Nigerian J. Anim. Sci. 2018, 20 (2): 85-91

Effect of Two Direct-Fed Microbials and Mycotoxin Binder on Performance and Blood Parameters of Weaned Pigs

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***Target Audience**: Pig farmers, Feed millers, Animal Scientists, Veterinarians*

Abstract

The study was conducted to evaluate the effect of multi-strain direct-fed microbials, mycotoxin binder and yeast culture on the growth performance and blood parameters of weaned pigs. Sixty four (64) crossbred (Landrace × Large White) weaned pigs with an average initial weight of 5.9 ±0.3 kg (± SE) were assigned to four dietary treatments in a completely randomised design. Each treatment had four replicates of four pigs each. Pigs on T1 were given control diet without any additive, while pigs on T2, T3 and T4 were fed the same diet as in T1, to which multi-strain direct-fed microbials, mycotoxin binder (bentonite) and yeast culture were added respectively and the study lasted for 56 days. There were significant (P<0.05) differences in daily feed intake and daily weight gains, thereby resulting in variation in the final body weights across the groups. Pigs on T2 had the highest daily feed intake (kg) and cost of feed consumed per day (₦) but had the least feed conversion ratio and feed cost per kilogram weight gain (₦/kg). The highest final body weight (20.90kg) was observed in pigs on treatment 2, while the least final body weight of 16.95kg was recorded with for pigs on treatment 4. The haematological parameters were within the normal range for healthy pigs and there was a significant effect of the feed additives on the serum biochemical parameters such as AST, ALT, ALP, glucose and albumin. It could be concluded that multi-strain direct-fed microbial promoted growth over and above the mycotoxin binder and yeast culture in weaned pigs and none of the feed additives had any deleterious effect on the health status of the weaned pigs.

***Keywords**: Direct fed microbials, bentonite, feed additives, pigs, blood.*

Description of Problem

Piglet mortality after weaning has been one of the major challenges facing the pig industry. Some of the possible causes, arising from an upset in the balance of the gastrointestinal flora due to abrupt nutritional and environmental changes, and other stress factors associated with weaning, that piglets experience after weaning, has been reported as diarrhoea and enteritis (1). The incidence of diarrhoea after weaning of piglets has resulted in increased mortality rate, reduction in body weight gains and increased feed conversion ratio, hence, reduction in profitability. Therefore, there is need for effective and safe alternative therapies for managing post-weaning diarrhoea in piglets raised under antibiotic-free feeding regimen. Some of the additives used to combat post-weaning diarrhoea in piglets include acidifiers, high levels of zinc and copper salts, probiotics, prebiotics, herbs and spices (2).

Some of the benefits of feeding piglets with additives include higher daily weight gains, better feed conversion, increased feed intake by animals as a result of improved palatability and improvements in health and welfare status, as reflected in the lower frequency of diarrhoea and reduced mortality of piglets (3). Direct-fed microbials (probiotics) are known to reduce the incidence of diarrhoea and its duration, hence, decrease in mortality rate of piglets (4). Direct-fed microbial can be of single-strain or multi-strain, however, multi-strain direct-fed microbial have been found to be more effective than single-strain direct-fed microbial and this could be due to multiple effect of each strains on the host (5). There has been an increased interest in the use of probiotics as binders to reduce the toxic effect of mycotoxins, and lactic acid bacteria and yeasts are two important microbes among many microbes that have been reported to show potentials for the detoxification of mycotoxins (6).

Similarly, mycotoxin binders (mainly clay minerals e.g. kaolinite) are known to adsorb aflatoxins, plant metabolites, heavy metals, toxins, pathogens and enterotoxins, and are used to control diarrhoea and digestive disorders in humans, but the dose of clay minerals for counteracting the severe effect of post-weaning diarrhoea is inconclusive in literature (7).

This study therefore, compared the effect of single-strain direct-fed microbial (yeast culture), multi-strain direct-fed microbial and mycotoxins binder (bentonite) on growth performance and blood profile of weaned pigs.

