EFFECTS OF DIFFERENT LEVELS OF YEAST (*Saccharomyces cerevisiae*) ON BINDING PROPERTY OF OKRA (*Abelmoschus esculentus*) IN FISH FEED

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ABSTRACT

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Three binders were tested: mature okra, immature okra and starch. All binders were added at 5%. Nine diets were prepared each containing 35% crude protein. Three feeds had no yeast (control for each of the three dietary groups). Three feeds contained 5% yeast while the other three contained 10% yeast each. The physical characteristics of the feeds were assessed using the following parameters: floatability, bulk density, water absorption index, thickness swelling, water stability and nutrient retention. The highest floatability was recorded on mature okra with 10% yeast "$Y^{10}M$" (360.80±15.59) while starch with no yeast "Y^0S" had the least (20.67±0.33). Y^0S had the highest water absorption index while immature okra with 10% yeast "$Y^{10}I$" had the least. $Y^{10}M$ had the highest thickness swelling while mature okra without yeast "Y^0M" had the least. There was a significant difference (P<0.05) in floatability, water absorption index and thickness swelling of diets. There was no significant difference (P>0.05) in bulk density of the diets. Mature okra with 10% yeast gave the highest floatability and thickness swelling while starch bounded feed with 0% yeast had the least floatability and highest water absorption index. Mature okra with 10% yeast would give a better fish feed pellet.

Keywords: *Binders, floatability, nutrient retention, okra, yeast.*

INTRODUCTION

Aquaculture is the cultivation of aquatic produce such as plants, fish and other animals. Fish feed is of great importance in aquaculture. Fish feeding is a major factor that determines the profitability of an aquaculture venture. Feeding accounts for 60-80% of the total cost of fish production in Africa (Orire and Sadiku, 2014; Adekunle *et al*., 2012). Cost of production, to a large extent, determines the viability and profitability of a fish farming enterprise. A well bound feed allows delivery and utilization of nutrients by fish and minimize leaching (Tiamiyu *et al*., 2003). The medium of culture for fish is water. And water being a universal solvent, could dissolve and leach the bonded nutrients into the water. Such leaching will pollute the water and also make the leached nutrients unavailable to fish. So, the aquatic environment would not favour the use of mash in feeding fish. Several methods have been employed in making sure that fish feed does not leach easily into the water media. One of such methods is to make use of binders which could be in the feed material or externally added. Starch based binders are commonly used in fish feed. Natural feed ingredients having high starch contents like millet, wheat grain, corn, guinea corn, yam and cassava have been used as binders of fish feed pellets. Some other materials commonly used as binders are agar, guar gum, carboxyl-methyl-cellulose, and carrageenan. Factors limiting the use of some of these binders includes: contribution to the nutritional value of the feed, cost and availability.

Local ingredients will reduce cost of production, encourage farmers to produce and also contribute to sustainable utilization of such ingredients. One of such methods is to make use of binders which could be in the feed material or externally added. Starch based binders are commonly used in fish feed. Natural feed ingredients having high starch contents like millet, wheat grain, corn, guinea corn, yam and cassava have been used as binders of fish feed pellets. Some other materials commonly used as binders are agar, guar gum, carboxyl-methyl-cellulose, and carrageenan. Factors limiting the use of some of these binders includes: contribution to the nutritional value of the feed, cost and availability. There has been little report on the use of mucilage or mucilaginous plants as feed binders. Okra (*Abelmoschus esculentus*) known in many English-speaking countries as lady's fingers or gumbo, is a flowering plant in the mallow family. Okra is an annual herb that is widely cultivated for its edible green seed pods in tropical, sub-tropical and warm temperate climates. Okra is a hardy plant that can grow even with low level of water and in hot conditions. *Abelmoschus esculentus* (Okra) contains mucilage, has appreciable nutritional value and it is readily available. The objective of this research is to evaluate the physical characteristics of fish pellets using two stages of okra (*Abelmoschus esculentus*) maturity at a fixed level as binder and yeast at varying inclusion level as a rising agent. The starch diets serves as the control.

