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# Soya bean meal increases litter moisture and foot pad dermatitis in maize and wheat based diets for turkeys but maize and non-soya diets lower body weight

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# ABSTRACT

1. A 2  $\times$  2 factorial experiment was conducted to compare the effects of wheat or maize based diets differing in dietary electrolyte balance (DEB) on litter moisture and foot pad dermatitis (FPD) at 4, 8 and 12 weeks of age in heavy-medium turkeys. A second objective was to investigate the effects on foot pad dermatitis of the interaction between dietary composition and artificially increasing litter moisture by adding water to the litter.

2. High DEB diets contained soya as the main protein source whereas low DEB diets did not contain soya bean meal. Diets were formulated to be iso-caloric and iso-nitrogenous in each of 3 successive 4-week phases following recommended dietary compositions. DEB concentrations were 330, 290 and 250 mEq/kg in high DEB diets and 230, 200 and 180 mEq/kg in low DEB diets.

3. Litter moisture and mean FPD score were higher in turkeys fed on high DEB diets compared with low DEB diets whereas there was no difference between maize and wheat.

4. Food intake was similar and body weight was lower after litter moisture was artificially raised in the wet compared with the dry litter treatment and there was no interaction with dietary composition.

5. Mean body weight and feed intake were higher in turkeys fed on wheat compared with maize and in high DEB compared with low DEB diets at 12 weeks of age.

6. Lowering dietary DEB for turkeys may improve litter moisture and lower the prevalence of FPD in commercial turkey flocks.

#### ARTICLE HISTORY

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#### **KEYWORDS**

Cereal; dermatitis; diet; electrolyte balance; feed; feed intake; litter moisture; protein

# Introduction

The prevalence of foot pad dermatitis (FPD) in growing poultry is an adverse welfare and economic factor that is closely related to high concentrations of litter moisture (Wu and Hocking 2011; Weber Wyneken et al. 2015). The banning of animal by-products as protein sources in diets for livestock in Europe (European Commission Decisions No. 98/272/CE and 2000/374/CE) and the high cost of alternative protein sources, has resulted in the presence of high concentrations of soya bean meal in rations for poultry. Soya bean meal contains high concentrations of potassium and  $\alpha$ -galactosides that have been associated with increased water intake and wet litter and an increase in the prevalence of FPD (Hurwitz et al. 1983; Bedford 1995; Smith et al. 2000; Eichner et al. 2007).

In a companion paper, Veldkamp et al. (2017) demonstrated that decreasing the dietary electrolyte balance (mEq) by removing soya bean meal in diets for turkeys from hatch to slaughter resulted in drier, more friable litter and a lower prevalence of FPD. They also showed that decreasing the crude protein concentration of the diets had variable results on litter moisture but did result in lower average FPD scores to 84 d of age. However, there were no differences between two lines of heavy-medium turkeys after 28 d of age.

The aim of the present experiment was to evaluate the effects of low dietary electrolyte balance (DEB), achieved by avoiding the use of soya bean meal in rations for turkeys

from hatch to 12 weeks of age, on litter moisture and FPD in both wheat- and maize-based diets. A second objective was to investigate the interaction between dietary treatment and experimentally increased litter moisture on the prevalence of FPD.

# **Materials and methods**

## Animals and husbandry

A total of 432 male B.U.T. Premium turkey poults (Aviagen Turkeys Ltd, Chester, UK) were obtained from a commercial hatchery at 1 d of age. The poults were beak trimmed by the infra-red method in the hatchery and vaccinated for Newcastle disease and turkey rhinotracheitis. The birds were distributed at random to 24 pens at a stocking rate of 18 poults/pen and individually identified by a wing band. The pens were 2.50 m wide  $\times$  1.77 m deep and were littered with pine wood shavings to a depth of about 50 mm (~2 kg/  $m^2$ ). On arrival, the poults were given water only for one h followed by feed for 2 h followed by 3 h of darkness and 3 h light repeated until the onset of the planned dark period. Thereafter the photoperiod was 16 h light and 8 h darkness per 24 h (0730-2330). The poults were brooded under a heat lamp from hatch to 26 d. Minimum ambient temperatures were set at 28°C for the first week, 26°C for the second week decreasing to 23°C at 3 weeks, 20°C at 7 weeks, 18°C at 8 weeks and 16°C at 11 weeks of age. Light intensities at d

