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## Utilization of unconventional dietary energy source of local origin in aquaculture: Impact of replacement of dietary corn with tapioca on physical properties of extruded fish feed

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### Abstract

A study was conducted to investigate the effects of graded replacement of dietary corn with tapioca starch on the physical properties of extruded fish feeds. Five (5) isonitrogenous fish feeds of 28% crude protein were formulated utilizing locally available feed ingredients wherein tapioca starch replaced corn starch at 0% (D1-Control), 25% (D2), 50% (D3), 75% (D4) and 100% (D5). The proximate compositions of the ingredients were determined and the physical properties of different extruded fish feeds were investigated. The results showed that replacement of corn starch with tapioca starch had significant effects ( $p<0.05$ ) on physical properties of the extruded feeds. The significantly highest bulk density (BD) was observed for D1 (541.5 g L<sup>-1</sup>) while the lowest BD observed in D5 (454.8 g L<sup>-1</sup>). Significant differences ( $p<0.05$ ) in water stability (WS) of extruded fish feed were evident both after 15 minutes and 30 minutes exposure to water with the highest values for D1 (89%) and D2 (84.9%), respectively. On the other hand, the lowest values were noted for D5 (85.9% and 80.5%) after both 15 minutes and 30 minutes of exposure to water, respectively. The result of the study revealed that replacement of corn with tapioca starch led to a significant but marginal reduction in BD and WS. Further, up to 50% replacement level no reduction in WS was noted.

**Keywords:** Extruded feed, physical properties, starch, tapioca

### 1. Introduction

Globally, aquaculture is the fastest growing food producing sector and is likely to continue to grow in light of increasing demand of fish vis-a-vis stagnating to declining fish production from capture fisheries. Notably, aquaculture also makes substantial contributions in socioeconomics of numerous nations including India with respect to increasing rural income, improving food and nutritional security, and creating greater employment opportunities [14]. Further, it is notable that the growth in production has been faster for fed-species than for non-fed species indicating that adoption of feed-based aquaculture as the major driver of aquaculture production growth [12]. It is remarkable that more than two-third of world food fish production through aquaculture now comes from feed-based culture systems [12]. The adoption of feed-based aquaculture is likely to further intensify for enhancing the productivity considering the declining land and water resources. Feeding fish in intensified aquaculture systems adequately to meet growth, reproductive and health requirements of fish in the era of finite resources is indeed a great challenge. Therefore, the sustainability of growth of aquaculture is intricately linked with the availability of sufficient feed ingredients. On the other hand, feed also represents the 50-70% of operating cost of the fish production [19]. The situation is increasingly becoming more and more serious considering that prices of traditional feed ingredients have been increasing at faster rates than that of fishes leading to remarkable reductions in the profit margins of fish farmers. These issues have set off the quest for less expensive, alternative, locally accessible feedstuff [22].

The local fish production of the north-eastern region is grossly inadequate to meet the local demand. Consequently, large quantities of fishes are imported into the region from not only other parts of India but also neighbouring countries including Bangladesh and Myanmar on daily basis. Therefore, it has been a priority for state governments to increase the local aquaculture production primarily through adoption of feed-based culture practices.

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But, the region is also deficit in traditional food and feed grains including maize, mustard oil cake, soybean, other beans, wheat, fish meal, etc. Consequently, most of these ingredients and/or fish feeds are also imported into the region from distant other parts of the country making the feed even more expensive due to high transportation cost. Further, the supply is also prone to anthropogenic and natural disruptions besides of increasing the carbon foot print of the fish production in north-eastern region. Therefore, looking for alternative unconventional feed ingredients of local origin is crucial for sustainable aquaculture in the region<sup>[17]</sup>.

In this regard it is notable that the region is endowed with a large variety of tapioca or Cassava (family Euphorbiaceae) which grows throughout the year without much care and has remarkably high starch productivity potential. The plants are widely cultivated in tropical and sub-tropical regions of the world and are the third largest source of food carbohydrates in the tropics, after rice and maize. The tapioca plant gives the third highest yield of carbohydrates per cultivated area among crop plants, after sugarcane and sugar beets<sup>[19]</sup>. Hence, it can be one of the potential local energy sources for fish feeds in the region in lieu of non-local and more expensive corn, wheat and even rice.