MATERIALS AND METHODS

Feed ingredients were purchased from a feed mill in Akure, Ondo State, Nigeria. The feed ingredients were reduced to fine particle. Nine feeds, each containing 35% crude protein, were formulated using Pearson square method (Table 1). Three types of binders were tested: mature okra, immature okra and starch. The mature and immature okra were separately mixed at 5%w/v, 75 g of okra to 1500 ml of water. The starch was prepared by mixing 75 g of dry cassava starch with 1200 ml of cold water, then 300 ml of hot water. It was then allowed to cool before mixing with other ingredients. All binders were added at 5% inclusion level. Each of the binder was added to other ingredients and thoroughly mixed until a homogenous paste (dough) was obtained.

Three sets of feeds were produced from each of the binders. Three feeds contained 0% yeast, these served as the control for each of three groups. Three contained 5% yeast while the other three contained 10% yeast each. The ingredients were weighed using an electronic sensitive weighing balance (Model JY10S-01). The dough was pelleted using a Hobart A-200T mixing and pelleting machine. The pellets were 6mm in diameter. The pelleted feeds were sundried on raised platform. The dried feeds were packed in polythene bags and labelled. Samples of the pelleted feeds were taken to the laboratory for proximate analysis. The proximate analysis was done following standard procedures of AOAC (2005).

Determination of physical properties of feed

The physical characteristics of the pelleted feed were assessed by measuring the following parameters; floatability, bulk density, water absorption index, thickness swelling, hardness, nutrient retention (Falayi *et al.*, 2004; Orire *et al.,* 2010; Adeparusi and Famurewa 2011). All physical characteristics were done in triplicates.

Feed floatability test

Floatability tests for the feeds were performed using a glass measuring cylinder filled to the 250 ml mark with borehole water. Triplicates samples of the each pelleted feed bonded with starch or okra with varying inclusion level of yeast were placed in the measuring cylinder. The time period for the feeds to float on the water was noted using stopwatch.

Determination of bulk density

The bulk density was calculated using the formula as described by Adeparusi and Famurewa, 2011;

$$\text{Bulk Density (BD)} = \frac{M2 - M1}{V}$$

where,

M2=Final mass of sample after soaking (g)

M1=Initial weight of sample before being soaking (g)

V=Volume of cylinder (cm3)

Determination of Water Absorption Index

The water absorption index was determined using the formula (Adeparusi and Famurewa, 2011);

$$\text{Water Absorption Index (WAI)} = \frac{M2 - M1}{M1}$$

where:

M2 = mass of pellet after immersion in water

M1 = mass of pellet before immersion in water

Determination of Thickness Swelling

The thickness strength of the pellet sample was determined by dividing the difference of T1 and T2 by T1, i.e. according to Adeparusi and Famurewa, 2011

$$\text{Thickness strength} = \frac{T2 - T1}{T1}$$

where,

T1= the thickness of the pellet sample before immersion (cm)

T2= the thickness of a pellet sample after being immersed in water for three Minutes (cm).

Determination of Nutrient Retention

Fifteen grams (15g) of each diet was soaked in water for 30 minutes. The nutrients retained in the feeds were determined with the procedure used for the proximate analysis (AOAC, 2005). The values of nutrients retained were expressed in percentage remaining basis (Falayi *et al*, 2003).

$$\text{Nutrient retention (\%)} = \frac{N2 \text{ x } 100\%}{N1}$$

where,

N1 = the initial nutrient composition of the pellet before immersion in water

N2 = the final nutrient composition of the pellet after immersion in water

Determination of water stability

Triplicates samples (10g each) of each pelleted feed were placed in a nylon sieve tied with rope and slowly immersed in 500 cm^3 of tap water. Aeration was done using ceiling fan. There was occasional shaking for 10 seconds every 2 minutes. The pellets were allowed in the water medium for a period of 20 minutes. At the end of each test time, the sacks were removed with aid of the rope. The crumbles were allowed to drain for one minute and the content put in a petri-dish and oven dried at 105°C for 2 hours. It was then cooled in desiccator and weighed. The new weight represents the left over from the original 10 g.