Materials and Methods

The study was carried out at the AK Research Farm situated at Eleyele, Ibadan, Oyo State. Sixty four crossbred (Large White × Landrace) weaned pigs with an average initial weight of 5.9 ±0.3 kg (± SE) were assigned to four dietary treatments in a completely randomized design in an experiment. The pigs were allotted to four dietary treatments (Table 1) of sixteen weaned pigs per treatment. Each treatment had four replicates of four pigs per replicate. Pigs on T1 were given the control diet, without any additive, while pigs on T2, T3 and T4 were fed the same diet as in T1, but to which multi-strain direct-fed microbials (DFM), mycotoxin binder (MB) and yeast culture (YC) were added respectively. Experimental diets were formulated using the nutrient requirement recommendations for weaned pigs of National Research Council (8). Multi-strain direct fed microbials was supplemented at 1.5 ml/kg diet, while the mycotoxin binder (bentonite) and yeast culture were supplemented at 1.5g/kg diet respectively. The multi-strain DFM contained 99.9% water, 1×10^{8} CFU/g *Lactobacillus sp*, 4×10^{12} CFU/g *Bacillus sp* and 11×10^{5} CFU/g *Saccharomyces cerevisiae*, while the yeast-based DFM contained yeast culture and dried brewer's yeast. The mycotoxin binder was an alluminosilicate (activated clay) that contained mainly bentonite. The animals were allowed *ad libitum* access to the feed and water on the concrete-floored pens where they were kept throughout the 56-day duration of the trial and the performance was monitored.

Table1: Gross composition of experimental diets

	Treatments[1]			
Ingredients %	T1 (Control)	T2 (DFM)	T3 (MB)	T4 (YC)
Fish meal	2.5	2.5	2.5	2.5
Groundnut cake	20	20	20	20
Palm kernel cake	9	9	9	9
Maize	50	50	50	50
Wheat bran	2.5	2.5	2.5	2.5
Palm oil	1.5	1.5	1.5	1.5
Limestone	1.25	1.25	1.25	1.25
Di-calcium phosphate	2	2	2	2
Salt	0.5	0.5	0.5	0.5
Methionine	0.2	0.2	0.2	0.2
Lysine	0.30	0.30	0.30	0.30
Vit-Min premix[2]	0.25	0.25	0.25	0.25
Additives	-	150ml	150g	150g
Total	**100**	**100**	**100**	**100**
Calculated analysis				
Metabolizable energy	2881.91	2881.91	2881.91	2881.91
Crude protein (%)	19.44	19.44	19.44	19.44
Crude fibre (%)	4.14	4.14	4.14	4.14

[1]DFM – Multi-strain direct fed microbial, MB- mycotoxin binder and YC-yeast culture
[2]Grower premix supplied the following per kg diet: vitamin A 10,000,000 IU; vitamin D 32,000,000 IU; vitamin E 8,000 IU; vitamin K 2,000 mg; vitamin B1 2,000 mg; vitamin B2 5,500 mg; vitamin B6 1,200 mg; vitamin B12 12 mg; biotin 30 mg; folic acid 600 mg; niacin 10,000 mg; pantothenic acid 7,000 mg; Choline chloride 500,000 mg; vitamin C 10,000 mg; iron 60,000 mg; Mn 80,000 mg; Cu 800 mg; Zn 50,000 mg; iodine 2,000 mg; cobalt 450 mg; selenium 100 mg; Mg 100,000 mg; anti-oxidant 6,000 mg.

At the end of the experiment, 10ml of blood sample was taken per animal from 8 animals per treatment (four male and four female) through the jugular vena cava. Blood sample from each animal was stored in 2 separate sample bottles, with one of the bottles containing ethylene diamine tetraacetic acid (EDTA). The blood samples stored in EDTA bottles were used for haematological analysis while the serum collected from sample bottle without EDTA after clotting was used in the biochemical analysis of the blood. Data obtained from the study were subjected to one-way analysis of variance and significant means were separated using Duncan multiple range test (9) of Statistical Analysis System (SAS) software for Windows (2013).

Results and Discussion

The growth performance of the weaned pigs fed diets supplemented with different feed additives is shown in Table 2. There were significant ($P<0.05$) differences in final body weight, body weight gain, daily weight gain and daily feed intake. The highest final body weight (20.90kg) was recorded with pigs fed diet supplemented with multi-strain direct fed microbials (T2) and the least final body weight (16.95kg) was recorded with pigs fed diet supplemented with the yeast culture (T4). The superior performance of pigs fed on diet supplemented with the multi-strain DFM (T2) over all other treatments may be due to improvement in the gut health and increased population of beneficial microbes at the exclusion of pathogenic strains in the gut and or broader enzymatic activities of multi-strain direct fed microbial on the weaned pig, which has been reported (10; 11; 12) to be greater at early stage of life for improved utilization of nutrients. The control diet and the diet supplemented with mycotoxin binder were comparable (P>0.05) in term of final body weight, body weight gain, daily weight gain and average daily feed intake. The supplementation of yeast culture in T4 significantly (P<0.05) decreased final body weight, daily weight gain and daily feed intake in weaned pigs. This is contrary to reports in literature in which yeast culture improved daily weight gain and feed conversion ratio in broilers (13) and daily feed intake and daily weight gain in nursery pigs (14). There was no significant effect of the additives on the feed conversion ratio across the dietary treatments.

