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Evaluation of the effect of different wheats and xylanase supplementation on performance, nutrition and energy utilisation in broiler chicks

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1Evaluation of the effect of different wheats and xylanase supplementation on
2performance, nutrient and energy utilisation in broiler chicks
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16Abstract

17The aim of this study was to evaluate the performance, nutrient utilisation and energy 18metabolism of broiler chicks fed eight different wheat samples, supplemented or not 19with xylanase. Seven-hundred sixty eight male broilers (1-day old) were distributed to 2016 experimental treatments (six replicates per treatment). The treatments were in a 21 factorial arrangement with eight different wheats and two levels of xylanase (0 or 2216,000 BXU/kg). The predicted apparent metabolisable energy (AME) of the wheat 23samples ranged from 13.0 and 13.9 MJ/kg and all diets were formulated to contain the 24same amount of wheat. Body weight gain (BWG) and feed intake (FI) were measured at 2521 d, as was jejunal digesta viscosity, and feed conversion ratio (FCR) calculated. On 26day 24, one representative bird pen was selected to calculate whole body energetics. At 2721 d, three chicks per replicate were randomly allocated to metabolism cages for energy 28and nutrient utilisation determinations, and were continued on the experimental diets 29until 24-d-old. No interactions were observed for any performance response variables, 30ileal nutrient utilisation or digesta viscosity. Xylanase improved BWG and reduced FCR 31and digesta viscosity (P < 0.05). Wheat influenced dry matter (DM) utilisation and 32xylanase increased ileal digestible energy (P = 0.04). Xylanase also improved (P < 0.04). 330.05) DM and nitrogen retention. Apparent metabolisable energy and AME corrected 34for nitrogen (AMEn) were subject to an interaction whereby wheats 2 and 6, which 35returned the lowest AME and AMEn values, responded to xylanase supplementation 36and the remainder did not. Net energy for production and the efficiency of energy use 37 for production were not influenced by xylanase, but were affected by wheat (P < 0.05). 38Despite the significant differences between wheats with regards to their nutrient 39utilisation and energy metabolism in birds, xylanase removed this variance and resulted 40in more homogeneous performance.

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41Keywords: Wheat, Near-infrared spectroscopy, Xylanase, Animal performance, Nutrient 42release, energy, Broiler chickens 431. Introduction

44Variation in the nutritive value of wheat samples is a reflection of genetic and 45environmental effects, and the economic impact of these variations on poultry 46performance highlights the need for improved predictors of wheat quality (Yegani and 47Korver 2012). This is a concern for plant breeders, farmers and animal nutritionists. 48Thus, nutritionists need to know the nutritional requirements of commercial poultry, and 49be able to determine or predict the nutritive value of each batch of raw material in an 50accurate and timely manner (van Kempen and Simmins 1997).

51 The use of Near-Infrared Spectroscopy (NIRS) provides an opportunity to determine 52the chemical composition of feedstuffs and their nutritive value before inclusion in the 53diet (Olukosi et al., 2011; Owens et al., 2009). The information from NIRS can be used 54to reduce or minimize nutrient imbalances in commercial rations fed to the animals. 55However, there are potential errors associated with NIRS technology such as sample-56related and chosen reference method errors which can lead to high values for coefficient 57 of variation (Yegani and Korver 2012), and as a result care must be taken in establishing 58NIRS calibration to ensure it is robust, precise and accurate. Near-Infrared Spectroscopy 59 calibrations now exist which can predict non-starch polysaccharide (NSP) and energy 60contents of wheat. In particular xylans, is often considered an anti-nutrient in wheat, and 61as a result variation in content of this component between wheat samples may 62contribute to differences in nutritive value. Xylanases are the major enzymes involved 63in arabinoxylan degradation, hydrolysing the 1,4-β-D-xylosidic linkage between xylose 64 residues in the backbone in a random manner (Mendis et al. 2016), therefore it is 65hypothesised that their supplementation in poultry feed may balance animal 66performance although differences in the nutritive value of different wheat origins. This 67 work was undertaken to determine if such a calibration by NIRS accurately predicts

68animal performance, and if so whether the application of an NSP-degrading xylanase 69would reduce the performance differences between samples of wheat which differ in 70NSP content (Bedford, 2000).

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732. Materials and Methods

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75All the experimental procedures received prior approval from the Scotland's Rural 76College's Animal Experiment Committee.

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782.1. Birds and experimental design

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80A total of 768 one-day old male broiler chicks (Ross 308) obtained from a commercial 81hatchery were used in the study for two experiments to determine growth performance 82and whole-body energy metabolism (Exp. 1) and nutrient utilisation (Exp. 2) responses. 83For Exp. 1 (n = 768) and for Exp. 2 (n = 288), birds were allocated to 16 experimental 84treatments in a randomized complete block design with an 8 × 2 factorial arrangements 85of treatments (eight wheat samples and two levels of xylanase), having in both 86experiments six replicates per treatment. Throughout the study, feed and water were 87supplied *ad libitum* and animals were raised under controlled conditions of light and 88temperature, as breeder recommended.

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902.1.1. Experiment 1

92Birds were reared up to day 24 in floor pens. All broiler chickens and feed were 93weighed on day 0 and 21 to calculate growth performance responses: body weight gain 94(BWG), feed intake (FI) and feed conversion ratio (FCR). On day 21, two chickens 95were randomly selected and euthanized by an overdose of sodium pentobarbital and 96jejunal digesta were collected for viscosity measurement. On day 24, one representative 97bird (on BW basis) per floor pen was selected and fasted prior to euthanasia to calculate 98the whole body energetics.