Water stability of the pellet was calculated using the equation as described by Solomon *et al.* (2011)

$$\% \text{ Water stability } = \frac{\text{Weight of retained whole pellets} \quad x\ 100}{\text{Initial total weight of pellets}}$$

Crushing load test (hardness test)

The crushing load test of the experimental feeds was carried out using a Californian Bearing Ration machine (Model no BS1377) at the Department of Applied Geology, FUTA. The load-measuring device was connected to compression machine. The mould with the sample and the surcharge weight was placed in the machine. A plunger was seated on top of the specimen and it was done to allow free movement of the plunger from the surcharge weight. The seating load was applied by weight. The displacement-measuring device was adjusted to zero. The motor drive was switched on the loading started with a loading rate of 1mm/min. The point at which the experimental feeds begin to crumble was noted. The breaking point in mm was multiplied with the applied force of 23.8N.

Statistical analysis

Data were subjected to analysis of variance (ANOVA) and separation of means was done with Duncan multiple range test.

Table 1: Gross composition (g per 100g) and proximate composition of experimental feeds

Ingredients	Y^0M	Y^5M	$Y^{10}M$	Y^0I	Y^5I	$Y^{10}I$	Y^0S	Y^5S	$Y^{10}S$
Fish Meal (72%)	15.02	15.02	15.02	15.02	15.02	15.02	15.02	15.02	15.02
Groundnut cake (40%)	30.04	25.04	20.04	30.04	25.04	20.04	30.04	25.04	20.04
Wheat Offal (16%)	11.81	11.81	11.81	11.81	11.81	11.81	11.81	11.81	11.81
Maize (10%)	23.63	23.63	23.63	23.63	23.63	23.63	23.63	23.63	23.63
Bakers' Yeast (48.25%)	0	5	10	0	5	10	0	5	10
Okra (24.32%)	5	5	5	5	5	5	0	0	0
Starch	0	0	0	0	0	0	5	5	5
Lysine	1	1	1	1	1	1	1	1	1
Methionine	1	1	1	1	1	1	1	1	1
Vitamin and Mineral Premix*	4	4	4	4	4	4	4	4	4
Vegetable Oil	3	3	3	3	3	3	3	3	3
Bone Meal	5	5	5	5	5	5	5	5	5
Salt	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Total	100	100	100	100	100	100	100	100	100
Proximate composition of experimental diets (% dry matter basis)									
Moisture	4.67	5.33	4.33	5.67	5.33	6.33	4.67	5.33	5.00
Crude Protein	35.41	36.11	36.79	36.49	37.01	37.35	35.01	35.78	36.03
Lipid	10.00	12.01	14.02	14.02	16.01	10.01	9.90	10.01	11.79
Crude fibre	8.12	8.54	8.11	6.16	7.79	7.56	7.76	8.24	7.41
Ash	14.01	13.99	12.02	16.36	16.01	14.01	16.05	15.94	17.94

Key: Y^0M= Mature okra without yeast, Y^0I= Immature okra without yeast, Y^0S= Starch without yeast, Y^5M= Mature okra with 5% yeast, Y^5I= Immature okra with 5% yeast, Y^5S= Starch with 5% yeast, $Y^{10}M$=Mature okra with 10% yeast, $Y^{10}I$= Immature okra with 10% yeast, $Y^{10}S$ = Starch with 10% yeast.

*Vitamin/Mineral premix – A Hi-Nutrients® International Ltd product.

