EVALUATION OF MONO-ENZYME (NUTRASE XYLA) AND MULTI-ENZYMES (NATUZYME AND BIOZYME) SUPPLEMENTATION ON THE PERFORMANCE OF LAYING BIRDS

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ABSTRACT
To assess the efficacy of enzyme type, 480 laying birds (ISA Brown; 43 weeks old) were allocated to four treatments (6 replicates, 20birds each) in a completely randomized design. Hens were fed a corn-groundnut cake-soybean meal diet over a 6-wk period. The control diet (2,650 kcal/kg ME, 18.5% CP, 0.94% lysine and 0.46% methionine), had a 3.5% greater nutrient density than the enzyme supplemented diets. Enzymes supplemented, following manufacturers instruction, were Nutrase Xyla (endo-xylanase, 100g/ton), Natuzyme (a multi-enzyme, 350g/ton) and Biozyme (a multi-enzyme, 350g/ton). Enzyme supplementation did not influence egg weight, feed intake, feed:egg (g/egg) and metabolizable energy intake (P > 0.05). Egg production (%) and egg mass were higher with multi-enzyme (P<0.05). however, feed:egg (g/g) was poorer on Biozyme (P < 0.05). Feed cost, per egg was lowest on Biozyme, while per kg egg was lowest with Nutrase-xyla. Natuzyme and Biozyme yielded a higher percentage of extra-large/jumbo eggs than the control group (P<0.05). Enzyme supplementation improved egg production and size, with a greater effect observed with multi-enzymes. However, enzyme contribution to feed cost will determine choice of enzyme.
Keywords: poultry, enzyme, egg production, egg size, feed cost

INTRODUCTION

Over time, enzymes have become increasingly popular as feed additives (Cowieson and Bedford, 2009). The sudden increase in the use of enzymes can be attributed to an expanding need to increase efficiency of feed ingredients to decrease costs (O'Neill *et al*., 2012). With rising capital investment in feed manufacturing, enzyme supplementation may enable producers use feed ingredients that would normally be poor in apparent metabolizable energy (ME). These enzymes hydrolyze and neutralize the negative effects produced by viscous compounds in these cereals, such as water-soluble non-starch polysaccharides (NSPs). Studies have shown beneficial effects of supplemental enzymes on feed intake, growth performance and egg production in poultry (Olukosi *et al*., 2015). However, evidence supporting benefits of enzyme supplementation in corn-based diets are inconsistent. According to Ravindran (2013), poultry producers supplementing enzymes face a dilemma of variable and unpredictable responses by birds. However, continuing and intensive research and development of feed enzymes has the potential to increase their use in regions where corn is fed. Therefore, products which contain a specific blend of enzymes may be able to provide multiple benefits, as compared with mono-component enzymes such as xylanases, the use of which may offer little or no benefit in corn-soya diets. In the tropics, like Nigeria, indiscriminate use of enzymes without consideration for feed matrix and substrate specificity still exists. Thus the objective of this study was to ascertain the efficacy of some of these enzymes as professed by distributors of such products.

MATERIALS AND METHODS

The study was carried out on a commercial farm (Dedora Farms Nigeria Limited), located in Ogun state, south-western, Nigeria. Four hundred and eighty laying birds (ISA Brown strain, 43-weeks old) were randomly allocated to four treatments (6 repliactes; 20birds each) in a completely randomized design. Hens were given access to 120g of feed/day, with water provided constantly via nipple drinkers. Experimental diets (iso-caloric and iso-nitrogenous, excluding the control diet; Table 1) were offered for a 6-wk period, with one week allowed for adjustment. The control diet was formulated to a nutrient density 3.5% greater than the enzyme supplemented diets. Three different enzymes with differing activities were utilised; Diet 2 was supplemented with Nutrase-Xyla (Nutrex, Hoogbuul 24, Olen 2250, Belgium), an endo-xylanase produced from *Bacillus subtilis*, Diet 3 was supplemented with Natuzyme (Bioproton Pty. Ltd. QLD, Acacia Ridge, Australia), a multi-enzyme, and Diet 4

Biozyme (Bios AgriCorp Ltd., Life Science Centre, Keilaranta 16, 02150 Espoo, Finland), a multi enzyme. Feed intake (FI), estimated metabolizable energy intake (MEI; kcal/hen /day), and egg production (EP) were recorded on a daily basis. Feed conversion (FC) was calculated as feed:egg (g/g), and also as feed:egg (g/egg). Eggs were weighed daily for egg weight (EW), while egg mass (EM) was calculated as a factor of EW and EP. Feed cost estimates were determined as cost per egg and cost per kg of egg. At the final week of data collection, eggs were collected and individually weighed, sorted and graded using both the United States and the European Union egg size grades (Table 2). Data on egg production were subjected to analysis of variance using SPSS version 20, and significance separated using Tukey's HSD ($P < 0.05$). Egg grade data was cross tabulated and subjected to Pearson's correlation, while a z-test (at α .05) was used to compare egg size proportions.