In the present paper, we utilized tapioca meal to replace dietary corn meal, a traditional starch source in fish feed. Although, tapioca has been utilized as dietary starch source for fishes viz. *Clarias gariepinus*<sup>[1]</sup> and *Oreochromis niolticus*<sup>[25]</sup>. There is general dearth of information pertaining to impact of its inclusion on physical properties of diets such as bulk density, sinking velocity and water stability. Considering that physical properties are important determinants of feed utilization efficiency in aquaculture, objectives of the present study was to determine the impact of graded replacement of dietary corn with tapioca on physical properties of extruded fish feed.

## 2. Materials and Methods

### 2.1 Collection and processing of Tapioca tubers, wolffia meal and other ingredients

The tapioca tubers were procured from ICAR research complex Lembucher, Tripura (W), India in a single lot to avoid variability in biochemical compositions. The tubers were manually cleaned to remove soil and sliced into chips of 1-3 mm thickness and oven dried for 24-36 hours at 65-70°C. Dried chips of Tapioca tubers were fine grounded using a hammer mill grinder (1 mm sieve) and were packed in a polythene bag until used. On the other hand, fresh *Wolffia globossa* was harvested from a culture pond and dried in oven for 40-48 hours at 65-70°C and grounded using the same hammer mill grinder. Rest of other traditional ingredients including corn, soybean meal, mustard oil cake and dry fish were procured from local market and were dried if required and grounded using the same hammer mill.

### 2.2 Proximate composition of ingredients

The proximate compositions of all the ingredients were analysed using standard methods<sup>[2]</sup>. In summary, the moisture content was determined by oven drying a pre-weighed sample at 105°C to constant weight. The ash content was determined by incinerating the dried samples in a Muffle furnace at 550°C for 5-6 hours. The percentage of protein ( $N \times 6.25$ ) was estimated in Kjeltec system (Tecator™ Digestor and Kjeltec™ 8400 Analyzer Unit, FOSS) after digestion with sulphuric acid and catalyst ( $K_2SO_4$  and  $CuSO_4$  in 9:1). The percentage of lipid was determined by using the petroleum

ether method in Soxtec system (Labtec ST310 Solvent, FOSS) and crude fibre was estimated in de-fattened sample using 1.25% Sulphuric acid and 1.25% Sodium hydroxide alkali digestion in Fibre tech system (Fibraplus – FES 06 AS, PELICAN). The proximate compositions of different feed ingredients are presented in Table 1.

### 2.3 Feed formulation and production

Five (D1-D5) extruded isonitrogenous experimental fish feeds (crude protein, 28%) were formulated using locally available ingredients namely fish meal, mustard oil cake, wolffia meal, rice bran, soybean meal, broken corn and tapioca using MS Excel. Tapioca starch (TS)-free fish feed where corn starch (CS) was included at a level of 30% (W/W) was treated as control (D1). In the rest other four experimental isonitrogenous fish feeds, the tapioca starch replaced corn starch at levels of 25% (D2; CS 22.5% and TS 7.5%), 50% (D3; CS 15% and TS 15%), 75% (D4; CS 7.5% and TS 22.5%) and 100% (D5; CS 0% and TS 30%). Thus, the highest inclusion level for both corn (in D1) and tapioca (in D5) were 30% (W/W). The ingredient compositions of different experimental fish feeds are presented in Table 2.

The sinking diets were produced using a twin-screw extruder (LT 70, Shandong light M & E Co. Ltd. China). The required quantities of grounded ingredients were weighed and carefully mixed in a horizontal mechanical mixture in a batch of 20 kg where water was added at a level of about 20% (volume/weight). The dough of mixed ingredients were transferred to pre-conditioning chamber through a disc-conveyor and then into the extruder and were passed through 1 mm die. The production conditions including temperature in the extruder (150° C, 160° C and 170° C of zone I, zone II and zone III respectively), main motor speed (29 rpm), feeder speed (25 rpm), cutter speed (23 rpm), die and diameter size (1 mm) were kept same for all fish feeds. The extruded pellets (diameter- 1 mm) were transferred into a conveyor belt drier (120° C) through air conveyor as soon as these came out of the extruder for drying to a moisture level of 6-8%.