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1, d 2 and from d 4 were 100, 50 and 12 lx, respectively. Mean relative humidity (SD) was recorded every 15 min during weeks 3–4, 7–8 and 11–12 and averaged 30 (4.2), 61 (3.8) and 67 (5.3), respectively. At 10 weeks of age, the litter in half the pens (3 replicates per treatment) was moistened with the same volume of water from a garden watering can to create litter with a score 4 on the Tucker and Walker (1999) scale (wet, soggy, leaving an imprint) to investigate the interaction between dietary treatment and litter moisture. Thereafter, the same volume of water was added to all pens at the same time whenever it was needed to maintain a litter score of 4.

Diet composition and calculated analyses for important nutrients are presented in Table 1 and for all nutrients in the supplementary file (Supplementary Table). The feed ingredients wheat, maize, hipro soya meal, sunflower seed meal, rapeseed meal, maize gluten, peas, potato protein, fishmeal and wheat feed were analysed for crude protein (CP) content prior to feed formulation and analysed CP contents of these feed ingredients were used for final feed formulation. Feed was provided according to a three phase feeding programme in 4-week phases (0-28, 28-56 and 56-84 d of age). Wheat as well as maize based diets were formulated with or without soya bean meal and diets were formulated to be iso-caloric and iso-nitrogenous for successive 4-week phases. Energy and crude protein (CP) concentrations for 0-4, 4-8 and 8-12 weeks of age were 11.8, 12.0 and 12.2 MJ/kg and 295, 275 and 235 g CP/kg, respectively. Experimental diets were supplemented with L-lysine HCl,

DL-methionine, and L-arginine to have similar digestible amino acid contents in each feeding phase (Supplementary Table). Diets formulated with soya bean meal contained 500, 400 and 300 g soya bean meal/kg in the respective feeding phases. In diets without soya bean meal, the protein sources were sunflower meal, rapeseed meal, peas, potato protein, fish meal and wheat gluten meal. Samples of all diets were analysed at a commercial laboratory (Sciantec Analytical Services, Cawood, UK) for crude protein content by the Dumas method (N x 6.25) and were within acceptable ranges of calculated values (Supplementary Table). Electrolyte balance in the respective feeding periods for diets formulated with soya bean meal was 330, 290 and 250 mEq/kg and in diets formulated without soya bean meal it was 230, 200 and 180 mEq/kg. The starter diet was provided as crumbs followed by the second and third diets as 3 mm pellets to 10 weeks and 3 d when it was replaced with coarse mash as the pelleted diets were likely to run out during the feed recording period (11-12 weeks of age). All diets were provided by Target Feeds Ltd., Whitchurch, UK. Feed was distributed in a floor pan from hatch to d 6 and thereafter solely in a suspended tubular feeder. Water was available at all times in a suspended bell drinker.

#### **Observations**

The number of dead poults and culls were recorded daily. Average pen body weight was determined by bulk weighing on d 1 and at 4 and 8 weeks of age. Individual body weights