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1002.1.2. Experiment 2

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102On day 21, three chicks were randomly selected from each of the 96 floor pens and 103transferred to 96 metabolism cages (for energy and nutrient utilisation trial) where 104chickens continued to receive the corresponding diets until 24 days of age. Excreta and 105ileal digesta were collected on day 24 and pooled on a cage basis for calculation of 106nutrient utilisation.

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1082.2. Diets and wheat selection

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110Starter experimental diets based on wheat and soybean-meal were formulated to be 111marginally lower in metabolisable energy (ME) than Ross 208 requirements (Table 1). 112Eight wheat samples originating from Germany and United Kingdom were obtained. 113Dry matter (DM), gross energy (GE), fat, nitrogen (N), calcium (Ca) and the 114phosphorous (P) contents of wheat samples were chemically analysed and further NIRS 115analyses were performed (Tables 2 and 3). A fixed amount of each wheat (58.6%) was 116used in the formula regardless of their chemical composition. Diets were predicted to 117contain 12.8 ME MJ/kg based on assumed average wheat apparent ME (AME) 58.6% 118came from wheat grain. Control diets were supplemented with 16,000 BXU/kg of 119xylanase following supplier recommendations (Econase XT, AB Vista, Marlborough, 120UK; 160,000 BXU/g), resulting in 16 experimental diets in total. All diets contained 121phytase supplemented at 500 FTU/kg (Quantum Blue, AB Vista, Marlborough, UK; 1225000 FTU/g). Activity of xylanase and phytase were determined using the reference 123method of analysis recommended by the supplier. Titanium dioxide (0.3%) was added 124to all the diets as an indigestible marker. Feed samples were taken at the beginning and 125throughout the experimental period for DM, N, fat and GE analysis.

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1272.3. Jejunal viscosity

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129Approximately 1.5 g (wet weight) of the fresh jejunal digesta were analysed according 130to Bedford et al. (1991). The viscosity (expressed as centipoise units, cP = 1/100 dyne 131sec/cm²) was determined using a Brookfield DV II digital viscometer.

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1332.4. Nutrient utilisation and total tract retention

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135Total tract retention and ileal nutrient utilisation were calculated using the index method 136(Olukosi et al., 2007), with titanium dioxide as the indigestible marker.

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1382.5. Net energy and nutrient accretion

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140Net energy for production (NEp), heat production (HP) and carcass fat and protein 141accretion were determined using the comparative slaughter technique as described by 142Olukosi et al. (2008). Briefly, six birds were euthanized at day 0 without feeding and 143kept frozen prior to processing and chemical analyses. On day 24, following euthanasia 144the carcasses were frozen and ground prior to freeze drying. Gross energy, N and fat 145contents were analysed. All the calculations for NEp, ME intake, HP as well as the 146efficiencies of energy for fat and protein retention (Fat-ER and CP-ER, respectively) are 147as described previously (Olukosi et al., 2008a). Net energy for production and HP were 148expressed per kg feed by dividing the total NEp (MJ) or HP (MJ) by kilogram of feed 149intake.

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1512.6. Chemical analyses

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153Ileal digesta and excreta were analysed for DM, N, fat and GE. Dry matter was 154determined by drying the samples in a drying oven (Uniterm, Russel-Lindsey 155Enginering Ltd., Birmingham, England, UK) at 105 °C for 24 h (method 934.01; 156AOAC, 2006). Total N content was determined by the combustion method (method 157968.06; AOAC, 2006). Gross energy was determined in an adiabatic oxygen bomb 158calorimeter (model 6200; Parr Instruments, Moline, IL) using benzoic acid as an 159internal standard. Titanium concentration in samples of diets and ileal digesta was 160determined using the method of Short et al. (1996).

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1622.7. Statistical analyses

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164Pen served as the experimental unit for FI, BWG and FCR, and cages as experimental 165unit for nutrient utilisation, jejunal viscosity, net energy and nutrient accretion. Data 166were analysed using the PROC MIXED command of SAS (SAS Inst. Inc., Cary, NC). 167When effects were found to be significant, treatment means were separated using 168Tukey's Highly Significant Difference test. Statistical significance was accepted at P < 1690.05 and trends were discussed at P < 0.10.

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1713. Results

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1733.1. Wheat nutritive value by NIRS and chemical analyses

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175The chemical analysis of the wheat samples indicates that they are all very similar in 176chemical composition and GE (Table 2). The predicted nutritive values by NIRS 177showed slightly more variability in nutrient composition between wheat varieties (Table 1783), but remained close to expected average values. The predicted GE was 179underestimated while the predicted fat content was higher than chemically analysed. 180The predicted AME of wheat varieties ranged from 13.0 to 13.9 MJ/kg (CV < 2%). 181There was a great deal of variability (CV > 10%) in the predicted contents for crude 182protein, acid detergent fibre, β -glucan, lignin and total non-starch polysaccharides, but 183low variability (CV < 8%) in all other analysed chemical components, including amino 184acids.

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1863.2. Feed enzyme activity, growth performance and jejunal viscosity

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188Enzyme activities in feed samples were close to expected (16,038 BXU/kg average 189value analysed in all the xylanase-supplemented diets). No interactions were observed 190in any of the performance parameters measured (Table 4). There were no effects of 191wheat on performance or jejunal digesta viscosity. Nevertheless, improvements in 192performance were observed when xylanase was supplemented, regardless of wheat 193sample. Xylanase application resulted in a near significant 20 g (P = 0.077) increase in 194BWG. Although FI was not influenced by xylanase, FCR was significantly improved by 195four points better (1.33 vs. 1.37, xylanase vs. control, respectively; P = 0.003). Xylanase 196supplementation also reduced viscosity of jejunal digesta (3.32 vs. 2.34 cP, for control 197and xylanase supplemented diets, respectively; P < 0.001). In the diets without xylanase 198supplementation, there were low and non-significant correlations between nutrient 199content of the wheats and bird FCR (Table 5). For the birds receiving xylanase 200supplemented diets, FCR was positively correlated with the analysed P and the 201predicted contents by NIRS of NDF, total and soluble AX as well as insoluble NSP. In 202addition, FCR was positively correlated with the analysed fat content.

