and will leave no injurious residue in food products.

Introduction

Egg yolk colour has a considerable influence on egg marketing (Gunawardana, Roland, & Bryant, 2008). Panaite, Bunduc, Criste, and Cornescu (2015) remarked that egg yolk colourants from plant sources were cheap, natural sources of plants as sources of xanthophylls, which could replace successfully the synthetic products used to enhance the colour of egg yolks. The study was to evaluate the concentrations of xanthophylls and beta carotene in natural and biodegradable raw materials to be used as potential egg yolk colourants.

Materials and methods

The experimental leaves used for this study were *Adansonia digitata*, *Aspilia africana*, *Chromolaena odorata*, *Moringa oleifera*, *Ocimum gratissimum*, *Parkia biglobosa* and *Tithonia diversifolia* which were sourced from various locations in Oyo and Ogun States. The leaves of the plants were destalked and washed properly first with tap water and then with distilled water to remove dirt. The leaves were subjected to three drying methods namely, air drying, oven drying and freeze drying. Fresh leaves to be air dried were spread in ventilated rooms at room temperature varying from 27–31°C till they were dried and crispy to touch. Fresh leaves to be oven dried were placed in large clean forced draught oven at 65°C in labelled envelopes till the leaves were dry and constant weights were obtained on an average of 3 days. Fresh leaves to be freeze dried were placed inside a lyophiliser at –80°C till the leaves were freeze dried on an average of 3 days. The already dried leaves were milled to pass through 1.0 mm sieve and stored in well-labelled sample bottles with

Table 1. Xanthophylls and beta carotene contents of differently processed potential egg yolk colourants.

Parameters		Leaf sample (LS)						
		LS 1	LS 2	LS 3	LS 4	LS 5	LS 6	LS 7
Beta Carotene (mg/100 g)	Air-dried	6.70	5.40	8.40	10.10 ^a	6.60	6.00 ^b	4.70
	Oven-dried	6.50	5.40	8.20	7.00 ^b	6.50	6.60 ^a	4.60
	Freeze-dried	6.80	5.50	8.30	9.00 ^b	6.70	6.50 ^a	4.70
	SEM	0.01	0.01	0.01	0.02	0.01	0.01	0.01
Xanthophyll (mg/100 g)	Air-dried	12.26 ^a	13.67	11.85 ^a	15.25 ^a	9.17	10.55 ^c	14.74
	Oven-dried	11.07 ^b	13.80	10.09 ^b	9.59 ^b	9.52	14.12 ^a	14.04
	Freeze-dried	11.26 ^{ab}	13.82	9.96 ^b	9.31 ^b	9.45	12.22 ^b	14.91
	SEM	0.31	0.41	0.48	0.98	0.34	0.16	0.30

^{abc} means in the same column having different superscripts differ significantly ($P < 0.05$).

stoppers until needed for xanthophyll and beta carotene analyses. Quantitative analyses of xanthophylls and beta carotene were carried out by the procedure according to Harbourne (1973). Results obtained were analysed using STAR 2.0.1. Drying methods were compared using Analysis of Variance and means were separated with Duncan Multiple Range Test at $P < 0.05$.

Results

Beta carotene values of most leaf samples were statistically similar ($P > 0.05$). These trends were also observed for xanthophylls in LS 2, 5 and 7, where air-dried values obtained for LS 1, 3 and 4 were found to be the highest ($P < 0.05$) compared to oven-dried and freeze-dried. All the results described above are shown in Table 1.

Conclusion

Beta carotene and xanthophylls in the leaves showed that all leaf samples appear to have good potentials of being used as natural egg yolk colourants in laying chicken feeds. With about 2 mg/kg yellow carotenoids in

feedstuffs, at 110 g/hen/day, 15 mg/kg of red at 10% canthaxanthin and 25 mg/kg of yellow at 10% apoester were recommended by the manufacturer of synthetic yolk colourants, to get yolk colour score of 10. The inclusion rates of the leaves in the diets of laying chickens would be estimated corresponding to the amounts recommended by the synthetic dyes manufacturers.

