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PRELIMINARY ASSESSMENT OF YEAST (*Saccharomyces cerevisiae)* PRODUCTS ON AFRICAN CATFISH PRODUCTION AND HEALTH

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ABSTRACT
A preliminary study was conducted to evaluate the effects of four products of yeast (Saccharomyces cerevisiae) on African catfish production and health. The catfish (11.77±0.05 g fish^{-1}, 100 fish 857L tank^{-1}) were fed either of diets supplemented with 0% yeast product (Con), 3% unextracted yeast (UnY), 0.3% hydrolysed yeast (HyY), 0.2% mannan-oligosaccharides (MoS) or 0.02% beta-glucan (Bg-S). After 63 days of feeding to apparent satiation, there was no significant difference (P < 0.05) observed in the final body weights, feed conversion ratios, specific growth rates, protein efficiency ratios and survivals of the catfish fed diets supplemented with the yeast products and those fed the control diet. Similarly, the somatic indices and haematological parameters measured were not significantly different (P > 0.05) among the catfish fed the experimental diets. It could be inferred from this study that yeast products (at current levels of inclusion) do not negatively affect the production nor impair the health of African catfish. However, a follow-up dose-response trial will be very informative for comprehensive assessment of each of the yeast products on African catfish production and health.

Keywords: *Clarias gariepinus*, immunostimulant, performance, welfare

INTRODUCTION

The aquaculture industry contributed 47% to global fish production in 2016 and accounted for more than half of the world's fish for direct human consumption (FAO, 2018). However, with increasing consumption of fish, natural fish stocks decrease while aquaculture continues to be responsible for the impressive growth in the supply of fish for human consumption (FAO, 2018). No doubt, aquaculture is a fast-growing food-producing industry in the world and will continue to be the engine that will drive growth in the global fish production for human consumption. To this end, there is need for the intensification of aquaculture practices to meet the rising human demand for fish consumption (Msangi *et al*., 2013). It is important to note however that the intensification of aquaculture operations is often accompanied by stress due to suboptimal environmental conditions resulting from overcrowding and overfeeding. This condition may be stressful for fish and subsequently compromise fish immune response leaving fish prone to infection and disease by opportunistic pathogens and consequently resort to decreased growth performance and poor yield. However, to keep up with increasing human demand for fish, the suboptimal environmental conditions associated with intensive aquaculture is likely to continue. The concept of immune-nutrition (production of high-quality feed with optimal growth and immune-boosting effects) could enhance the performance and up regulate the immunity of aquaculture species under intensive aquaculture operations characterise by stressful conditions (Nakagawa *et al*., 2007; Kiron, 2012).

One of the ways to achieve the concept of immunonutrition is by dietary supplementation of immunostimulants, which are classified as functional feed additives (Dawood *et al*., 2018). Yeast (*Saccharomyces cerevisiae*), one of the commonly used immunostimulants in aquaculture is capable of enhancing the performance, health and immunity of aquaculture species. In addition to its relatively high protein, amino acids, energy and micronutrients content, cell and cell wall of yeast (*S. cerevisiae*) contains nutraceutical compounds such as β-glucans, mannan-oligosaccharides and nucleotides that have been demonstrated to improve growth performance, health and diseases resistance of aquaculture species (Huyben *et al*., 2017; Xue *et al*., 2017; Shurson, 2018). The growth and health benefits of yeast and yeast products have been reported in *Labeo rohita* (Amir *et al*., 2018), rainbow trout (Huyben *et al*., 2017; Ji *et al*., 2017), turbot (Librán-Pérez *et al*., 2018), sea bream (Dimitroglou *et al*., 2010; Gultepe *et al*., 2011; Dawood *et al*., 2017), tilapia (Sado *et al*., 2008; Ozório *et al*., 2012; Pilarski *et al*., 2017; Hassaan *et al*., 2018), largemouth bass (Zhou *et al*., 2018), Pacific white shrimp (Zhang *et al*., 2012; Qiu and Davis, 2017; Jin *et al*., 2018), Jian carp (Yuan *et al*., 2017), gibel carp (Zhang *et al*., 2018), common carp (Momeni-Moghaddam *et al*., 2015), hybrid striped bass (Li and Gatlin, 2003), giant freshwater prawn (Prasad *et al*., 2013), Thai panga (Pongpet *et al*., 2016), channel catfish (Peterson *et al*., 2012),