Table 1: Composition of experimental diets

Ingredients, %	Diet 1	Diet 2[1]	Diet 3[2]	Diet 4[3]
Maize	51.80	48.10	48.10	48.10
Soybean meal	9.30	7.20	7.20	7.20
Groundnut cake	18.50	18.00	18.00	18.00
Wheat Offal	6.22	12.39	12.36	12.36
Limestone	9.95	10.20	10.20	10.20
Bone meal	2.10	1.90	1.90	1.90
Soya oil	0.85	0.85	0.85	0.85
Salt	0.31	0.31	0.31	0.31
Lysine HCl	0.25	0.25	0.25	0.25
Methionine DL	0.18	0.18	0.18	0.18
Methiorep[4]	0.05	0.05	0.05	0.05
Biosure[5]	0.10	0.10	0.10	0.10
Toxibond[6]	0.10	0.10	0.10	0.10
Choline Chloride	0.042	0.042	0.042	0.042
Min/Vit. Premix	0.30	0.30	0.30	0.30
Enzyme	-	0.01	0.035	0.035
Calculated nutrient content, %				
Metabolizable energy, kcal/kg	2651	2560	2560	2560
Crude protein	18.51	17.97	17.96	17.96
Crude fibre	3.32	3.74	3.74	3.74
Lysine	0.94	0.91	0.91	0.91
Methionine	0.46	0.44	0.44	0.44
Calcium	4.29	4.30	4.30	4.30
Available phosphorus	0.41	0.42	0.42	0.42
Linoleic acid	1.67	1.61	1.61	1.61

[1]Nutrase xyla (units per gram): bacterial (Bacillus subtilis) endo-1,4-beta-xylanase (IUB: EC 3.2.1.8; 9,000).
[2]Natuzyme (units per gram): cellulase ≥6,000, xylanase≥10,000, α-amylase ≥700, β-glucanase ≥700, phytase≥500, pectinase ≥70, protease ≥3,000, lipase ≥30. [3]Biozyme (units): α-amylase 800, protease 8,000, xylanase 500, β-glucanase 200, cellulase 7,000, lipase 80, fructooligosaccharides C.S.P. 1g. [4]Herbal derived methionine (65%). [5]Ammonium propionate, propionic acid, formic acid and methylene oxide. [6]Thermally Hydrated and Activated Sodium Calcium Aluminosilicate (HSCAS)

Table 2. Egg size grades

US Grade		EU grade	
Size	Minimum	Size	Range
Jumbo	70.9g	Extra large	≥ 73g
Extra large	63.8g	Large	≥63 and <73g
Large	56.8g	Medium	≥53 and <63g
Medium	49.6g	Small	<53g
Small	42.5g		
Peewee	35.4g		

RESULTS AND DISCUSSION

In the current study, it was observed that HP and EM ($P = 0.039$) were significantly higher with the enzyme cocktails (Natuzyme and Biozyme) supplementation compared with the control ($P<0.05$; Table 3). Xylanase supplementation did not influence HP and EM. However, feed conversion on whole egg basis tended to be better with Biozyme supplementation (P = 0.09). Surprisingly, feed:egg (g/g) was significantly poorer with Biozyme supplementation than other enzyme treatments as well as control group (P = 0.001). Feed conversion (g:g) for the Nutrase xyla and Natuzyme groups however did not differ from the control group.

Table 3: Effect of mono-component enzyme (Nutrase Xyla) and multi-enzyme (Natuzyme and Biozyme) supplementation on performance and feed cost of laying birds

	Control	NX	NZ	BZ	SEM	P value
Hen-day production (%)	72.5^{a}	77.15^{ab}	78.5^{b}	80.8^{b}	0.85	.026
Egg Weight (g)	64.04	63.78	64.32	63.09	0.15	.171
Egg Mass (g/d/hen)	46.45^{a}	49.17^{ab}	50.44^{b}	50.99^{b}	0.53	.039
Feed Intake (g/d/hen)	106.5	106.5	107.3	107.9	0.40	.865
FCR (g /egg)	147.7	139.7	137.7	135.3	1.66	.092
FCR (g /g)	1.66^{a}	1.68^{a}	1.68^{a}	1.73^{b}	0.005	.001
MEI (kcal/d/hen)	282.2	272.6	274.6	276.2	1.80	.298
Feed Cost	387.97	369.63	371.90	373.28	-	-
Feed Cost/Egg	57.29	51.64	51.21	50.52	-	-
Feed Cost/ kg Egg	644.03	620.98	624.79	645.77	-	-

^{a,b} means in a row with different superscripts are significantly different (p < 0.05)
MEI: metabolizable energy intake, FCR: feed conversion ratio, NX: nutrase xyla, NZ: natuzyme, BZ: biozyme