Acknowledgments

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Feeding a combination of xylanase and arabino-xylo-oligosaccharides to young broiler chickens

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Application

The positive response of supplementing xylo-oligosaccharides to poultry feed may be obvious over a longer feeding period.

Introduction

Breakdown products of arabinoxylans on use of a xylanase (XYL) have been demonstrated to positively influence animal performance and gut functionality in poultry (Bedford, 2018). The success of such improvements, however, has been hypothesised to be linked to the development of the gastrointestinal microbiota and cereal quality. Degradation of dietary arabinoxylans with XYL leads to the production of arabino-xylo-oligosaccharides (AXOS) that are metabolised by specific groups of bacteria producing short-chain fatty

acids, in particular butyric acid. Thus, the concomitant supplementation of XYL and a fermentable AXOS in broiler diets may improve gut health and growth performance of birds. The objective of this project is to determine the impact of a combination of supplementary commercial blend of AXOS/XYL (SIGNIS, ABVista, UK), on daily feed intake (FI), weight gain (WG), feed conversion ratio (FCR) and dietary apparent metabolisable energy (AME) when fed to broiler chickens from 0 to 10d age.

Materials and methods

The experiment was conducted at the National Institute of Poultry Husbandry and approved by the Harper Adams University Research Ethics Committee. The study involved 24 floor pens (12 pen with males + 12 pen with females), each

Table 1. The effect of a blend of xylanase and arabino-xylo-oligosaccharides on daily feed intake (FI), daily weight gain (WG), feed conversion ratio (FCR), and apparent metabolisable energy (AME) when fed to broiler chickens. There were no interactions between treatments ($P > 0.05$)

Parameter	Fibre		Sex		AXOS/XYL			Probabilities (P-value)		
	no	yes	F	M	no	yes	SEM	Fibre	Sex	AXOS/XYL
FI (g/b/d)	29.6	29.7	28.9	30.4	29.9	29.4	0.3	0.825	<0.001	0.246
WG (g/b/d)	25.4	25.0	24.5	25.9	25.2	25.2	0.2	0.297	<0.001	0.861
FCR (g:g)	1.161	1.181	1.171	1.170	1.176	1.166	0.012	0.217	0.960	0.567
AME (MJ/kg)	11.73	11.80	11.84	11.70	11.72	11.81	0.06	0.374	0.118	0.351

housing 20 Ross 308 birds. A basal diet used as a positive control (PC) contained 539 g/kg maize and 387 g/kg soybean meal as the main ingredients, and with a calculated 12.59 MJ/kg AME and 228 g/kg CP content. The diets were then split on two parts and one part was 'diluted' with 50 g/kg wheat bran at the expense of maize to generate the negative control (NC). Both, PC and NC were then further split on two and one part was supplemented with a blend of AXOS/XYL, resulting in 4 diets, or 8 treatments in total including 4 diets and 2 sexes. Each diet was fed to 12 floor pens from day old to 10d age following randomisation. At day 7, ten birds from each pen were allocated to a pen with mesh floor and excreta were collected for the last three days for AME determination. Data were analysed by ANOVA following 2x2x2 factorial design (including 2 fibre levels, with and without enzyme, and two sexes).

Results

The growth performance of the birds was similar, but slightly below breeder's recommendations (Aviagen Ltd., Edinburgh, UK). There were no differences ($P > 0.05$) in FI, WG or FCR between birds fed PC and NC. Male birds had higher FI and WG ($P < 0.001$) but there was no difference in FCR ($P > 0.05$). Feeding AXOS/XYL did not produce difference

in performance variables ($P > 0.05$), although FCR for birds fed the blend was numerically ($P > 0.05$) lower compared to control fed birds. Dietary AME was not affected by any of the factors ($P > 0.05$). The relatively low AME values may be attributed to the very young age of the birds.

Conclusion

Male birds consumed more feed and grew faster compared to females. The results suggest that the adaptation of the birds/microbiota to ferment arabino-xylo-oligosaccharides may require some time as the animals grow and the very short period if this study was likely not long enough for a benefit to accrue.