seabass (Torrecillas *et al*., 2011, 2007, 2015; Salem *et al*., 2016) and pacu (Sado *et al*., 2014). However, there is insufficient studies on the effects of yeast and yeast products on African catfish (*C. gariepinus*) production and health except for studies carried out on yeast as an alternate protein ingredient in the diets of African catfish (Hoffman *et al*., 1997; Aderolu *et al*., 2011; Ezenwaji *et al*., 2012; Solomon *et al*., 2017). Therefore, the objective of this study was to carry out a preliminary assessment of yeast (*S. cerevisiae*) products (unextracted yeast, hydrolysed yeast, mannan-oligosaccharides and β-glucans) on African catfish (*C. gariepinus*) production and health.

MATERIALS AND METHODS

Experimental design and diets preparation.

The trial was carried out in a flow-through aquaculture system of the Department of Aquaculture and Fisheries Management of Federal University of Agriculture, Abeokuta – Nigeria. The flow-through system contains 15 circular tanks (857 L capacity each) and were supplied with freshwater from a deep well. One thousand and five hundred African catfish (*C. gariepinus*) of mean weight 11.77±0.05 g obtained from a reputable hatchery were randomly distributed (100 catfish per tank) into the 15 tanks after two weeks of acclimatisation. The photoperiods and water temperatures were maintained at ambient condition.

Five iso-nitrogenous and iso-lipidic diets were formulated (Table 1) as Con (containing 0% yeast product), UnY (containing 3% unextracted yeast), HyY (containing 0.3% hydrolysed yeast, CeFi® ^{pro}), MoS (containing 0.2% mannan-oligosaccharides, Biolex® MB40) and Bg-S (containing 0.02% highly purified β-glucans, Beta-S) diets. The inclusion levels (3% for unextracted yeast, 0.3% for hydrolysed yeast, 0.2% for mannan-oligosaccharides and 0.02% for β-glucan) of the yeast products were based on the recommendation of the products' manufacturer, Leiber GmbH. The feed ingredients were thoroughly mixed, moistened (200 mL kg^{-1}) and then cold press extruded to produce 2mm pellets using a flat die pelleting machine. The diets were sun dried and their proximate composition analysed (Table 1) using AOAC protocols (AOAC, 1996). After drying, the diets were stored in airtight containers prior to use. The African catfish (*C. gariepinus*) were fed the experimental diets daily (09.00 h and 16.00 h) to apparent satiation for 9 weeks in two equal rations. Daily feed was adjusted according to tank biomass on a weekly basis by batch weighing following a 24 h deprivation period.

Growth, feed utilisation and somatic indices.

Growth performance, feed utilisation and somatic indices were assessed by final body weight (FBW), specific growth rate (SGR), feed conversion ratio (FCR), protein efficiency ratio (PER), hepatosomatic index (HSI), viscerosomatic index (VSI) and condition factor (K-factor). Calculations were carried out using the following formulae described by Adeoye *et al*., (2016a).

SGR = $((lnFBW - lnIBW)/T)X\ 100$
Where FBW = final body weight (g) and IBW = initial body weight (g)
FCR = FI/WG
Where FI = feed intake (g) and WG = wet weight gain (g)
PER = WG/PI
Where WG = wet weight gain (g) and PI = protein ingested (g)
HSI = $(LW/BW)X\ 100$
Where LW = liver weight (g) and BW = body weight (g)
VSI = $(VW/BW)X\ 100$
Where VW = visceral weight (g)
K-factor = $(100\ X\ BW)/\ [\![TL]\!]$ ^3
Where BW = body weight (g) and TL = total length (cm)

All fish were euthanized with clove oil at a concentration of 100 mg L^{-1} followed by destruction of the brain prior to sampling. At the end of the trial, three fish per tank were sampled and used to record viscera weight and whole-body weight to calculate the somatic indices (K-factor, HSI and VSI).