	Diet and age (weeks) when fed											
	0-4			4–8				8–12				
Ingredient (g/kg)	WS	WN	MS	MN	WS	WN	MS	MN	WS	WN	MS	MN
Wheat	388.9	226.4	_	_	471.2	363.7	_	_	578.7	467.0	_	_
Maize	-	-	388.6	224.7	-	-	463.4	360.4	-	-	584.2	469.6
Hipro soya meal	497.5	-	498.8	-	395.3	-	417.0	-	308.7	-	311.3	-
Sunflower 36%	-	170.0	-	170.0	-	120.0	-	120.0	-	100.0	-	100.0
Rape ext. double zero	-	80.0	-	80.0	-	60.0	-	60.0	-	50.0	-	50.0
Maize gluten 60	15.4	-	28.4	-	48.3	-	50.0	26.0	33.2	5.6	50.0	21.6
Peas	-	200.0	-	200.0	-	150.0	-	150.0	-	120.0	-	120.0
Potato protein	-	104.2	-	107.6	-	124.4	-	114.1	-	100.0	-	100.0
Fishmeal 66	-	84.8	-	89.2	-	60.0	-	60.0	-	30.0	-	30.0
Wheatfeed	-	50.0	-	50.0	-	50.0	-	50.0	-	50.0	-	50.0
Soya oil	40.1	49.4	26.1	44.7	32.2	36.4	17.3	23.7	30.8	39.0	5.8	20.1
Monocalcium phosphate	27.4	21.4	27.9	21.5	22.7	18.8	23.3	19.2	19.9	17.5	20.7	18.2
Limestone	12.3	1.2	12.3	0.5	12.8	4.8	12.7	4.8	12.3	8.2	12.3	8.2
Sodium chloride	2.1	-	2.1	-	2.0	0.8	2.2	0.9	2.1	1.6	2.1	1.7
Premix <sup>a</sup>	4.0	4.0	4.0	4.0	4.0	4.0	4.0	4.0	4.0	4.0	4.0	4.0
DL methionine	3.8	2.1	3.6	1.9	3.1	1.8	2.9	1.5	2.5	1.6	2.3	1.4
Lysine mono HCl	2.9	0.8	2.9	0.4	2.8	-	2.3	-	2.5	0.3	2.4	0.2
L threonine	1.1	-	0.9	-	0.5	-	0.2	-	0.3	-	-	-
L arginin	-	0.9	-	0.8	-	0.5	-	0.7	-	0.5	-	0.5
Sodium bicarbonate	2.2	2.4	2.2	2.3	2.6	2.5	2.3	2.3	2.5	2.3	2.5	2.1
Phytase <sup>b</sup>	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0
Xylanase <sup>c</sup>	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0
Elancoban G200 <sup>d</sup>	0.4	0.4	0.4	0.4	0.4	0.4	0.4	0.4	0.4	0.4	0.4	0.4
Calculated composition <sup>e</sup>												
Energy ME, MJ	11.8	11.8	11.8	11.8	12.0	12.0	12.0	12.0	12.2	12.2	12.2	12.2
Protein	296.0	296.0	296.0	296.0	275.0	275.0	275.0	275.0	235.0	235.0	234.4	235.0
Ca	13.5	13.5	13.5	13.5	12.5	12.5	12.5	12.5	11.5	11.5	11.5	11.5
Available P	7.6	7.6	7.6	7.6	6.7	6.7	6.7	6.7	6.1	6.1	6.1	6.1
DEB, mEq	335.2	228.2	327.4	224.1	290.6	204.7	291.5	194.2	251.1	186.3	240.2	174.4

WS: wheat soya; WN: wheat non-soya; MS: maize soya; MN: maize non-soya; DEB: dietary electrolyte balance (Na<sup>+</sup>+K<sup>+</sup>-Cl<sup>-</sup>).

<sup>a</sup>Supplying retinyl acetate 4.09 mg/kg, cholecalciferol 125 mμg/kg, α-tocopherol 100 mg/kg, menadione 4 mg/kg, thiamine 5 mg/kg, pyridoxine 7 mg/kg, cobalamin 0.04 mg/kg, niacin 150 mg/kg, pantothenic acid 25 mg/kg, folic acid 4 mg/kg, ascorbic acid 200 mg/kg, choline chloride 1200 mg/kg; I 2 mg/kg, Se 0.2 mg/kg, Cu 20 mg/kg, Fe 50 mg/kg, Mn 120 mg/kg, Zn 100 mg/kg, Co 0.5 mg/kg, Mo 0.5 mg/kg.

<sup>b</sup>Phytase enzyme.

<sup>d</sup>Monensin sodium coccidiostat, Elanco Animal Health, Basingstoke, UK.

<sup>e</sup>Full details are in the Supplementary Table.

 Table 1. Dietary ingredients and calculated composition.