Acknowledgments

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The impact of dietary mixture of xylanase and xylo-oligosaccharides on energy availability and caecal fermentation when fed to broilers

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Application

Feeding broilers a mixture of xylanase and xylo-oligosaccharides increases caecal short-chain fatty acid production that may be beneficial in stress conditions.

Introduction

The beneficial effects of exogenous xylanase in broiler feed have been attributed to several mechanisms, such as reduction of gut viscosity, release of digestible components from plant cell wall and generation of xylo-oligosaccharides (Bedford, 2018). Xylo-oligosaccharides (XOS) may be potential prebiotics that influence bird growth performance, gut microbiota and health. The aim of the study was to compare the effect of a mixture of xylanase and XOS (SIGNIS, AB Vista, Marlborough, UK) on dietary N-corrected apparent metabolisable energy (AMEn) and growth performance

variables including feed intake (FI), weight gain (WG), feed conversion ratio (FCR), when fed to broilers. The activity of beneficial microbiota in the chicken gut was assessed by determination of acetic (AA), butyric (BA), propionic (PA), and total short-chain fatty acid (SCFA) in caecal digesta.

Materials and methods

The experiment was approved by the Harper Adams University Research Ethics Committee. A total of 320 day-old Ross 308 broiler chicks that were placed to 16 floor pens (20 birds in each). Control wheat-maize-soybean broiler mash diets were prepared, these included starter-grower (0–20d age) and finisher (21–35d age) phases. A second diet was produced by replacing 100 g/t maize with a blend of xylanase and XOS (SIGNIS, AB Vista, UK) in both growing phases. All diets contained 20 g/kg acid insoluble ash as indigestible

Table 1. Effect of added blend of xylanase and xylo-oligosaccharides on the growth performance of broilers and the N-corrected apparent metabolisable energy (AMEn), and caeca concentration of acetic acid (AA), propionic acid (PA), butyric acid (BA) and short-chain fatty acids (SCFA) when measured at two different ages

Factor	AMEn (MJ/kg)	FI (g/b/d)	WG (g/b/d)	FCR	AA (mM)	BA (mM)	PA (mM)	SCFA (mM)
<i>Diet</i>								
Control	11.40	62.7	33.1	1.649	65.1	10.5	3.5	86.5
SIGNIS	11.56	64.9	33.3	1.684	70.3	15.8	2.7	95.9
SEM	0.07	0.90	0.67	0.029	6.9	2.7	0.7	9.2
<i>Bird age</i>								
21 d	10.81	50.0	27.4	1.568	77.6	18.1	3.3	107.7
35 d	12.16	77.6	38.9	1.765	57.7	8.2	2.9	74.7
SEM	0.05	0.63	0.47	0.020	4.9	1.9	0.5	6.5
<i>P-values</i>								
diet	0.029	0.775	0.717	0.776	0.006	0.005	0.771	0.007
age	<0.001	<0.001	<0.001	<0.001	0.013	0.003	0.532	0.003
diet*age	0.879	0.888	0.745	0.628	0.263	0.294	0.395	0.268

marker. The SIGNIS supplemented starter-grower diet had determined 625 FTU/kg phytase and 15,800 BXU/kg xylanase activities and the finisher diet had 651 FTU/kg phytase and 18,600 BXU/kg xylanase activities, respectively. Each diet was fed to 8 pens following randomisation. At the end of each dietary phase, from 17 to 20 and 32 to 35d age, excreta were collected from each pen, dried, milled and analysed for gross energy content. Caeca contents were also collected at the end of each phase and analysed for AA, BA, PA and total SCFA as described by Apajalahti, Vienola, Raatikainen, Holder, and Moran (2019). The results were statistically compared with ANOVA, using analysis of repeated measurements.

Results

There were no differences ($P > 0.05$) in FI, WG or FCR between birds fed different diets. Feeding SIGNIS increased AMEn ($P = 0.029$) compared to control diets. Dietary

SIGNIS also led to an increase caecal production of AA, BA and total SCFA ($P < 0.05$). There were no age x diet interactions ($P > 0.05$) in any of the measured variables.