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2043.3. Nutrient utilisation and total tract retention

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206No interactions between the main factors were observed for any of the ileal nutrient 207utilisation results (Table 6). The DM utilisation of wheat 3 was significantly lower 208compared with wheats 6, 7 and 8 (P < 0.05), whereas wheats 1, 2, 4 and 5 had 209intermediate values. Wheats 7 and 8 had greater energy utilisation (P < 0.001) compared 210with wheats 1, 2, 3 and 5, whereas wheats 4 and 6 where in between. Xylanase 211supplementation increased ileal utilisation of energy (IDE) measured as MJ/kg (P =2120.04), regardless of wheat. Ileal N utilisation tended to be influenced by wheat (P =2130.06), and was not influenced by xylanase supplementation.

214There were significant interactions of the main factors for all total tract measurements 215(P < 0.001). Xylanase supplementation improved the retention of DM and N as well as 216AME and AMEn for diets based on wheats 2 and 6. For those diets based on wheats 3,

2174, 5 and 8 xylanase, inclusion led to no effect or marginally lower results in total tract 218retention of N, AME and AMEn.

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2203.4. Net energy and nutrient accretion

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222There were no interactions between wheat and xylanase for any energy utilisation and 223efficiency responses, except for HP and Kre-protein, and no xylanase effect on any of 224the responses (Table 8). Net energy for production and K_{RE} were greater (P < 0.05) for 225wheat 2 compared with wheats 4, 5 and 7, but similar, although numerically higher, than 226the other wheats. Energy retained as protein was greater (P < 0.05) for wheats 3, 4 and 5 227compared with wheats 7 and 8. Energy retained as fat and Kre-fat was greater (P < 0.05) 228for wheat 2 than wheats 1, 3, 4 and 5. The interaction observed for HP (P = 0.02) was 229explained by xylanase supplementation increasing HP when birds were fed wheats 2 230and 6 (data not shown), but decreased HP for wheat 8, with no effect observed for the 231remaining wheats. The interaction noted for Kre-protein (P = 0.006; data not shown) 232was due to xylanase addition resulting in birds fed wheats 3 and 8 supplemented being 233more efficient in protein accretion, whereas it was reduced for wheats 2 and 6, with no 234effect on the remaining wheats.

235

2364. Discussion

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238It is well known that wheats, even of the same variety, can vary in both chemical 239composition and nutritive value (Theander et al., 1989). The current study investigated 240the effect of wheat sample and xylanase supplementation on the performance of broilers 241fed starter diets. In spite of the variability found between wheats in both the analysed

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242chemical composition and that predicted by NIRS, performance was not affected. 243Nevertheless, supplementation with xylanase improved BWG, FCR and reduced digesta 244viscosity, as has been shown in numerous studies (Olukosi et al., 2007; Wu et al., 2004; 245Zyla et al., 1999). Arabinoxylan is the main NSP in cereals, representing about 60-70% 246in the cell wall endosperm cells an aleurone layer. Although AX from different sources 247 differs in their substitution along the xylan backbone, a general structure can be 248assigned for AX: a backbone of β -(1,4)-linked xylose residues, which are substituted 249with arabinose residues on the C(O)-2 and/or C(O)-3 position and phenolic acids can be 250linked on the C(O)-5 position of arabinose. The structure of AX leads to high water 251holding capacity in the gastrointestinal tract resulting in high viscosity, and as a 252consequence animal production process is less efficient. Xyalanases cleave AX by 253 internally hydrolysing the β -1,4- β -D-xylosidic linkage between xylose residues giving 254small fragments of oligosaccharides with high or low degree of substitution (Mendis et 255al. 2016). The successful exposure of xylanase to different wheats with variations in the 256level content of soluble NSP makes them a feasible choice to mitigate the negative 257 effects of arabinoxylan (AX) in monogastrics (Bedford 2000).

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259Feed conversion ratio was not correlated with any of the analysed or predicted 260composition values of wheat without xylanase, but those supplemented with the enzyme 261had an unexpected positive correlation with the predicted contents of fat and fibre 262components (NDF, AX, soluble AX and total insoluble NSP). These findings are 263puzzling and suggest that the presence of more fibre (substrate) when the enzyme is 264present resulted in poorer performance but that the presence of this fibre in the absence 265of the enzyme was, if anything, marginally beneficial. Scott et al. (1999) found a 266significant relationship between predicted AME and FCR in wheat based diets with 267enzymes (r = -0.46), similarly found in this study (r = -0.45).

268

269Non-starch polysaccharide degrading enzymes reduce digesta viscosity in the animal by 270shortening the molecular weight of NSP and also partly remove the nutrient 271encapsulation effect of the cell wall components and, as a consequence, nutrient 272absorption is promoted and growth performance maximized (Masey O'Neill et al., 2732012, 2014a,b; Persia et al., 2002). In this study the measured intestinal viscosity of all 274samples was extremely low in comparison with the literature, which suggests that the 275wheat samples employed were not particularly challenging from a viscosity viewpoint. 276In a similar study, xylanase supplementation improved performance in broilers fed 277different Chinese maize samples varying in chemical composition, improving the 278homogeneity in animal flocks with NSP-ases addition (Masey O'Neill et al., 2012).