Table 2: Growth performance of weaned pigs fed experimental diets

Parameters	T1 (Control)	T2 (DFM)	T3 (MB)	T4 (YC)	±SEM
Initial weight (kg)	5.61	6.08	6.20	5.86	0.19
Final weight (kg)	18.50^{ab}	20.90^{a}	19.46^{ab}	16.95^{b}	0.51
Body weight gain (kg)	12.89^{ab}	14.82^{a}	13.26^{ab}	11.09^{b}	0.42
Daily weight gain (g)	230.18^{ab}	264.64^{a}	236.79^{ab}	198.04^{b}	6.64
Average daily feed intake (g)	725.49^{ab}	795.24^{a}	794.66^{a}	698.97^{b}	13.48
Feed conversion ratio	3.15	3.01	3.36	3.53	0.12

DFM – Multi-strain direct fed microbial, MB- mycotoxin binder and YC-yeast culture

The economic analysis of weaned pigs fed diets supplemented with the three different feed additives is shown in Table 3. The values used for calculation were based on the prevailing market price of the ingredients at the time of purchase and sale of pigs per kg live weight. Cost of feed (₦/kg), total feed intake (kg), average daily feed intake (kg), total cost of feeding (₦), average cost of feed per day (₦), feed/gain ratio and feed cost/kg weight gain (₦/Kg) were considered. It is of interest to know which of the treatment is economically recommended at this stage of the pig's growth, given the cost of feeding. There was a significant (P<0.05) difference among the treatment means of the feed cost (₦/kg), total cost of feeding (₦/kg) and feed cost per kilogram weight gain (₦/kg). All the treatments to which additives were added were of the same price due to equal price of the additives and the same quantity added to each of the diet. Pigs on treatment 2 had the highest total feed intake (kg), daily feed intake (kg), total cost of feeding (₦), average cost of feed per day (₦), least numerical feed conversion ratio and feed cost per kilogram weight gain (₦/kg). The combination of improved weight gain and best feed conversion efficiency may have been responsible for higher returns in diet 2 compared to other diets.

Table 3: Economic analysis of weaned pigs fed diets supplemented with three different feed additives

Parameters	T1 (Control)	T2 (DFM)	T3 (MB)	T4 (YC)	**±SEM**
Cost of feed (₦/kg)	94.00^{b}	97.00^{a}	97.00^{a}	97.00^{a}	0.20
Total feed intake (kg)	45.71^{ab}	50.10^{a}	50.06^{a}	44.04^{b}	0.85
Average daily feed intake (g)	725.48^{ab}	795.24^{a}	794.66^{a}	698.97^{b}	13.48
Total cost of feeding (₦)	4296.7^{b}	4859.7^{a}	4855.8^{a}	4271.9^{b}	83.78
Average cost of feed per day (₦)	68.19^{b}	77.13^{a}	77.08^{a}	67.80^{b}	1.33
Feed conversion ratio	3.15	3.01	3.36	3.53	0.12
Feed cost/kg weight gain (₦/kg)	346.76	342.94	378.88	393.01	11.90

DFM – Multi-strain direct fed microbial, MB- mycotoxin binder and YC-yeast culture

The haematological parameters of weaned pigs fed diets supplemented with the three feed additives are shown in Table 4. There were significant ($P<0.05$) effects of supplementation of feed additives on neutrophils, eosinophils, lymphocytes and platelet but not on PCV and hemoglobin ($P>0.05$), even though higher numerical values of 33.25 % and 11.05 g/dl were recorded for PCV and hemoglobin respectively in the pigs fed control diet compared to the test treatments.