Conclusion

Although supplementary SIGNIS did not affect growth performance variables of birds, it increased dietary AMEn and the caecal production of AA, BA and total SCFA, that all indicates an increase in intestinal microbial fermentation. The increased production of SCFA may be beneficial in periods of disease challenge or other stress.

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Susceptibility of different wheat varieties to enzymatic hydrolysis

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Application

Determining the level of oligosaccharides generated by different wheats could show which respond best to enzyme treatment.

Introduction

Xylanases are commonly used in wheat-based formulations to help improve their feeding value (Bedford, 2000), however different wheat cultivars vary in their susceptibility to hydrolysis by the same enzyme (Smeets, Nuyens, Van Campenhout, & Niewold, 2013). This may be due to variations in their viscosity (estimated by soluble xylan) and/or their ability to release fermentable xylo-oligosaccharides. Therefore, the current in vitro study was designed to determine which wheat cultivars may respond better to treatment by the same xylanase through estimation of variance in viscosity and prebiotic potential.

Materials and methods

Ten different wheats were selected for in vitro analysis. Methods were designed to determine the following xylan fractions from each wheat; soluble xylan, enzyme extractable xylan, soluble short chain xylans and enzyme extracted short chain total xylans. Wheat samples were ground through a 0.5 ml mesh screen in a Retsch Mill. 2 g of each wheat was incubated in 10 ml of 0.05 M sodium citrate buffer (pH 5.5) and incubated at 41°C for 1 hr with and without 16,000 BXU/kg Econase XT (AB Vista). Supernatants were collected after each incubation and a subsample precipitated in 4 vols of absolute ethanol (80% final EtOH concentration). The supernatants from all incubations were acid hydrolysed in 5 ml of 1.3M HCL at 100°C for 1 hr and neutralised once cooled, using 5 ml of 1.3M NaOH. Xylose content was determined using a Megazyme assay procedure for D-xylose (Megazyme International, 2016). Data were analysed as a complete factorial with wheat, enzyme and

Table 1. Different fractions of xylose content (mg/g) of the ten studied wheat samples.

Wheat	Soluble xylose content	Soluble xylose content after enzyme treatment	Oligosaccharide content of wheat	Oligosaccharide content after enzyme treatment
JB Diego	3.49	14.40	0.34	1.58
Bennington	3.58	10.31	0.26	0.94
Barrel	4.55	11.60	0.42	0.34
Dunston	3.81	8.19	0.25	0.85
Siskin	3.96	11.87	0.44	0.48
Reflection	5.13	14.33	0.05	1.05
Zyatt	3.65	10.97	0.54	1.07
Graham	4.18	7.85	0.20	0.74
Shabras	3.20	9.60	0.13	0.60
Kerrin	3.78	10.33	0.10	0.90
St err	0.89	1.71	0.22	0.31

Table 2. Effects of treatment

Treatment	P-Value
Wheat variety	0.1349
Econase	<0.001
Wheat variety*Econase	0.2388
Precipitated	<0.001
Wheat variety*Precipitated	0.1957
Econase*Precipitated	<0.001
Wheat variety*Econase*Precipitated	0.2766

precipitation as the factors using JMP 15th edition. Differences were considered significant with $P < 0.05$ and means separated, where appropriate, by Students t-test.

Results

Wheat clearly varies in soluble xylan content (Table 1) regardless of treatment with further variation when xylanase is added. As expected, oligosaccharide content is low and varies between wheats, but the addition of xylanase released a greater quantity of xylo-oligosaccharides, which is likely a result of further hydrolysis of soluble and insoluble xylan. The models show the main effects of wheat and the interaction between enzyme and precipitation (i.e. oligosaccharides

vs total xylan content) to be significant (Table 2). These data suggest that wheat differs in its susceptibility to release soluble xylan and oligosaccharides in the presence of a xylanase, which may explain part of the variation in response in wheat-based diets when a xylanase is added.

Conclusion

These data suggest there is a variability between wheat samples in soluble xylan and oligosaccharide content and even more variable response to those wheats when xylanase is added with regards to additional soluble xylan and oligosaccharide release. Further detailed analysis of the feeding value of these wheats will determine the relevance of these data in predicting wheat feeding value.