280Some studies have reported improved performance and energy utilisation when NSP-281enzymes are used in diets based on wheat, rye, barley (Bedford and Morgan, 1996; 282Bedford and Schulze, 1998) or maize (Masey O'Neill et al., 2012), but other studies 283have only shown improvements in animal performance without changes in nutrient 284utilisation (Hong et al., 2002; Wu et al., 2004). Wheat sample influenced ileal DM, N, 285energy utilisation and IDE. In particular, wheats 6, 7 and 8 were particularly good 286samples and this coincided with wheats 7 and 8 having the lowest viscosity. On the 287other hand, wheats 6, 7 and 8 also had lower contents of N (and predicted crude 288protein), acid detergent fiber, AX and NSP compared with the other wheats. Xylanase 289use increased energy utilisation and AME. Aside from the effect of reducing viscosity, 290there may be an additional benefit of increasing the permeability of the aleurone layer.

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291This may enhance contact with digestive enzymes and their substrates, for better 292nutrient utilisation (Parkkonen et al., 1997).

293

294The interaction of the main factors for all total tract measures of nutrient utilisation was 295significant. This was mostly due to the large responses of wheats 2 and 6 to xylanase 296addition due to their comparatively low nutrient utilisation in the absence of enzyme. 297Feed conversion ratio and measured AME significantly correlated in both wheats, 298suggesting the added benefit (r = -0.65, P = 0.023 and r = -0.49, P = 0.11, respectively; 299data not shown). This observation implies that xylanase may have greater effects in 300poorer quality samples, elevating their nutritive value and thus reducing the variability 301between samples. Nonetheless, none of the results from the chemical analysis or NIRS 302predictions suggested that these two samples may have had a poorer nutritive value than 303the others. In this regard, it is noteworthy the low correlation between the predicted 304AME and the measured AME (r = -0.16; P = 0.13) suggesting the limited capacity of 305NIR to predict animal performance (data not shown). Wheats 3, 4, 5 and 8 had higher 306nutrient utilisation in the absence of enzyme, which may be due to the presence of 307endoxylanase in the outer layer of wheat (Cleemput et al., 1997), being responsible for 308part of the degradation of AX (Dornez et al., 2006), or lesser content of xylanase 309inhibitors or both.

310

311The response of broilers to dietary intervention in general and enzyme supplementation 312in particular is usually measured using growth performance responses or ileal nutrient 313utilisation and total tract nutrient retention. These can be adequate for measuring gross 314efficiency of nutrient utilisation, but to further characterize the efficiency of nutrient 315utilisation it is important to delineate the weight gain into the composition of gain, i.e.,

316protein or fat, especially because of the differences in the efficiency with which these 317nutrients are deposited (Olukosi and Adeola, 2008). Net energy for production can be 318used as a more sensitive measure of energy utilisation by chickens receiving exogenous 319enzymes because it takes into account the efficiency of utilisation of ME for growth 320(Bhuiyan and Iji, 2015; Pirgozliev and Rose, 1999; Olukosi and Adeola, 2008; Olukosi 321et al., 2008a). Net energy for production is not only dependent on body weight but also 322on the amount of energy deposited in the carcass, which is an indication of how 323effectively the enzyme used facilitated energy utilisation. Net energy for production and 324K_{RE} were not influenced by xylanase supplementation, but wheat sample did. Wheat 325samples 2, 7 and 8 presented better indices of energy utilisation, which may be related 326to the fact that they have the lowest viscosities compared with the other wheats. Heat 327production and Kre-Protein varied depending on wheat and xylanase inclusion.

329Interestingly, xylanase supplementation of wheats 2 and 6 increased total tract AME 330retention, Nep and HP but reduced K_{RE} , Kre-CP and Kre-Fat and the efficiency of 331energy use for protein and fat accretion, as has been demonstrated previously (Bhuiyan 332and Iji, 2015; Daskiran et al., 2004; Olukosi and Adeola, 2008; Olukosi et al., 2008a). In 333the current study, the comparison between animal performance and energy utilisation 334must be considered with caution as only one bird from each replicate was selected. The 335extrapolation of the performance data derived from eight animals per replicate may thus 336have some mis-alignment with the energy utilisation results obtained from one 337individual bird.

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339The utilisation of ME was more efficient for energy deposition and less for protein and 340fat. Nevertheless the efficiency of protein accretion was almost two-fold that of fat

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341accretion, which was similarly shown by previous authors (Olukosi and Adeola, 2008; 342Olukosi et al., 2008b). The genetics and age of birds are important factors (Leeson and 343Summers, 1997; Lopez and Leeson, 2005). The higher proportion and retention of 344protein than fat is likely because the young broiler chicks were still actively growing 345and have not reached the stage at which fat deposition can overtake protein deposition 346(Bregendahl et al., 2002; Sanz et al., 2000).

347

3485. Conclusion

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350Under the current experimental conditions xylanase supplementation may compensate 351for the poorer nutritive value of some wheats, enabling more homogenous broiler chick 352performance. Unfortunately the predicted nutrient composition by NIR did not predict 353accurately animal performance, and moreover taken together the predicted nutrient and 354chemically determined contents of the wheats used in this study did not allow for 355accurate ranking of the samples prior to feeding, which may relate to the very low 356viscosity of the wheat samples employed. In this regard, the use of the xylanase as an 357insurance policy is justified.

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359Acknowledgments

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365Conflict of interests

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367All authors declare no conflict of interests.