Haematology.

At the end of the feeding trial, blood from three fish per tank (n = 9 per treatment) was taken from the caudal arch using a 25-gauge needle and 1 mL syringe after the fish were anaesthetized with clove oil (100 mg L^{-1}). Blood samples were prepared and analysed as previously described by Adeoye *et al*., (2016b).

Statistical analysis.

All data are presented as mean ± standard deviation. Data were analysed using one-way analysis of variance (ANOVA). Multiple comparisons were performed using Turkey post-hoc test. Differences were considered significant at a value of $P < 0.05$. The statistical analysis was carried out using SPSS for Windows (SPSS Inc., 24.0, Chicago, IL, USA).

RESULTS

Table 2 shows the growth performance and feed utilisation of the African catfish (*C. gariepinus*) fed the experimental diets. At the end of nine weeks of feeding to apparent satiation, the final body weights (g $fish^{-1}$) of the African catfish experienced more than five folds increase with specific growth rates that ranged from 3.05 – 3.18 % day^{-1}, feed conversion ratios that ranged from 1.27 – 1.32 and protein efficiency ratios that ranged from 1.64 – 1.72. However, there was no significant difference

(P > 0.05) observed in the final body weights, feed conversion ratios, specific growth rates, protein efficiency ratios and survivals of the catfish fed the experimental diets.

The somatic indices (K-factor, hepatosomatic indices, and viscerosomatic indices) and survivals of the African catfish (*C. gariepinus*) fed the experimental diets are shown in Table 2. After nine weeks of feeding the catfish with the experimental diets, the catfish K-factors ranged from 0.75 – 0.83, the hepatosomatic indices ranged from 0.78 – 1.07, the viscerosomatic indices ranged from 10.2 – 12.0 and the survivals ranged from 88.3 – 94.4%. However, the somatic K-factors, hepatosomatic and viscerosomatic indices were not significantly different (*P > 0.05*) among the catfish fed diets.

The haematological profile of the African catfish (*C. gariepinus*) fed the experimental diets are presented in Table 3. The haematological parameters such as haematocrit (%PCV), haemoglobin (g dL^{-1}), red blood cells count (10^{12} L^{-1}), white blood cells count, neutrophils (%), lymphocytes (%), basophil (%), eosinophil (%), monocytes (%), MCV (fL), MCH (pg) and MCHC (g dL^{-1}) were not significantly different (*P > 0.05*) among the fish fed diets supplemented with the yeast products and those fed the control diet.

Table 1. Formulation and composition of the experimental diets

Ingredients (%)	Con	UnY	HyY	MoS	Bg-S
Fishmeal (65% CP)	22.0	22.0	22.0	22.0	22.0
Whole shrimp meal	1.00	1.00	1.00	1.00	1.00
Soybean meal (solvent extracted)	40.0	40.0	40.0	40.0	40.0
Groundnut cake	14.0	11.0	13.7	13.8	14.0
Maize	9.49	9.49	9.49	9.49	9.49
Whole wheat meal	7.50	7.50	7.50	7.50	7.50
Soybean oil	4.00	4.00	4.00	4.00	4.00
Fish oil	1.00	1.00	1.00	1.00	1.00
Vitamin mineral premix	1.00	1.00	1.00	1.00	1.00
Brewers' yeast unextracted	0.00	3.00	0.00	0.00	0.00
CeFi® pro	0.00	0.00	0.30	0.00	0.00
Biolex® MB40	0.00	0.00	0.00	0.20	0.00
Beta-S	0.00	0.00	0.00	0.00	0.02
Anti-oxidant	0.01	0.01	0.01	0.01	0.01
Total	**100.0**	**100.0**	**100.0**	**100.0**	**100.0**
Composition (% dry weight)					
Moisture	11.7	11.5	12.6	11.9	11.4
Crude protein	36.1	38.8	36.5	38.5	37.5
Lipid	12.3	11.8	11.9	12.0	11.5
Ash	8.11	8.06	8.02	8.11	8.40
NFE	28.3	26.1	27.7	26.9	27.0
Crude fibre	3.48	3.69	3.35	2.72	4.21