RESULTS AND DISCUSSION

The result on Table 2 revealed that $Y^{10}M$ had the highest floatability (360.80±15.59 seconds) while Y^0S had the least floatability (20.67±0.33 seconds). Y^0M had significantly lower ($p<0.05$) thickness swelling than the other two feed containing 0% yeast. The water stability of Y^0S was significantly lower ($p<0.05$) than that of other feeds containing 0% yeast ($p<0.05$). Though there was no significant difference between Y^0M and Y^0I but Y^0M was slightly higher than Y^0I. At 5% yeast inclusion level, water stability of Y^5M was significantly higher ($p<0.05$) than the other two feed (Y^5I and Y^5S). At 10% yeast inclusion level, $Y^{10}M$ was significantly higher than $Y^{10}I$ and $Y^{10}S$ (Table 2). Crushing load test showed that Y^0I was the hardest of the feed containing 0% yeast while Y^0M and Y^0S had same value. Y^5I had the highest crushing load amongst diet containing 5% yeast. $Y^{10}M$ was the highest for feed containing 10% yeast

Generally, feeds bounded with either immature or mature okra had higher floatability than feeds bounded with starch at different inclusion level of yeast. This shows that the binders have effect on the floatability of the feeds. There was no significant difference ($P>0.05$) in the bulk density of the nine feeds. This result was possible because all variable and fixed ingredients were the same in all the feeds. Only the binders were different and are not weighty. The regression analysis shows that there is a weak relationship ($R= 0.17$) between floatability and bulk density. Y^0M had lower thickness swelling than the other two feed containing 0% yeast. This shows that mature okra expands easily without disintegrating into water. Y^5S thickness swelling was lower than Y^5M and Y^5I at 5% inclusion level.

With increase in the yeast inclusion, there was an increase in water absorption index and thickness swelling. $Y^{10}M$ had the highest thickness swelling for the nine feed while Y^0M had the least thickness swelling. Higher thickness swelling means that the feed had more ability to absorb water and stay glued together. Correlation analysis showed a relationship of 0.68 between floatability and thickness swelling. Highest water absorption index was recorded in Y^0S and least in $Y^{10}I$. High water absorption index and high bulk density may have resulted in the low floatability of Y^0S. This is in line with the report of Adeparusi and Famurewa, (2011). The water absorption index in this study when compared to the works of Momoh *et al.* (2016) had a better result.
The nutrient retention shows that there was higher retention of protein after soaking in water for 30 minutes compare to the lipid in all the diets (figure 1). Y^0M had the highest protein retention of 94.05% while $Y^{10}S$ had the lowest retention of protein (81.54%). $Y^{10}M$ had the lowest lipid retention of 42.85% while $Y^{10}I$ had the highest lipid retention of 79.92%. This might have been due to the protein content of the binder. The values for lipid retention were lower to those in the work of Falayi *et al.,* (2004), were diets retained 95.1 to 99.0% lipid.

Table 2: Physical Characteristics of Experimental Feeds

Parameters	Y^0M	Y^5M	$Y^{10}M$	Y^0I	Y^5I	$Y^{10}I$	Y^0S	Y^5S	$Y^{10}S$
Floatability (Secs)	85.40±0.20c	206.40±1.80e	360.80±15.59g	143.4±1.80d	208.4±1.60e	316.80±3.00f	20.67±0.33a	36.00±1.00ab	42.00±1.00b
Bulk Density (g per cm³)	0.004±0.00	0.004±0.00	0.004 ±0.00	0.004±0.00	0.004±0.00	0.004±0.00	0.012±0.00	0.004±0.00	0.004±0.00
Water Absorption Index (g)	0.35±0.01ab	0.33±0.00a	0.41±0.01b	0.55±0.05c	0.33±0.00a	0.28±0.03a	1.01±0.01d	0.34±0.01a	0.30±0.03a
Thickness Strength (mm)	0.33±0.00a	0.66±0.00c	0.85±0.01e	0.50±0.01b	0.67±0.01c	0.71±0.00d	0.51±0.01b	0.51±0.01b	0.71±0.00d
Water Stability (%)	85.20±0.10b	88.10±0.12d	88.90±0.46e	85.17±0.08b	87.13±0.12c	88.23±0.08d	83.53±0.08a	87.17±0.09c	88.10±0.12d
Crushing Load Test (N)	71.40±0.00	95.20±0.00	119.00 ± 0.00	95.20±0.00	119.00±0.00	71.40 ± 0.00	71.40±0.00	71.40±0.00	71.40±0.00