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463Tables

| 464Table 1 Ingredient and | calculated com | position as-fed | of the ex | perimental | diets |
|---------------------------|----------------|-----------------|-----------|------------|-------|
| | | | | | |

| Item | Control | + Xylanase |
|------------------------------------|---------|------------|
| Ingredient, g/kg | | |
| Wheat - feed | 585 | 585 |
| Soybean meal 48 | 325 | 325 |
| Soy oil | 44.4 | 44.4 |
| Salt | 3.00 | 3.00 |
| Sodium bicarbonate | 1.87 | 1.87 |
| DL-methionine | 2.99 | 2.99 |
| Lysine HCl | 2.46 | 2.46 |
| Threonine | 0.77 | 0.77 |
| Limestone | 7.86 | 7.86 |
| Dicalcium phosphate | 15.5 | 15.5 |
| Vitamin premix ¹ | 4.90 | 4.90 |
| Phytase ² | + | + |
| Xylanase ³ | - | + |
| Calculated nutrient composition, % | | |
| Crude protein | 22.4 | 22.4 |
| Ca | 0.90 | 0.90 |
| Р | 0.74 | 0.74 |
| Available phosphorous | 0.45 | 0.45 |
| Fat | 5.72 | 5.72 |
| Fibre | 2.55 | 2.55 |
| Met | 0.62 | 0.62 |
| Cys | 0.38 | 0.38 |
| Met + Cys | 1.00 | 1.00 |
| Lys | 1.35 | 1.35 |
| His | 0.55 | 0.55 |
| Trp | 0.28 | 0.28 |
| Thr | 0.88 | 0.88 |
| Arg | 1.45 | 1.45 |
| Ile | 0.92 | 0.92 |
| Leu | 1.64 | 1.64 |
| Phe | 1.05 | 1.05 |
| Val | 1.00 | 1.00 |
| AME, MJ/kg | 12.8 | 12.8 |

465AME = apparent metabolisable energy.

466¹ Vitamin/mineral premix supply per kilogram of diet: vitamin A, 16,000 IU; vitamin 467D₃, 3000 IU; vitamin E, 25 IU; vitamin B₁, 3 mg; vitamin B₂, 10 mg; vitamin B₆, 3 mg; 468vitamin B₁₂, 15 μg; nicotinic acid, 60 mg; pantothenic acid, 14.7 mg; folic acid, 1.5 mg;

469biotin, 125 μg; choline chloride, 25 mg; Fe as iron sulfate, 20 mg; Cu as copper sulfate, 47010 mg; Mn as manganous oxide, 100 mg; Co as cobalt oxide, 1.0 mg; Zn as zinc oxide, 47182.222 mg; I as potassium iodide, 1 mg; Se as sodium selenite, 0.2 mg; and Mo as 472molybdenum oxide, 0.5 mg.

473² Quantum Blue 5G, AB Vista, Marlborough, UK; 5000 FTU/g.

474³ Econase XT 25P, AB Vista, Marlborough, UK; 160,000 BXU/g.

475Table 2 Analysed nutrient composition and coefficient of variation (CV) of the wheat

| Item | Wheat samples | | | | | | | | |
|---------------------|---------------|------|------|------|------|------|------|------|----|
| | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | CV |
| Gross energy, MJ/kg | 18.0 | 18.1 | 18.1 | 18.0 | 18.2 | 17.9 | 18.0 | 18.1 | <1 |
| Viscosity, cP | 10.5 | 8.50 | 12.8 | 13.0 | 11.3 | 11.2 | 7.60 | 7.80 | 21 |
| Dry matter, % | 87.2 | 87.4 | 87.8 | 87.5 | 87.1 | 87.2 | 88.6 | 87.6 | <1 |
| Fat, % | 1.49 | 1.37 | 1.48 | 1.37 | 1.26 | 1.15 | 1.24 | 1.94 | 17 |
| Nitrogen, % | 2.22 | 1.88 | 2.37 | 2.10 | 2.02 | 1.79 | 1.55 | 1.79 | 13 |
| Calcium, % | 0.03 | 0.03 | 0.03 | 0.03 | 0.03 | 0.02 | 0.02 | 0.05 | 31 |
| Phosphorous, % | 0.28 | 0.32 | 0.34 | 0.33 | 0.38 | 0.29 | 0.27 | 0.33 | 11 |
| Phytic acid, % | 0.75 | 0.77 | 0.64 | 0.72 | 0.81 | 0.92 | 0.53 | 0.53 | 19 |