Xylanase is a common non-starch polysaccharide enzyme added to poultry diets that hydrolyzes bonds between sugars in xylan molecules, including arabinoxylans (Bigge *et al*., 2018). However, the effect can vary depending on the chosen ingredient (Cowieson and Bedford, 2009). Increased egg production (EP) has been reported after using xylanase in wheat containing diets (Mirzae *et al*., 2012), and corn–soybean meal-dried distiller's grain diet (Bobeck *et al*., 2014). Contrarily, Bigge *et al*. (2018) reported that xylanase supplementation on a corn-soybean meal- dried distiller's grains and soluble (DDGS) diet did not influence EP. In fact, EW and EM were significantly lower in the negative control (NC) + xylanase compared with positive (PC) and NC. Hann-Didde and Purdum (2014) examining the effect of a xylanase/amylase/protease complex in laying hen diets containing DGGS, reported that birds fed diets with an enzyme complex and low ME tended to consume more feed than their counterparts. This however did not increase production; as enzyme complexes have been shown to recoup energy within the diet (Novak *et al*., 2007). Recently, an enzyme cocktail added to corn–soybean meal diets of broilers notably improved FCR and enhanced production efficiency (Al-qhtani *et al*., 2022). This influence could be linked to hydrolysis of the different components of NSPs found in the diet. Unlike individual enzymes which target one substrate, multi-enzymes have a greater effect due to several enzymatic activities that target multiple components of feed and contribute to improved feed efficiency (Sureshkumar *et al*. 2023).

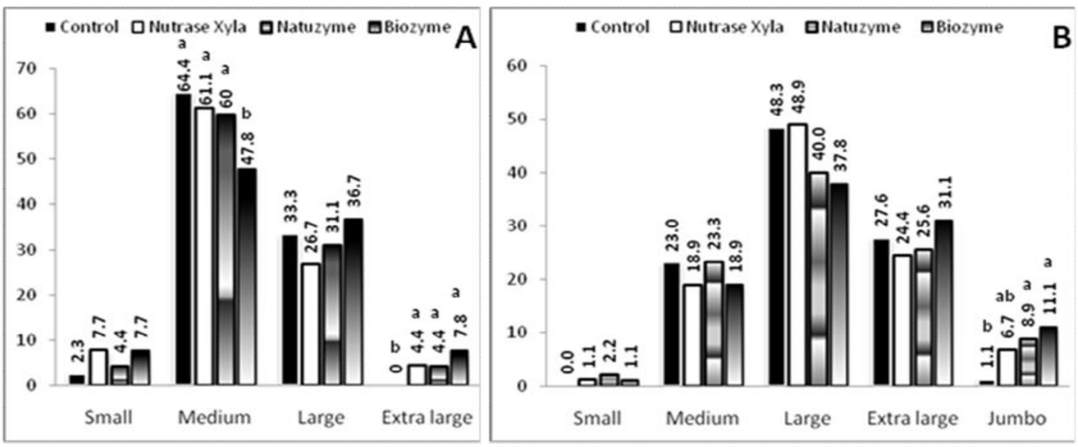

Figure 1. Percentage (%) of egg size from laying birds fed a mono-component or multi-enzymes in diets: (**A**) US grade, (**B**) EU grade system.
a, b. Alphabets denote column proportions which differ significantly at α .05.

Feed cost estimation revealed that on whole egg basis, the higher density control group cost the most, while the Biozyme group cost the least, likely a result of the higher egg production, and apparently lower FC per egg. Xylanase supplementation however produced the least feed cost/kg of egg, while feed cost/kg egg was highest with Biozyme. Using the EU grading system, Biozyme supplementation resulted in a lower percentage of medium sized eggs (-17%), with a corresponding increase in extra-large eggs (+7%) compared with the control ($P < 0.05$). In fact, enzyme supplementation produced a higher proportion of extra-large eggs ($P < 0.05$). On the US grading system, egg size proportions did not differ between the treatments, except for Jumbo size, which was greater for the Natuzyme and Biozyme supplemented diets ($P < 0.05$). Notably, both Natuzyme and Biozyme have a protease enzyme included in the cocktail (> 3000 and 8000 u/g respectively). The addition of a protease, such as noted in enzyme complexes, improves protein and nitrogen retention (Hahn-Didde and Purdum, 2014), which could influence the size of the eggs positively.

CONCLUSION

In **CONCLUSION**, while enzyme supplementations significantly improved egg production indices, these were varied among the treatments. Multi-enzymes supplementation showed marked advantage over the mono-component enzyme, with regards to quantity and size of eggs produced, probably due to the presence of protease in the complex. However, cost-benefit could be a major factor in the choice of enzyme, since xylanase was as cost effective as the multi-enzymes.

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