Acknowledgments

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Determination of optimum post-mortem blood sampling and storage methods for blood antioxidative capacity testing in broilers

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Application

The use of anticoagulants and subsequent storage influences the accuracy of results in oxidative assays. Care must be taken when sampling to ensure the more representative results.

Introduction

Substances such as polyunsaturated fatty acids (PUFA's), proteins and DNA are all potentially oxidizable by free

radicals and reactive oxygen species (ROS). A complex antioxidant defence system normally protects the oxidizable products within cells. When this is overwhelmed damage to these biological molecules occur compromising cell health (Katerji, Filippova, & Duerksen-Hughes, 2019). This is referred to as oxidative stress. Oxidative stress in poultry affects meat and egg quality as well as bird performance. It is caused by pathological factors, nutrition and environmental factors, such as heat stress and high ammonia levels. It may be possible to mitigate oxidative stress through dietary supplementation of antioxidant additives capable of savaging

Table 1. Anticoagulants and subsequent storage effects the accuracy of results in oxidative assays

Parameters		VEA $\mu\text{mol/L}$ ($\pm\text{SE}$)	P-value
<i>Effect of blood storage</i>	Fresh	1029.21 \pm 41.59 ^a	0.003
	Defrost	863.22 \pm 34.59 ^b	
<i>Effect of blood tube additives</i>	EDTA	805.73 \pm 29.17 ^b	<0.001
	Heparin	894.23 \pm 30.51 ^b	
	None	1095.04 \pm 66.49 ^a	

^{abc}superscript letters denote differences ($P < 0.05$)

ROS (Mishra & Jha, 2019). The ABEL-40 M2 kit measures the relative effective antioxidant capacity within blood plasma and or serum. Vitamin E is included as a standard and the kit reports results as Vitamin E analogue equivalent units (VEA). This method could be potentially valuable for determining the efficacy of antioxidant additives in research trials, and as a measure of oxidative stress in birds in challenging environments. In order to achieve useful data from this ABEL-40 M2 kit it is important to first validate sample collection and sample storage conditions.

Materials and methods

Blood samples were taken post-mortem from thirty day-35 Ross 308 broilers culled by cervical dislocation. Samples were divided into three blood tubes containing either EDTA, Heparin or nothing (for serum). These tubes were centrifuged at 3000rpm for 10 minutes. The plasma/serum was removed and tested within 16 h using the ABEL-40 M2 kit from Knights scientific according to the manufacturer's protocol. The remaining plasma was then frozen for 10 days at -20°C , before defrosting and testing using the kit in the same procedure as previously to assess the effect of short-term storage. Each plate also contained 6 duplicates of reconstituted freeze-dried serum of known VEA. They were evenly distributed throughout the plate that acted as an Internal standard. Data was analysed by IBM SPSS v26 using univariate analysis and Duncan's multiple range post-hoc test to elucidate differences.

Results

The addition of EDTA or Heparin in the blood sampling tube reduced the VEA value of the blood sample (Table 1), as did the process of freezing and defrosting (Table 1). There was no significant interaction between factors found in the univariate analysis.

Conclusion

In order to reduce external factors when analysing for antioxidant effects, samples should be collected as serum rather than plasma as anticoagulants appear to have a detrimental effect. It is also advisable to test samples immediately as storage at -20°C was also shown to reduce antioxidant readings. To confirm these conclusions the experiment should be repeated.

Acknowledgments

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Determining the effects of housing system on skeletal integrity at both early and mid-lay

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Application

Through understanding the developmental changes in bone development in different housing systems, we can create appropriate interventions where necessary.

Introduction

The egg industry is a large part of the UK economy, and as preference for chicken produce from high welfare grows, the industry must adapt to ensure welfare standards continue to rise. Bone strength in laying hens is a current welfare issue. Laying hens become sexually mature from around 16–24 weeks (Hester, 2017) so large volumes of dietary calcium are partitioned towards medullary rather than cortical bone

formation, leaving the still-growing chicken with minimal amounts of calcium for robust skeletal development. Therefore, fractures are common in laying hens and can be dependent on the housing system used. The aim of this study was to compare the effect of housing system on development of selected key bones at both early and mid-lay.