<sup>&</sup>lt;sup>c</sup>Xylanase enzyme, DSM Nutritional Products Ltd., Basel, Switzerland.

were recorded at 12 weeks of age. Pen mean daily feed intake was measured at 3–4, 7–8 and 11–12 weeks of age. Litter moisture content was determined by sampling the full depth of litter from 4 locations approximately 80 cm from the corner of each pen. After thoroughly mixing the litter a subsample of 100–200 g of litter was placed in a disposable container and dried in a fan oven (Gallenkamp size one fan incubator) at 60°C for 7 d as described by Mayne et al. (2007). Litter moisture was calculated as the percentage loss of weight of the fresh litter. Foot pad lesions were scored by one person at 4, 8 and 12 weeks of age on a 5 point scale (0 = unaffected and 4 = more than half the footpad affected), as described previously (Hocking et al. 2008). The highest score for both feet only was recorded.

# **Bird welfare**

All experiments and procedures were conducted after ethical approval under project licence number PPL60/4506.The health of the turkeys was inspected on a daily basis and severely affected birds were humanely killed with an intravenous injection of sodium pentobarbital (Euthatal, Merial, Toulouse, France). All turkeys were killed at the end of the experiment by an approved method (Euthatal injection or Cash captive bolt Poultry Killer).

## Statistical analyses

The experiment was a randomised block design with each pen assigned to one of the 8 treatments. The statistical model for mean pen body weight, feed intake and litter moisture included effects for block, pen and treatment effects for protein (soya vs. non-soya) and cereal (wheat vs. maize), with age (4, 8 and 12 weeks) nested within pen. All 2- and 3-way interactions were also evaluated. Litter treatment at 10 to 12 weeks (wet vs. dry) was included in the model. Body weight and feed intake were transformed by taking natural logarithms to overcome a positive mean-variance relationship in the combined analysis for all three ages. The 12-week data including mean FPD score were analysed by a model with effects for block, litter treatment, cereal, protein source and their 2- and 3-way interactions. Overall mortality from hatch to 12 weeks was modelled by a Generalised Linear Model with binomial errors. All analyses of variance were conducted in GenStat v.13.

# Results

There were no observations of birds with FPD at 4 or 8 weeks of age. The number of surviving turkeys with FPD scores of 0, 1, 2, 3 and 4, respectively, at 12 weeks were 334, 20, 20, 13 and 6. Two- and three-way interactions were not statistically significant. Mean pen FPD score was higher in turkeys fed on soya compared with non-soya protein sources (0.47 vs. 0.13, SED 0.139, P = 0.028). Pen mean scores for birds fed on maize compared with wheat diets, respectively, were 0.42 and 0.19 (SED 0.139, not significant) and 0.29 and 0.32 (SED 0.139, not significant), respectively, for wet and dry litter.

The combined analyses of litter moisture, body weight and feed intake at three ages showed a highly significant effect of protein source on litter moisture (Table 2). Litter moisture was higher in turkeys fed on soya compared with non-soya diets at 4, 8 and 12 weeks of age (Table 3). Mean **Table 2.** Summary of the analysis of variance (probability of statistical significance of effects) of the combined 4, 8 and 12 week pen means for body weight, feed intake, water intake and litter moisture of male BUT Premium turkey poults fed on iso-nutritional diets composed of maize or wheat as the cereal source and soya or non-soya sources of protein.

Effect	Litter moisture, %	Body weight, ln (kg)	Feed intake, In (g/d)
Cereal	0.688	0.014	0.670
Protein	<0.001	0.253	0.048
Cereal $\times$ protein source	0.642	0.094	0.036
Litter treatment	0.107	0.460	0.604
Age	<0.001	<0.001	<0.001
Age $\times$ cereal	0.920	0.348	0.059
Age $\times$ protein source	0.392	<0.001	<0.001
Age $\times$ cereal $\times$ protein	0.815	0.565	0.507
source			

Figures in bold are statistically significant (P < 0.05).

Table 3. Litter moisture, body weight and feed intake at 4, 8 and 12 weeks of age analysed separately of male BUT Premium turkey poults fed on isonutritional diets composed of maize or wheat as the cereal source and soya or non-soya sources of protein.