| Item | Wheat samples | | | | | | | | |
|------------------------------|---------------|--------|------|------|------|------|------|------|----|
| | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | CV |
| Energy, MJ/kg and ether extr | act, % | | | | | | | | |
| GE | 16.5 | 16.5 | 16.6 | 16.6 | 16.6 | 16.4 | 16.3 | 16.4 | <1 |
| AME | 13.4 | 13.4 | 13.2 | 13.0 | 13.3 | 13.6 | 13.9 | 13.4 | 2 |
| Fat | 2.10 | 2.23 | 2.16 | 2.40 | 2.28 | 2.25 | 1.95 | 2.13 | 6 |
| Fibre | | | | | | | | | |
| NDF | 15.1 | 16.4 | 16.1 | 19.0 | 17.2 | 16.1 | 14.0 | 16.1 | 9 |
| ADF | 2.44 | 2.82 | 2.94 | 3.94 | 3.15 | 2.99 | 2.11 | 3.02 | 18 |
| Lignin | 0.72 | 0.97 | 0.89 | 1.15 | 1.06 | 1.02 | 0.95 | 0.94 | 13 |
| AX | 7.66 | 8.10 | 7.97 | 9.19 | 8.48 | 7.83 | 7.30 | 8.02 | 7 |
| Soluble AX | 0.56 | 0.61 | 0.57 | 0.62 | 0.63 | 0.59 | 0.58 | 0.57 | 4 |
| β-glucan | 1.21 | 1.66 | 1.66 | 2.36 | 1.88 | 1.63 | 1.78 | 1.90 | 18 |
| Total insoluble NSP | 10.2 | 11.4 | 11.1 | 13.2 | 12.1 | 11.1 | 10.4 | 11.3 | 8 |
| Total soluble NSP | 1.84 | 2.47 | 2.36 | 3.23 | 2.75 | 2.43 | 2.56 | 2.64 | 15 |
| Protein,% and amino acid pro | ofile, g/100 |) g CP | | | | | | | |
| CP | 13.4 | 11.44 | 13.6 | 11.7 | 11.9 | 10.6 | 8.36 | 10.2 | 15 |
| Lysine | 3.01 | 3.35 | 2.96 | 3.42 | 3.26 | 3.18 | 3.07 | 3.19 | 5 |
| Methionine | 1.56 | 1.68 | 1.60 | 1.65 | 1.69 | 1.70 | 1.78 | 1.67 | 4 |
| Leucine | 7.37 | 6.64 | 7.07 | 6.47 | 6.43 | 6.98 | 7.22 | 7.10 | 5 |
| Threonine | 3.30 | 3.42 | 3.25 | 3.42 | 3.34 | 3.41 | 3.41 | 3.42 | 2 |
| Tryptophan | 1.16 | 1.21 | 1.16 | 1.21 | 1.19 | 1.22 | 1.30 | 1.27 | 4 |
| Tyrosine | 3.21 | 3.13 | 3.20 | 3.09 | 3.12 | 3.23 | 3.28 | 3.23 | 2 |
| Valine | 4.75 | 4.91 | 4.76 | 4.96 | 4.86 | 4.90 | 4.99 | 4.95 | 2 |
| Phenylalanine | 4.43 | 4.39 | 4.55 | 4.53 | 4.47 | 4.44 | 4.53 | 4.50 | 1 |
| Histidine | 2.57 | 2.62 | 2.59 | 2.61 | 2.63 | 2.62 | 2.60 | 2.58 | <1 |
| Isoleucine | 3.48 | 3.43 | 3.53 | 3.45 | 3.46 | 3.47 | 3.46 | 3.46 | <1 |
| Arginine | 5.18 | 5.43 | 5.09 | 5.52 | 5.33 | 5.07 | 4.54 | 4.99 | 6 |
| Alanine | 4.28 | 3.76 | 4.11 | 3.89 | 3.67 | 4.04 | 4.33 | 4.22 | 6 |
| Asparagine | 6.06 | 6.22 | 5.79 | 6.27 | 5.98 | 6.04 | 5.59 | 6.00 | 4 |
| Cysteine | 2.19 | 2.34 | 2.20 | 2.27 | 2.34 | 2.33 | 2.40 | 2.32 | 3 |
| Glutamine | 24.9 | 23.5 | 26.0 | 23.2 | 24.7 | 24.1 | 24.0 | 23.4 | 4 |
| Glycine | 3.99 | 4.27 | 3.98 | 4.19 | 4.23 | 4.22 | 4.19 | 4.15 | 3 |
| Proline | 9.23 | 8.99 | 9.78 | 9.23 | 9.39 | 9.28 | 9.91 | 9.40 | 3 |
| Serine | 4.88 | 4.83 | 4.83 | 4.70 | 4.79 | 4.84 | 4.81 | 4.78 | 1 |

480coefficients of variation (CV) of the wheat samples

| 481GE= gross energy; AME = apparent metabolisable energy; NDF = neutral detergent |
|---|
| 482fibre; ADF = acid detergent fibre; AX = arabinoxylan; NSP = non-starch |
| 483polysaccharides; CP = crude protein. |
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| Item | Weight gain, g/bird | Feed intake, g/bird | Feed conversion ratio, g/g | Jejunal viscosity, cP |
|-----------------|---------------------------|------------------------|-------------------------------------|-----------------------------|
| Wheat effect | | | | |
| 1 | 824 | 1105 | 1.341 | 2.81 |
| 2 | 816 | 1103 | 1.353 | 3.13 |
| 3 | 820 | 1087 | 1.329 | 2.89 |
| 4 | 817 | 1097 | 1.345 | 3.01 |
| 5 | 781 | 1087 | 1.394 | 2.77 |
| 6 | 791 | 1079 | 1.369 | 2.94 |
| 7 | 787 | 1049 | 1.341 | 2.40 |
| 8 | 791 | 1065 | 1.349 | 2.67 |
| SEM | 16 | 16 | 0.019 | 0.06 |
| Xylanase effect | | | | |
| 0 BXU/kg | 793^{b} | 1088 | 1.373ª | 3.3 2ª |
| 16,000 BXU/kg | 813 ^a | 1079 | 1.331 ^b | 2.34^{b} |
| SEM | 8 | 8 | 0.010 | 0.03 |
| <i>P</i> -value | | | | |
| Wheat | 0.465 | 0.192 | 0.367 | 0.380 |
| Xylanase | 0.072 | 0.496 | 0.003 | < 0.001 |
| Interaction | 0.951 | 0.950 | 0.894 | 0.845 |

507Table 4 Animal performance and jejunal digesta viscosity¹

 $508^{a,b}$ Means in the same column with different letters differ at P < 0.05.

509¹ Mean values for six replicate cages with eight broilers per replicate cage.