CeFi® pro, hydrolysed yeast produced by Leiber GmbH; Biolex® MB40, Mannan-oligosaccharides produced by Leiber GmbH; Beta-S, highly purified β-glucans produced by Leiber GmbH; NFE, Nitrogen-free extract

Table 2. Growth, feed utilisation and somatic indices of African catfish fed the experimental diets

Parameters	Experimental diets				
	Con	**UnY**	**HyY**	**MoS**	**Bg-S**
IBW (g $fish^{-1}$)	11.8±0.05	11.8±0.05	11.8±0.05	11.8±0.05	11.8±0.05
FBW (g $fish^{-1}$)	65.4±2.42	63.3±1.08	62.2±2.74	61.4±4.97	64.8±3.10
SGR (% day^{-1})	3.18±0.09	3.11±0.04	3.08±0.09	3.05±0.19	3.16±0.09
FCR	1.31±0.05	1.29±0.03	1.31±0.05	1.32±0.09	1.27±0.04
PER	1.67±0.07	1.69±0.04	1.66±0.08	1.64±0.14	1.72±0.06
K-factor	0.80±0.09	0.81±0.12	0.79±0.05	0.83±0.12	0.75±0.08
HIS	0.87±0.16	0.78±0.28	1.07±0.19	0.95±0.19	1.01±0.19
VSI	10.7±0.96	10.2±1.06	11.5±1.54	12.0±3.02	10.4±1.18
Survival (%)	88.3±0.94	93.7±1.25	90.0±1.41	92.3±1.89	94.4±6.55

There was no significant difference (*P < 0.05*) in the growth, feed utilisation and somatic indices of African catfish fed the experimental diets. IBW, initial mean body weight; FBW, final mean body weight; SGR, specific growth rate; FCR, feed conversion ratio; PER, protein efficient ratio; HSI, hepatosomatic index and VSI, viscera-somatic index.

Table 3. Haematological parameters of African catfish fed the experimental diets

Parameters	Experimental Diets				
	Con	**UnY**	**HyY**	**MoS**	**Bg-S**
Haematocrit (%PCV)	28.2±0.69	27.4±4.35	27.11±1.90	25.9±1.07	28.6±0.57
Haemoglobin (g dL^{-1})	9.47±0.36	9.24±1.49	9.17±0.62	8.75±0.31	9.58±0.20
WBC (10^9 L^{-1})	110±15.8	126±9.36	111±8.03	83.1±5.94	114±13.2
RBC (10^{12} L^{-1})	1.63±0.01	1.59±0.27	1.59±0.10	1.47±0.09	1.68±0.05
Neutrophil (%)	20.2±6.74	23.6±8.13	18.7±5.55	21.6±7.90	24.4±4.79
Lymphocytes (%)	76.1±5.93	70.4±9.74	79.2±4.67	75.6±7.25	70.4±6.49
Basophil (%)	0.56±0.20	0.56±0.39	0.11±0.20	0.78±0.51	0.67±0.27
Eosinophil (%)	1.67±1.20	2.56±1.39	0.89±0.70	1.22±0.70	2.56±0.57
Monocytes (%)	1.45±0.39	2.89±1.68	1.11±1.02	0.89±0.38	1.89±1.10
MCV (fL)	173±3.44	173±3.12	171±1.94	175±2.56	170±3.87
MCH (pg)	58.1±1.87	58.2±0.70	57.9±0.55	59.8±1.66	57.0±1.64
MCHC (g dL^{-1})	33.5±0.41	33.7±0.30	33.8±0.32	33.9±0.34	33.6±0.15

There was no significant difference ($P < 0.05$) in the haematological parameters of African catfish fed the experimental diets. WBC, leucocytes; RBC, red blood cells; %, mean percentage of total leucocytes; MCV, mean corpuscular volume (haematocrit (%PCV) × 10) / RBC (10^9 L^{-1}); MCH, mean corpuscular haemoglobin (haemoglobin (g dL^{-1}) × 10) / RBC (10^{12} L^{-1}); MCHC, mean corpuscular haemoglobin concentration (haemoglobin (g dL^{-1}) × 100) / haematocrit (%PCV).