		Age (weeks)				
Cereal	Protein	4	8	12		
Litter moisture, %						
Wheat	Soya	18	41	51		
	Non-soya	14	29	43		
Maize	Soya	19	39	49		
	Non-soya	14	31	43		
SED		2.5	4.3	4.5		
Significance						
Cereal		NS	NS	NS		
Protein		*	**	*		
Cereal $\times$ protein		NS	NS	NS		
Body weight, kg						
Wheat	Soya	1.45	5.83	11.45		
	Non-soya	1.49	5.94	11.04		
Maize	Soya	1.46	5.77	11.20		
	Non-soya	1.44	5.76	10.60		
SED		0.024	0.077	0.104		
Cereal		NS	*	***		
Protein		NS	NS	***		
Cereal $\times$ protein		NS	NS	NS		
Feed intake, g/d						
Wheat	Soya	109	371	586		
	Non-soya	116	381	538		
Maize	Soya	114	360	615		
	Non-soya	115	361	527		
SED		2.4	9.8	11.1		
Significance						
Cereal		NS	*	NS		
Protein		*	NS	***		
Cereal $\times$ protein		NS	NS	*		

NS: not significant; \**P* < 0.05; \*\**P* < 0.01; \*\*\**P* < 0.0.

litter moisture at 12 weeks for the soya and non-soya treatments were 50.1 and 43.0%, (SED 1.07%, P < 0.001) and for the wheat and maize diets they were 45.8 and 47.3% (SED 1.07%, not significant), respectively. Litter moisture for the dry and wet treatments at 12 weeks were 40.7 vs. 52.4% (SED 1.07%, P < 0.001); there was no difference in mean food intake (558 vs. 553 g/d, SED 11.5) between the dry and wet litter treatments whereas body weight was lower in the wet compared with the dry litter treatment (10.92 vs. 11.22 kg, SED 0.074, P < 0.01).

The combined analyses showed a highly significant (P < 0.001) protein x age interaction for both body weight and feed intake (Table 2). The data for each age were therefore analysed separately using the same model without the effect of age and by omitting the effect of litter treatment for the 4- and 8-week data. The results of these analyses are presented in Table 3.

Mean body weights in the combined analysis were similar at 4 weeks whereas at 8 and 12 weeks body weights were higher in turkeys fed on the soya compared with the non-soya diets leading to a significant age x protein interaction. Marginal means (back transformed, kg) for soya and non-soya diets were 7.280 (1.45) and 7.289 (1.46) at 4 weeks, 8.665 (5.80) and 8.674 (5.85) at 8 weeks and 9.334 (11.32) vs. 9.289 (10.82) (ln(kg), SED 0.011). At 12 weeks, the mean body weights from the separate analysis were 11.25 compared with 10.90 kg (P < 0.001, SED 0.073), respectively. Body weight in the combined analysis was also higher (P < 0.05) in wheat compared with maize diets: means (back transformed, kg) were 8.411 (4.50) and 8.433 (4.60) (ln(kg), SED 0.0078), respectively. At 12 weeks the mean body weights were (P < 0.001, 11.25 vs. 10.90 kg, SED 0.073).

Feed intakes in the combined analysis of turkeys fed on the different diets were numerically similar at 4 weeks whereas at 8 weeks feed intake was higher in turkeys fed on the wheat diets compared with the maize diets (P < 0.05). However, at 12 weeks, feed intake was higher (P < 0.001) in soya compared with non-soya diets leading to an age  $\times$  protein source interaction (P < 0.001): means (back transformed, g/d) were 4.714 (111) and 4.749 (115) at 4 weeks, 5.899 (365) and 5.919 (372) at 8 weeks and 6.402 (603) vs. 6.277 (532) at 12 weeks, respectively, (ln(g/d), SED 0.0180). (Mean feed intakes at 12 weeks in the separate analysis for the soya and non-soya diets respectively were 587 vs. 524 g/d (SED 11.5, P < 0.001, Table 3). There was also a significant cereal × protein source interaction (P < 0.05) in the combined analysis for feed intake: marginal means (back transformed, g/d) for maize and wheat were 5.682 (294) and 5.662 (288) for the soya diets compared with 5.634 (280) and 5.663 (288) for the non-soya diets (ln(g/d), SED 0.0153).