510Table 5 Correlation of feed conversion ratio (FCR) with the analysed chemical 511composition and the predicted values by near-infrared spectroscopy (NIRS) of wheat in 512diets supplemented with or without xylanase

| | Pearson's Corr | Pearson's Correlation | | | | |
|----------------------------|-----------------|-----------------------|--|--|--|--|
| | coefficients wi | vith FCR | | | | |
| Itere | Without | Mith mileness | | | | |
| Item | xylanase | With xylanase | | | | |
| Analysed composition | | | | | | |
| GE | -0.27 | 0.38 | | | | |
| Fat | 0.14 | -0.26 | | | | |
| Nitrogen | -0.47 | 0.07 | | | | |
| Calcium | -0.43 | 0.36 | | | | |
| Phosphorous | -0.35 | 0.70* | | | | |
| NIRS predicted composition | | | | | | |
| СР | 0.06 | 0.07 | | | | |
| Fat | -0.27 | 0.68* | | | | |
| GE | -0.09 | 0.49 | | | | |
| AME | 0.53 | -0.45 | | | | |
| ADF | -0.34 | 0.63 | | | | |
| NDF | -0.31 | 0.69* | | | | |
| Total AX | -0.36 | 0.73* | | | | |
| Soluble AX | 0.25 | 0.85* | | | | |
| β-glucan | -0.34 | 0.55 | | | | |
| Lignin | 0.07 | 0.62 | | | | |
| Total insoluble NSP | -0.26 | 0.74* | | | | |
| Total soluble NSP | -0.23 | 0.60 | | | | |

513GE = gross energy; NIRS = near-infrared spectroscopy; CP = crude protein; AME =

514apparent metabolisable energy; ADF = acid detergent fibre; NDF = neutral detergent

515fibre; AX = arabinoxylan; NSP = non-starch polysaccharides.

516 *P < 0.05

| Item | Dry matter, % | Nitrogen, % | Energy, % | IDE, MJ/kg |
|-----------------|--------------------|-------------|----------------------|----------------------|
| Wheat effect | | | | |
| 1 | 68.0 ^{bc} | 78.0 | 70.8^{bc} | 13.2 ^b |
| 2 | 66.9 ^{bc} | 74.8 | 70.0 ^c | 13.1 ^b |
| 3 | 65.2 ^c | 74.4 | 69.5° | 13.2 ^b |
| 4 | 68.8 ^{bc} | 78.2 | 72.0 ^{abc} | 13.7^{ab} |
| 5 | 66.9 ^{bc} | 75.9 | 69.6 ^c | 13.0 ^b |
| 6 | 70.2 ^{ab} | 78.1 | 73.2 ^{abc} | 13.8 ^{ab} |
| 7 | 70.4^{ab} | 79.4 | 73.9 ^{ab} | 14.0 ^a |
| 8 | 73.0ª | 79.7 | 76.0 ^a | 14.4 ^a |
| SEM | 1.47 | 1.42 | 1.47 | 0.28 |
| Xylanase effect | | | | |
| 0 BXU/kg | 67.8 | 76.9 | 70.9 ^b | 13.35 ^b |
| 16,000 BXU/kg | 69.5 | 77.8 | 72.9 ^a | 13.77 ª |
| SEM | 0.74 | 0.71 | 0.74 | 0.14 |
| <i>P</i> -value | | | | |
| Wheat | 0.012 | 0.062 | 0.019 | 0.004 |
| Xylanase | 0.111 | 0.205 | 0.057 | 0.039 |
| Interaction | 0.550 | 0.104 | 0.571 | 0.577 |

517Table 6 Ileal nutrient utilisation of nutrients¹

518IDE = ileal utilization of energy.

 $519^{\text{a-c}}$ Means in the same column with different letters differ at *P* < 0.05.

520¹Mean values for six replicate cages with eight broilers per replicate cage.

521

522

| Wheat & X | ylanase effect | Dry matter, % | Nitrogen, % | AME, MJ/kg | AMEn, MJ/kg |
|-------------|---------------------|----------------------|---------------------|----------------------|----------------------|
| Wheat | Xylanase, BXU/kg | | | | |
| 1 | 0 | 69.4^{cde} | 62.9 ^{de} | 13.5^{ef} | 13.0 ^e |
| 1 | 16,000 | 71.2^{bc} | 60.7^{ab} | 14.1^{cde} | 13.5 ^{cd} |
| 2 | 0 | 65.4 ^g | 57.9^{f} | 12.9 ^h | 12.4 ^g |
| 2 | 16,000 | 73.4ª | 65.7ª | 14.5^{ab} | 14.0^{ab} |
| 3 | 0 | 68.6 ^{de} | 58.1 ^{cd} | 13.8^{fg} | 13.2 ^e |
| 3 | 16,000 | 68.4 ^e | 57.4 ^{cd} | 13.8^{fg} | 13.2 ^{ef} |
| 4 | 0 | 71.5^{abc} | 63.3 ^{abc} | 14.2 ^{abc} | 13.8 ^{abc} |
| 4 | 16,000 | 70.6 ^{cd} | 62.7 ^{cd} | 14.1 ^{cd} | 13.6 ^{cd} |
| 5 | 0 | 69.6 ^{cde} | 64.6 ^e | 13.5^{def} | 13.0 ^{de} |
| 5 | 16,000 | 66.1^{fg} | 57.7 ^{ef} | 13.0 ^{gh} | 12.4^{fg} |
| 6 | 0 | 65.2 ^g | 55.1^{ef} | 13.0 ^h | 12.4 ^g |
| 6 | 16,000 | 73.1^{ab} | 66.9 ^{abc} | 14.4^{ab} | 13.9 ^{ab} |
| 7 | 0 | 68.0 ^{ef} | 62.0 ^{ef} | 13.6 ^{fg} | 13.1 ^{ef} |
| 7 | 16,000 | 69.9 ^{cde} | 65.3° | $14.1^{	ext{ef}}$ | 13.7 ^e |
| 8 | 0 | 72.8 ^{ab} | 65.8 ^{abc} | 14.5 ^ª | 14.0^{a} |
| 8 | 16,000 | 71.4^{abc} | 64.1 ^{abc} | 14.2 ^{bc} | 13.7^{bc} |
| Pooled SEM | N | 0.73 | 0.79 | 0.12 | 0.12 |
| P-value | | | | | |
| Wheat | | < 0.001 | < 0.001 | < 0.001 | < 0.001 |
| Xylanase | | < 0.001 | < 0.001 | < 0.001 | < 0.001 |
| Interaction | | < 0.001 | < 0.001 | < 0.001 | < 0.001 |