### 2.4 Determination of physical properties

Physical properties of extruded fish feeds were analysed by measuring bulk density (BD), sinking velocity (SV) and water stability (WS) of different experimental fish feeds in triplicates. The BD ( $g L^{-1}$ ) was measured to determine the property which describes the weight of particular feed per unit volume. Each experimental fish feed was poured into a 1000 ml measuring cylinder in triplicates and weight of contained fish feed was noted in kg up to 3 decimal precision<sup>[24]</sup>. The SV was measured by using 1000 ml measuring cylinder filled with distilled water in which ten uniform sized pellets of each experimental fish feed were dropped and the time taken for each piece to reach the bottom (T) were recorded. The height of the water column (H) was noted and sinking velocity was determined as “ $SV (m s^{-1}) = H/T$ ”. The WS test of experimental fish feeds were determined by placing 4 g of different experimental fish feeds that were dried to constant weights (105°C) in small bags of nylon sieve materials of 0.1 mm mesh in triplicates. After tieing the bags from top the bags with feeds were immersed in 100 ml distilled water for 15 minutes and 30 minutes. The bags were removed and oven dried to constant weight by drying at 105°C for 24 hours at start as well as again dried in oven and weighed to obtain dry matter weight<sup>[16]</sup>.

a. Bulk Density ( $g L^{-1}$ ) = Weight of experimental diet (g) / Volume (l)

- b. Sinking Velocity ( $\text{m s}^{-1}$ ) = Height of the water column (m) / Time taken for each piece to reach the bottom (s)
- c. Water Stability (%) = (Weight of retained whole pellets / Initial total weight of pellets)  $\times 100$

## 2.5 Statistical analysis

The statistical analyses were performed using MS excel and Statistical Package for Social Sciences (SPSS, version 16.0 for windows). One-way analysis of variance (ANOVA) was performed to determine the differences between the mean values of different treatments. Ranking of differences if any in means were obtained by Duncan's New Multiple Range test at  $p<0.05$  level [8].

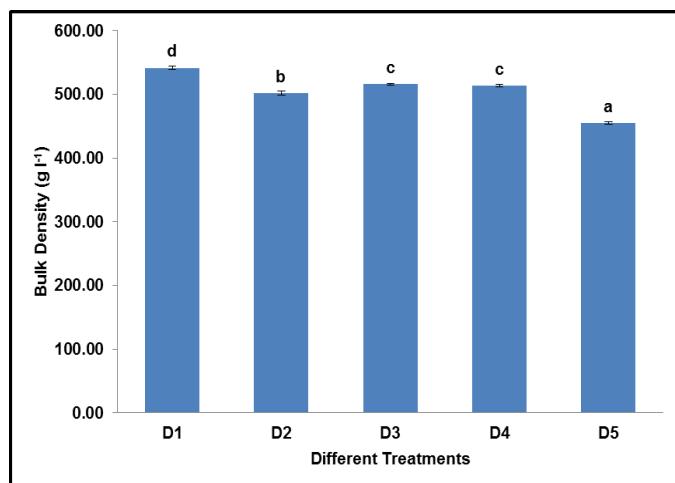
## 3. Results

As may be apparent, tapioca and corn had remarkably high nitrogen-free extract (85.6-88.2%) and crude fibre (2.34-5.12%) and lower crude protein (3.03-8.35%) and crude lipid (0.78-2.30%) contents in comparison to fish meal, mustard oil cake and soybean meal (Table 1). Notably, the crude fibre and nitrogen-free extract contents in tapioca were somewhat higher than those of corn while crude protein and crude lipid contents were relatively lower. The respective mixtures of ingredients (20 kg) corresponding to isonitrogenous (28%) test diets D1, D2, D3, D4 and D5 containing graded levels of tapioca starch were extruded through 1 mm diameter die in a twin screw-extruder under exactly the same manufacturing conditions. The pellets were directly transferred to a conveyor belt-dryer through air conveyor-cum-hoister and were collected after gradual cooling to room temperature in double drum roller. In general, the pellets of all experimental feeds were of cylindrical shape, similar size and resilient.