DISCUSSION

This study is the first trial (to our knowledge) to assess the dietary effects of different yeast (*S. cerevisiae*) products (i.e. brewers' yeast unextracted, hydrolysed yeast, mannan-oligosaccharides and highly purified β-glucans) on African catfish (*C. gariepinus*) production and health, except for studies carried out by Hoffman *et al*., (1997), Aderolu *et al*., (2011), Ezenwaji *et al*., (2012) and Solomon *et al*., (2017) on the effect of yeast as an alternate protein ingredient in the diets of African catfish. In this study, diet supplemented with 3% brewers' yeast unextracted (UnY) would not impair the growth performance of the African catfish when compared with those fed the control diet; the final body weight of the catfish experienced more than five folds increase after nine weeks of feeding to apparent satiation with specific growth rate of 3.11 % day^{-1}, feed conversion ratio of 1.29 and protein efficiency ratio of 1.69. Our findings are quite contrary to the report of Solomon *et al*., (2017) who reported best growth performance (less than five folds increase in final body weight of the catfish with specific growth rate of 2.7 % day^{-1}, feed conversion ratio of 2.24 and protein efficiency ratio of 1.29) among African catfish fed diet with no brewers' yeast when compared to those fed diets containing increasing levels of brewers' yeast (Solomon *et al*., 2017). This difference could possibly be attributed to the different levels of inclusion of brewers' yeast in the two studies; the inclusion levels of brewers' yeast
by Solomon *et al*., (2017) ranged from 14.45 – 57.8% unlike 3% level of inclusion used in the present study.

It was then inferred that increasing levels of brewers' yeast appeared not to support the growth of African catfish noting that palatability of diets

reduced with increasing levels of brewers' yeast. Solomon *et al*., (2017) therefore concluded that optimal range of including brewers' yeast in the diet of African catfish is between 1 – 14%. Unlike the findings of Solomon *et al*., (2017), Aderolu *et al*., (2011) reported that yeast at all levels of inclusion (0%, 2%, 4%, 6% and 8%) enhanced the growth performance and feed utilisation of African catfish noting that the best performance was however recorded among the catfish fed diet supplemented with 4% yeast. The improved growth performance and feed utilisation in the catfish was attributed to improved nutrient digestibility of diets supplemented with yeast (Aderolu *et al*., 2011). Even though, Solomon *et al*., (2017) reported that dietary supplementation of yeast did not appear to enhance African catfish performance (at 14.45 - 57.8% levels of inclusion), nonetheless the findings of Aderolu *et al*., (2011) somewhat agrees with the final suggestion of Solomon *et al*., (2017) that the optimal range of inclusion level of yeast in the diet of African catfish is between 1 – 14%. The level of inclusion of yeast (3%) in the present study also falls within the range suggested by Solomon *et al*., (2017). In another study where, African catfish was fed diets containing different inclusion levels (1.53 – 15.45%) of yeast, it was reported that all the African catfish died within the first week of being fed diet containing 15.45% yeast (Ezenwaji *et al*., 2012). However, those within the range (1 – 7.72%) suggested by Solomon *et al*., (2017) survived till the end of the trial with slight increase in growth

response and best growth performance recorded among the catfish fed diet containing 7.72% yeast.

The African catfish fed diet supplemented with 0.3% hydrolysed yeast (HyY) did not show significant increase in growth performance when compared with those fed the control diet (Con). However, a significant improvement in the growth of Jian carp was reported when fed diet supplemented with 3% hydrolysed yeast (Yuan *et al*., 2017). The difference observed in the growth performance could be attributed to higher level of inclusion of hydrolysed yeast in the experimental diets for the Jian carp.