Materials and methods

This small study was conducted as a WPSA Summer Studentship and forms part of a larger project. Ethical approval was granted by the School of ARES Ethical review group. Six Lohmann Brown Classic hens from two farms per free range system and a caged system were collected and euthanised at two

Table 1. Effect of age and housing type on bone breaking strength of key laying hen bones.

Age	24 weeks		48 weeks		SEM	P-values		
	Free range	Caged	Free range	Caged		Age	Housing	Interaction
L Humerus	283.11 (±16.166)	189.59 (±9.183)	231.99 (±15.480)	192.64 (±11.496)	9.47	<0.001	<0.001	NS
R Humerus	286.58 (±14.764)	203.73 (±13.947)	260.67 (±21.859)	167.23 (±10.025)	11.13	<0.05	<0.001	NS
L Tibia	318.46 (±14.536)	258.14 (±23.681)	256.64 (±21.020)	222.41 (±17.569)	13.80	<0.05	<0.05	NS
R Tibia	324.37 (±14.198)	330.53 (±44.262)	268.48 (±16.271)	236.51 (±10.342)	17.79	<0.001	NS	NS
Keel	188.54 (±31.936)	189.28 (±42.981)	195.13 (±26.174)	207.62 (±26.319)	15.93	NS	NS	NS

time points – 24 weeks (early lay) and 48 weeks (mid lay). Body weight was recorded then the left and right humerus and tibia, along with the keel bone were dissected out from each bird. The bones were then de-fleshed and morphometric measurements were taken, though only breaking strength was used in this study. The strength of each bone was measured using a TA.XT texture analyser (Guildford, UK). Each bone type was placed on the machine in identical orientation to reduce error. A 2-way ANOVA (SPSS, IBM statistics 26) was used to examine the differences in bone strength between housing systems over two different ages.

Results

Free range hens had stronger tibia and humerus than cage hens across both age groups but there was no significance difference in keel bone strength at any age. In both humerus and tibia, substantial differences between left and right bones were observed.

Conclusion

The lateral asymmetry was a surprising finding which requires further investigation. More likely to associated with the eggshell (and hence calcium) mass required at this stage in production and the prolonged negative calcium balance that leads to reductions in cortical bone.

Acknowledgments

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Energy metabolisability and ileal digesta viscosity of growing turkeys fed diets containing malted sorghum sprouts supplemented with enzyme or yeast

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Application

Malted sorghum sprouts (MSP) supplementations reduced energy utilization in growing turkeys. Energy metabolisability was improved with additive supplementation compared with MSP singly.

Introduction

Nigerian livestock enterprise has suffered more than any other industries as a result of rising costs of production as feed alone accounts for up to 75% of total cost of broiler production. Moreover, the competition between man and livestock for these feed ingredients necessitate the use of alternative sources of ingredient. Oke et al. (2010) reported that a commercial enzyme and yeast supplementation in Malted Sorghum Sprout diet improved broiler performance. This research work therefore determined the energy metabolizability and ileal digesta viscosity of growing turkeys fed MSP diets supplemented with enzyme or yeast.

Materials and methods

One hundred and twenty 28d old British United Turkeys were randomly allocated to 6 dietary treatments. The six experimental diets were formulated such that each treatment contained either enzyme or yeast at 200ppm in diets containing MSP at 0, 50, or 100 g/kg respectively. The exogenous enzyme used is a commercial blend of xylanase (EC3.2.1.8), β glucanase (EC3.2.1.4) and protease (EC 3.4.2.1). At day-84, 6 groups of 6 turkeys each were allocated to 6 experimental diets while an additional group of 6 turkeys were used for estimation of endogenous losses. Three days of acclimatization in metabolic cage were allowed for the birds before the commencement of the study. Known weights of feed were given and leftovers were measured daily. Excreta were collected for three days, unlike the former group of birds where only water was administered to determine the endogenous losses. At the lapse of 24 hours, excreta were collected for two days. Apparent metabolisable energy (AME), AME corrected for nitrogen retention (AMEn), true metabolisable energy (TME) and TME corrected for nitrogen retention were determined according to the method of Sibbald (1979). At day-112,