524Table 7 Total tract retention of nutrients¹

525AME = apparent metabolisable energy; AMEn = AME corrected for nitrogen ;

526^{a-e} Different letters mean significant differences between treatments, highlighting the

527 statistical interaction between main factors wheat x xylanase (P < 0.05).

528 ¹Mean values for six replicate cages with three broilers per replicate cage.

| Te | Energy utilisa | Energy utilisation, MJ/kg | | retained, MJ/kg | Efficienc prote | Efficiencies of energy use for energy, protein and fat retention accretion | | |
|-----------------|-----------------------|---------------------------|-----------------------------|---------------------|--------------------|--|-----------------------|--|
| Item — | Nep ² | HP ³ | Protein- ER ⁴ | Fat-ER⁵ | ${\rm K_{RE}}^6$ | Kre-Protein ⁷ | Kre-Fat ⁸ | |
| Wheat effect | | | | | | | | |
| 1 | 5.59^{abc} | 6.84 ^{bc} | 3.92 ^{ab} | 2.20 ^{bc} | 0.45 ^{bc} | 0.275 ^{ab} | 0.154^{abcd} | |
| 2 | 5.94 ^a | 6.35 ^d | 3.89 ^{ab} | 2.48 ^a | 0.48^{a} | 0.275^{ab} | 0.175ª | |
| 3 | 5.53 ^{abc} | 6.54^{cd} | 3.94 ^a | 2.04 ^c | 0.46^{ab} | 0.289ª | 0.149^{bcd} | |
| 4 | 5.53° | 7.24 ^a | 3.95 ª | 2.04 ^c | 0.43 ^c | 0.269^{bc} | 0.139^{d} | |
| 5 | 5.32 ^c | 6.74 ^{bc} | 3.96 ^a | 2.05 ^c | 0.44^{bc} | 0.277^{ab} | 0.143 ^{cd} | |
| 6 | 5.64 ^{abc} | 6.60^{bcd} | 3.89 ^{ab} | 2.24 ^{abc} | 0.46^{ab} | 0.275^{ab} | 0.158^{abcd} | |
| 7 | 5.72 ^{bc} | 6.44 ^{bcd} | 3.83 ^b | 2.25 ^{abc} | $0.47^{\rm b}$ | 0.278^{ab} | 0.163 ^{ab} | |
| 8 | 5.77 ^{ab} | 7.22 ^{ab} | 3.84 ^b | 2.41 ^{ab} | 0.44^{bc} | 0.258 ^c | 0.162 ^{abc} | |
| SEM | 0.135 | 0.155 | 0.031 | 0.098 | 0.011 | 0.0050 | 0.0078 | |
| Xylanase effect | | | | | | | | |
| 0 BXU/kg | 5.54 | 6.68 | 3.92 | 2.18 | 0.454 | 0.275 | 0.153 | |
| 16,000 BXU/kg | 5.72 | 6.81 | 3.89 | 2.24 | 0.457 | 0.274 | 0.158 | |
| SEM | 0.068 | 0.077 | 0.016 | 0.049 | 0.005 | 0.0025 | 0.0039 | |
| <i>P</i> -value | | | | | | | | |
| Wheat | 0.029 | 0.001 | 0.032 | 0.008 | 0.012 | 0.017 | 0.029 | |
| Xylanase | 0.950 | 0.060 | 0.135 | 0.373 | 0.327 | 0.869 | 0.354 | |

534Table 8 Energy utilisation, energy retained and efficiencies of energy use¹

| Interaction | 0.198 | 0.018 | 0.429 | 0.922 | 0.284 | 0.006 | 0.984 | |
|---|--|---------------|-------|-------|-------|-------|-------|--|
| 535 ¹ Mean values for six replicate cages with one broiler per replicate cage. | | | | | | | | |
| 536 ² NEp - net energy for produc | tion. | | | | | | | |
| 537 ³ HP - heat production. | | | | | | | | |
| 538 ⁴ Protein-ER - energy retained as protein. | | | | | | | | |
| 539^{5} Fat ER - energy retained as f | fat. | | | | | | | |
| $540^6\ K_{\text{\tiny RE}}$ - efficiency of energy us | se for production. | | | | | | | |
| 541 ⁷ Kre-Protein - efficiency of e | nergy use for protei | in accretion. | | | | | | |
| 542 ⁸ Kre-Fat - efficiency of energ | 542 ⁸ Kre-Fat - efficiency of energy use for fat accretion. | | | | | | | |
| 543 ^{a-d} Means in the same column with different letters differ at $P < 0.05$. | | | | | | | | |
| | | | | | | | | |
| | | | | | | | | |