Mean ±S.E. with different superscript are significantly different from each other (P<0.05)

In the three groups of feed, mature okra bounded feed had the highest water stability. This might be due to the high viscosity of the mature okra in cold water. This is because the viscosity of mature okra is higher than some binders used as feed binder including starch. The most water stable diet is $Y^{10}M$ while Y^0S was the least stable feed in water. The range of the water stability result (83.53 to 88.90) was similar to the result of Falayi et al, (2004), for cassava tuber starch and wheat grain starch. Also, the result of water stability has some similar results with the works of Momoh *et al.* (2016). Solomon *et al.,* (2011), reported a water stability range of between 40-95% for their experiment. The binders used in their experiment were corn, wheat, millet, guinean corn, and cassava tuber starch at 12% inclusion with yeast as an additive for buoyancy while this study make use of okra and starch at 5% inclusion level. Water stability and crushing load had a regression coefficient of 0.97 which shows a strong relationship between the parameters.
The result for crushing load test shows that good water stability does not necessarily translate to hardiness. A diet may be stable in water but may not be able to withstand load before crushing. The hardness might have resulted from the proper gumming of the particles by the binder. The crushing load test shows that feed $Y^{10}M$ can withstand handling processes such has bagging and transporting.

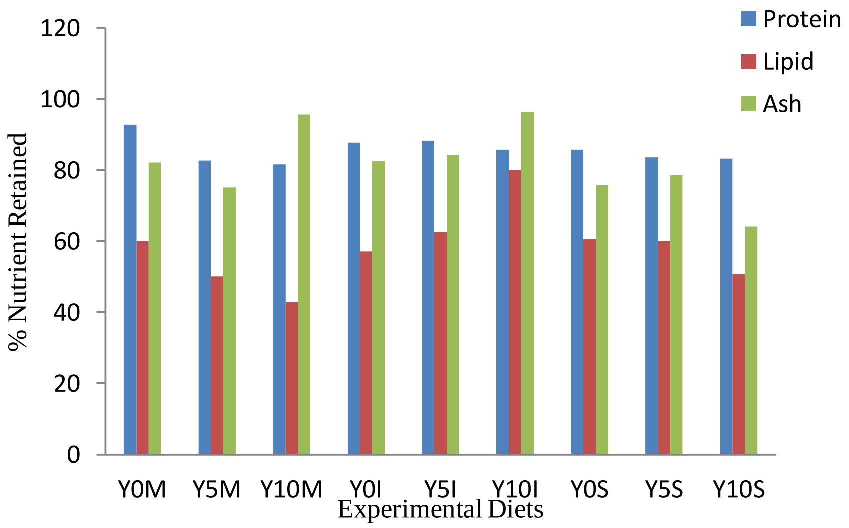

Fig 1: Nutrient retention (%) of experimental feed

CONCLUSIONS AND RECOMMENDATIONS

Results from the study shows that ingredients used binders in feed formulation have effect on the physical characteristics of feed pellets. The use of mature okra as binder with 10% yeast gave highest floatation time and thickness strength. The high water stability will reduce leaching of nutrients to the environment resulting in higher fish production and lower feed cost since higher percentage of the nutrient will be available for fish. It will also prevent feed wastage and is therefore recommended for use in fish feed. Mature okra that is consider as waste on farm and in market can be made use of as binder in the production of fish pellet feed. This will help in converting waste to products useful in fish feed production.

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