Table 1. Effects of Malted sorghum sprouts (MSP) inclusion and enzyme or yeast supplementation on the apparent and true metabolizable energy of growing turkeys (9–12 weeks)

Measurements	MSP Inclusion (g/kg)				Additives (mg/kg)					P value
	0	50	100	SEM	L	Q	Enzyme	Yeast	SEM	
AME (MJ/kg)	15.29 ^b	15.63 ^a	15.77 ^a	0.08	0.019	0.025	15.57	15.56	0.12	0.14
TME (MJ/kg)	16.43	16.75	16.78	0.07	0.057	0.112	16.66	16.65	0.13	0.17

^{a, b}Means on the same row having different superscript differ significantly ($P < 0.05$)

Table 2. Effects of Malted sorghum sprouts (MSP) inclusion and enzyme or yeast supplementation on viscosity of growing turkeys

Speed	MSP Inclusion (g/kg)				Additives (mg/kg)					P value
	0	50	100	SEM	L	Q	Enzyme	Yeast	SEM	
50	297.8 ^b	329.9 ^a	308.8 ^b	4.56	0.232	0.000	313.1	311.2	3.72	4.12
60	250.7 ^b	277.4 ^a	274.1 ^a	4.38	0.008	0.003	267.7	267.1	3.74	4.23

^{a, b}Means on the same row having different superscript differ significantly ($P < 0.05$)

ileal digesta viscosity (2 birds per replicate, 48 turkeys were selected based on final live weight, slaughtered and digesta content collected from the Merckel's diverticulum to the ileo-caecal junction into a labeled sample bottles. Uniform weight of digesta was diluted to a volume of 400 ml using a brookfield DV-E viscometer. The readings were taken at 50 and 60 revolution per minute. Data generated and Polynomial contrast (Linear and quadratic) for determination of enzyme or yeast inclusion were analysed using SPSS (1999).

Results

Birds fed 50 and 100 g/kg MSP had higher ($P < 0.05$) AME while the least values were recorded for birds fed control diet. The TME values showed no effect ($P > 0.05$) on dietary inclusion of MSP and supplementation with enzyme or

yeast. Ileal viscosity increased ($P < 0.01$) with increasing concentration of MSP with or without additives.

Conclusion

Turkeys metabolized MSP at 50 g/kg and 100g/kg for improved AME, irrespective of the additive used. Supplementation with 200 mg/kg enzyme or yeast did not significantly affect ileal digesta viscosity.

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Developing a model to test the efficacy of alternatives to antibiotics in broiler diets

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Application

There is a potential to develop a model to elicit an inflammatory response as a means to test the efficacy of alternatives to antibiotics in broiler diets.

Introduction

Antibiotics are used to treat disease on veterinary prescription and, as a side effect, reduce an exacerbated mucosal inflammatory immune response to microbial infection which leads to necrosis and diarrhoea. Establishing the relationship between inflammatory immune response and performance/digestibility is an important first step in designing strategies to maintain bird health, welfare and productivity in a reduced antibiotic-use system (Kuttappan et al., 2015). To date, no model has been effectively developed within broilers to stimulate an inflammatory immune response within the

gastrointestinal tract which could be used to test the efficacy of alternatives to antibiotics. The aim of this study was to elicit an inflammatory immune response by the administration of dextran sodium sulphate (DSS) or by feed withdrawal (FW) in broilers and to establish the relationship between performance/digestibility and inflammatory immune response.