The growth performance of the African catfish fed diet supplemented with 0.2% mannan-oligosaccharides (MoS) showed no significant difference when compared to the catfish fed the control diet (Con). This contradict findings by Ali *et al*., (2017) who reported a significant increase in the growth performance of Asian seabass (*Lates calcarifer*) fed diet supplemented with 1% mannan-oligosaccharides. Apart from difference in species, the difference in the growth performances between the African catfish and the Asian sea bass could be due to lower level of inclusion of MoS (recommended by the product's manufacturer) in the present study as against higher level of inclusion by Ali *et al*., (2017).

The growth performance of the African catfish fed diet supplemented with 0.02% highly purified β-glucans (Bg-S) did not show significant improvement when compared to the catfish fed the control diet (Con) in terms of growth performance. However, this result is contrary to report from Ji *et al*., (2017) who fed rainbow trout (*Oncorhynchus mykiss*) with diets supplemented with graded levels of β-glucans. The rainbow trout demonstrated better growth performance when fed diets supplemented with 0.1% and 0.2% β-glucans compared to those fed the control diet (containing no β-glucans). In another vein, Pilarski *et al*., (2017) reported a different growth performance when Nile tilapia were fed different β-glucans of same dosage (0.01%); one influenced the growth of the Nile tilapia positively but the second did not. To this end, the effect of β-glucans on fish growth performance could not only be attributed to different dosage levels but also the source and biotechnical methods employed by different manufacturers to extract the β-glucans.

The insignificant difference observed in the values of the somatic indices among the African catfish fed the yeast products (UnY, HyY, MoS and Bg-S) indicated that dietary supplementation of the yeast products (at current levels of inclusion) had no negative effect on the liver functionally and no excessive hepatic accumulation of fat or carbohydrate as the values were within the normal range (Piccolo *et al*., 2017). In addition, since the condition factor and survival among the African catfish fed diets supplemented with the yeast products showed no significant difference compared with those fed the control diet, then it could be inferred from this study that the yeast products would not impair liver or intestinal physiological well-being and survival of the African catfish.

In this study, haematological parameters such as haematocrit, haemoglobin and red blood cells count which are indicators of health status in fish (NRC, 2011) as well as parameters such as white blood cells count and differential leucocyte (important parameters in the non-specific immunity of fish) were measured for preliminary assessment of the health status of the African catfish. These parameters were not markedly different among the catfish fed diets supplemented with the yeast products and those fed the control diet that contain no yeast product. This indicates that dietary inclusion of the yeast products (at current levels of inclusion) do not have deleterious effect on African catfish haematology. This finding is not different from what Aderolu *et al*., (2011) reported when African catfish were fed graded level of yeast; no significant difference were observed in the haemoglobin, haematocrit, red blood cells count, white blood cells count, mean corpuscular haemoglobin and mean corpuscular volume of the catfish fed the experimental diets. On the contrary, Ezenwaji *et al*., (2012) reported a significant increase in African catfish haematocrit, haemoglobin, red blood cells count and white blood cells count when fed diets supplemented with 3.09% and 7.72% brewers' yeast compared to those fed the control diet. Again, this difference could be partly attributed to lower dosage level of brewers' unextracted yeast (3%) used in the present study in additon to different sources and methods employed by manufacturers of yeast products.

CONCLUSION

In conclusion, it could be inferred from the present study that at current levels of inclusion (3% brewers' yeast unextracted, 0.3% hydrolysed yeast, 0.2% mannan-oligosaccharides and 0.02% highly purified β-glucans), the yeast products do not negatively affect the growth performance and haematological parameters of the African catfish when compared to the those fed the control diet (that did not contain any of the yeast product). To this end, this study could be regarded as preliminary work and a proof of concept on which a follow-up dose response trial can be planned for comprehensive and more informative assessment of each of the yeast products in African catfish production and health.

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