The mean total platelet counts (164/10^{3}ul) was significantly ($P<0.05$) higher in the control diet compared to all the additive supplemented diets. RBC and WBC were numerically higher ($P>0.05$) in the pigs fed DFM treatments, though not statistically significantly different from the control and mycotoxins binder treatments. The haematological values were within the normal range for healthy pigs (15; 16)

Table 4: Haematological parameters of weaned pigs fed diets supplemented with three different feed additives

Parameters	T1 (Control)	T2 (DFM)	T3 (MB)	T4 (YC)	**±SEM**
PCV (%)	33.25	31.25	33.00	31.63	0.81
Haemoglobin (g/dl)	11.05	10.38	10.98	10.50	0.27
RBC(10^{6}ul)	4.97	5.86	4.99	5.51	0.17
WBC (10^{3}ul)	6.56	7.13	5.93	7.12	0.36
Neutrophils (%)	31.13^{ab}	31.13^{ab}	27.00^{b}	38.50^{a}	1.87
Eosinophils (%)	1.75^{b}	2.75^{ab}	3.50^{a}	2.5^{ab}	0.27
Monocytes (%)	3.50	3.50	3.25	4.00	0.24
Lymphocytes (%)	63.63^{ab}	61.63^{ab}	66.00^{a}	55.00^{b}	1.75
Platelet (10^{3}ul)	164.38^{a}	133.00^{ab}	112.38^{b}	108.88^{b}	8.97

DFM – Multi-strain direct fed microbial, MB- mycotoxin binder and YC-yeast culture
PCV - packed cell volume, RBC - red blood cells, WBC – white blood cells

Blood serum was analysed and the result is presented in Table 5. Serum total protein, albumin, creatinine, urea, and glucose concentrations have been described as valuable parameters of hepatic injury and function. Likewise activity of serum enzymes such as ALP, ALT and AST provides a sensitive and specific measure of hepatic function or injury

(17; 18). The result showed significant effect of the additive on the AST, ALT, ALP, glucose and albumin. Only ALP (10 vs. 257 U/L) and total protein (59 vs. 84 g/L) were below the normal reference range for healthy pigs for both control and other diets. Likewise, apart from cholesterol (142 vs. 45 mg/dL) and urea serum values (34 vs. 20 mg/dL) that are higher than the normal value for healthy pigs, all other haematological parameters were within the normal range reported for normal pigs and variations observed was not beyond those reported for normal healthy pigs (19).

Table 5: Serum Biochemical Parameters of weaned pigs fed diets supplemented with three different feed additives

Parameters	T1 (Control)	T2 (DFM)	T3 (MB)	T4 (YC)	±SEM
AST (U/L	41.22^{bc}	46.82^{ab}	35.54^{c}	52.37^{a}	2.02
ALT (U/L)	41.33^{ab}	37.80^{b}	50.61^{a}	49.64^{a}	1.84
ALP (U/L)	8.71^{b}	9.92^{a}	9.87^{a}	10.39^{a}	0.22
Glucose (mg/dL)	92.03^{b}	101.49^{a}	98.27^{ab}	93.36^{ab}	1.45
Total protein (g/dL)	6.05	6.14	5.85	5.61	0.18
Albumin (g/dL)	3.16^{a}	2.18^{b}	2.75^{ab}	2.99^{a}	0.13
Cholesterol (mg/dL)	136.71	141.25	149.86	141.25	3.04
Creatinine (mg/dL)	1.36	1.83	2.02	1.66	0.13
Urea (mg/dL)	34.60	33.04	35.17	34.60	1.36

DFM – Multi-strain direct fed microbial, MB- mycotoxin binder and YC-yeast culture; ALT - Alanine aminotransferase, ALP – Alkaline phosphatate, AST - Aspartate aminotransferase

Conclusion and Application

1. It can be concluded that multi-strain DFM enhanced the growth performance of weaned pigs more than yeast culture and mycotoxin binder, while none of the feed additives had adverse effect on haematological and serum biochemical parameters.
2. The use of antibiotics as antimicrobial growth promoter in pigs should be discouraged, while multi-strain direct fed microbial could be embraced for improved growth performance and better health status of post weaned pigs.

Acknowledgement

The authors are grateful to Dr K. Oppong-Anane (Director of Basic Environmental Systems and Technology, Accra, Ghana) and Dr David Akinde (Fusion BioSystems GmbH, D-49393 Lohne, German) for the supply of the DFM (RE3) and Fusion (Bentonite) respectively. We also appreciate the supply of Bg-Max (Yeast culture) from Arm & Hammer Animal Nutrition, Princeton, USA.

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