Materials and methods

The trial was approved by the Animal Welfare Ethical Review Body at the Agri-Food Bioscience Institute. Male Ross 308 broilers ($n = 480$) were placed in pens (20 birds/pen) at 0d and assigned to one of three experimental treatments; 1) control, 2) DSS (Alfa Aesar, Heysham, UK) and 3) FW resulting in eight pen replicates/treatment. Birds were offered standard starter (12.7 MJ/kg AME, 215 g/kg crude

Table 1. The effect administration of dextran sodium sulphate (DSS) or by feed withdrawal (FW) and sampling time on fold change upregulation cf. control in broilers

Target inflammatory genes	Fold change DSS			Fold change FW			Treatment		Time		Interaction	
	2 hr	24 hr	48 hr	2 hr	24 hr	48 hr	SEM	P	SEM	P	SEM	P
IL1B	4.86	1.40	2.46	6.58	1.40	-0.13	0.831	NS	1.178	<0.05	1.666	NS
IL8L1	2.0	1.7	1.1	13.2	4.8	0.4	1.88	NS	2.30	NS	3.25	NS
CCL	2.23	1.26	3.51	3.23	0.96	1.66	1.95	NS	1.15	NS	1.576	NS
TNFSF11	1.30	1.03	0.68	1.34	-0.23	0.49	0.389	NS	0.476	NS	0.674	NS

protein (CP)), grower (12.9 MJ/kg AME, 198 g/kg CP) and finisher (13.1 MJ/kg AME, 179 g/kg CP) rations from 0–14d, 14–21d and 21–35d respectively. On day 5, birds were weighed and returned to their pen. DSS was administered to DSS assigned birds at a rate of 0.45 g/bird by oral gavage at 10am on day 5 and repeated on day 6. Feeders were removed from FW assigned bird pens on day 5 to result in an 18 hr feed withdrawal period. At 2hr, 24 hr and 48 hr after the second dose of DSS, five birds/pen were euthanised and dissected to obtain samples of the jejunum, ileum and caecum and rapidly immersed in RNAlater™ solution (Sigma, Dorset, UK). The ileum samples were pooled on a pen basis and used in customised RT2 Profiler gene expression PCR assay plates containing four target inflammatory genes (IL1b, IL8L1, CCL4 & TNFSF11), a housekeeping gene and three essential control assays. Production performance was determined on a per pen basis (feed intake (FI), liveweight gain (LWG) and feed conversion ratio (FCR) from 0–35d) and two birds/pen were euthanised at 21d and 35d to obtain ileal contents to determine ileal digestibility of dry matter (DM) and CP of the starter/grower ration respectively. Upregulation in comparison to the control treatment was expressed as fold changes. Results were analysed by ANOVA and linear regressions were performed between ileal DM and CP digestibility and fold changes IL8L1 expression.

Results

There were no significant treatment effects on bird performance. On average, 35d weight was 3.0 kg (SED = 0.036, P = NS), overall FI was 4329 g (SED = 94.2, P = NS), overall LWG was 2953 g (SED = 36.2, P = NS) and overall FCR was 1.47 (SED = 0.038, P = NS). Ileal digestibility of CP in the grower diet was lower for birds offered the DSS and FW treatments than for birds offered the control treatment (0.838 and 0.831 vs 0.860 respectively, SEM = 0.0037, P < 0.001). There was no significant interaction on fold change upregulation due to treatment or time of sampling (Table 1). Three of the assays (IL1B, IL8L1 and CCL4) were numerically upregulated relative to the control but the

differences were non-significant. There was a significant effect of sampling time on upregulation with sampling at 2 hr resulting in the highest level of upregulation for both DSS and FW treatments (Table 1). There were weak, non-significant relationships (P > 0.05) between ileal digestibility and IL8L1 expression.

Conclusion

The exceptional performance of the birds, thought to be attributed to the experimental research conditions, indicates that the treatments may not have sufficiently compromised the gut health of the bird to incite a large inflammatory response. However, despite a non-significant effect on overall performance, the reduction of digestibility of CP in the grower phase with FW and DSS indicates that there was a disruption in the digestive process. Three of the assays (IL1B, IL8L1 and CCL4) showed some potential as gut inflammatory markers in that they were upregulated relative to the control although not significantly. There was large variability within the treatment groups and many more birds would be required to make a robust model. The optimum sampling time would appear to be at 2 hrs after treatment and this is the time point where future work should focus. In conclusion, DSS and FW disrupted the digestive system and caused an up regulation of specific genes therefore, with more replication, there is the potential to develop a robust model.

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