There were no statistically significant differences in mortality between the diets. Back transformed means on the original scale for wheat and maize diets, respectively, were 8.5 and 8.4% and for soya compared with 6.7 and 10.5% for the nonsoya diets (predicted means on the binomial scale for maize and wheat diets were -2.38 and 2.39 and for soya and low nonsoya diets they were -2.63 and 2.15; SED 0.365).

# Discussion

The current results support the hypothesis that the increased reliance on soya bean meal has been accompanied with an adverse effect on litter moisture and FPD. The effect of soya bean meal on litter moisture and FPD was similar in both maize- and wheat-based diets. The lower body weight of turkeys after exposure to wet litter is consistent with other evidence that wet litter may depress economic performance (Wu and Hocking 2011; Hocking and Wu 2013) as well as increasing the susceptibility of turkeys to develop FPD (Mayne et al. 2007).

The maize-based diets were associated with lower body weights and feed intakes at 8 weeks of age. At 12 weeks of age live weight was higher in wheat- than in maize-based diets and also in soya compared with non-soya diets. In contrast to the higher body weights, mean feed intake was lower at 12 weeks of age in the birds fed on the wheatcompared with maize-based diets, implying a better feed conversion ratio (FCR). Moreover, the difference between the soya and non-soya diets was greater in the maizecompared with the wheat-based diets resulting in a significant cereal  $\times$  protein source interaction. It is possible that the line of turkeys in this experiment show adaptation to a wheat-soya based diet similar to that used in the breeding programme.

The mean body weight of the birds was up to 16% heavier than the published breed target at 12 weeks of age and feed intake was greater than anticipated. It was clear after 10 weeks of age that there would be insufficient pelleted feed for the last week of the experiment and further supplies had to be ordered that, because of time constraints, could only be supplied in meal form. To ameliorate the effects of changing from pellets to meal during the feed intake recording period at 11-12 weeks, the meal form of each diet was introduced at 10 weeks and 3 d of age. The authors cannot therefore be certain that diet related differences in body weight and feed intake at 12 weeks were not affected by palatability differences or selection of feed ingredients associated with differences in particle size. However, the major conclusions of this research regarding the differential effects of cereal and protein source on litter moisture and FPD are not affected by the change in diet form.

Anticipated feed intake at 12 weeks of age from technical information provided by the breeding company (http:// www.aviagenturkeys.com, accessed 2 November 2017) was 429 g/d/bird compared with 527–615 g/d in this study. However, the expected feed intake of males weighing 11 kg was about 450 g/d and consumption of +20% feed above this gives an estimated feed intake of 540 g/d. The latter is comparable with the present results given that some spillage was likely in these mash diets, although noticeable spillage was only observed in the maize soya diet.

The diets were designed to be iso-caloric and iso-nitrogenous but wheat also contains high concentrations of nonstarch polysaccharides (NSPs) and crude fibre. Enzymes (Table 1) were therefore added to all diets to negate the possible effect on litter moisture and FPD of undigested NSPs in wheat compared with maize diets. Higher concentrations of crude fibre in the wheat based diets (Supplementary Table) were unlikely to affect performance or litter condition: crude fibre can be utilised to some extent by turkeys and concentrations of 60 g fibre/kg in the diet did not result in decreased performance after 6 weeks of age and may in fact be beneficial (Sklan et al. 2003).

Taken together, the results of the current experiment and the parallel study in Italy (Veldkamp et al. 2017) confirm the importance of DEB and the role that soya bean meal has in increasing litter moisture and the prevalence of FPD. Combined with evidence that FPD may be painful (Sinclair et al. 2015; Weber Wyneken et al. 2015), it is imperative that regular monitoring and amelioration of litter conditions are incorporated into flock management procedures to minimise FPD; specifically, the increase in litter moisture with time must be minimised (Wu and Hocking 2011; Weber Wyneken et al. 2015). A simple but effective system for litter scoring was recently published (Giacomelli et al. 2018) that will facilitate this process even if manipulation of dietary composition and DEB will be limited by practical and economic constraints. Further research to determine the optimum dietary concentration of soya to minimise DEB and the prevalence of FPD in growing turkeys is required.

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## **Disclosure statement**

No potential conflict of interest was reported by the authors.

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