Some of the physical properties including BD, SV and WS after 15 minute and 30 minute soaking of different experimental fish feeds are shown in Figure 1-4. In general, the results showed that replacement of corn starch with tapioca starch had significant effects ( $p<0.05$ ) on physical properties of the extruded fish feeds.

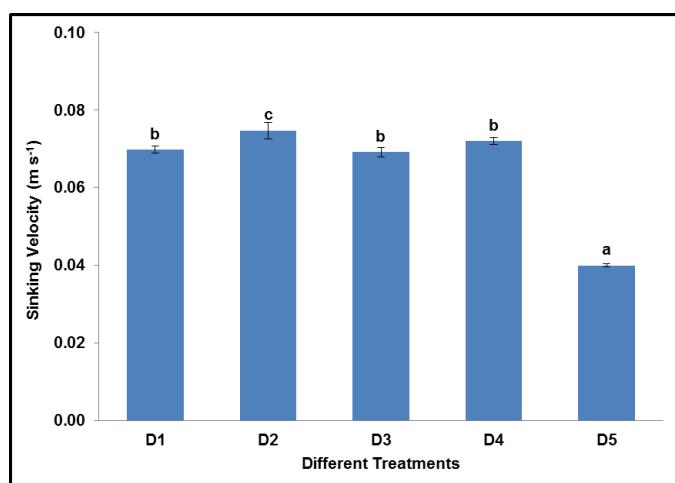
Specifically, the BD was significantly ( $p<0.05$ ) highest (541.5  $\text{g L}^{-1}$ ) for D1 viz. control fish feed (tapioca-free) while it was the lowest (454.8  $\text{g L}^{-1}$ ) for D5 (corn starch-free). In general, BDs of experimental feeds decreased concomitant to replacement of corn starch with tapioca starch, it indicated that the feeds with tapioca as the main starch source had greater expansion in comparison to the feed with corn as the main starch source. Accordingly, SV was somewhat faster for D1 in comparison to that of D5 ( $0.04 \text{ m s}^{-1}$ ) also significantly varied ( $p<0.05$ ) among treatments having highest for D2 (0.07  $\text{m s}^{-1}$ ) while the lowest for D5 (0.04  $\text{m s}^{-1}$ ).

More notably, there were significant differences ( $P<0.05$ ) in WS of extruded fish feeds both after 15 minutes (Figure 3) and after 30 minutes (Figure 4) exposure to water. The significantly high ( $p<0.05$ ) WS after 15 minutes and 30 minutes were observed for D1 (89%) and D2 (84.9%), respectively while significantly lowest value at both durations were observed for D5 with values of 85.9% and 80.5%, respectively indicating reduction in water stability due to replacement of corn starch with tapioca starch. However, no significant differences ( $p>0.05$ ) in WS were apparent among D1, D2 and D3 indicating that up to 50% replacement of corn starch with tapioca starch did not have any adverse impact on water stability.



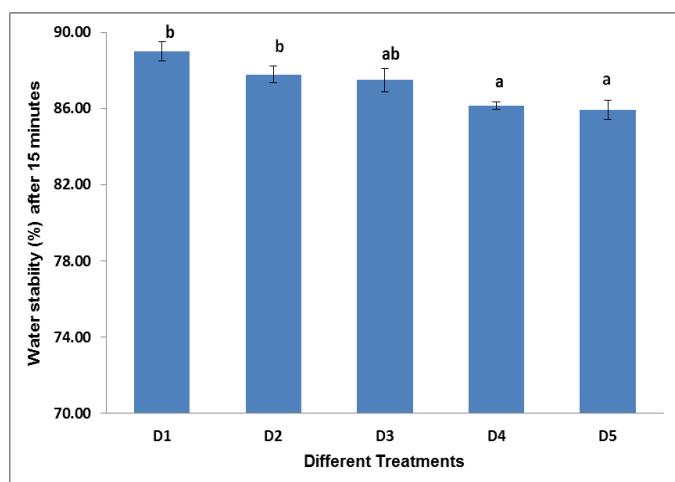
**Fig 1:** Mean bulk density ( $\pm 1\text{SE}$ ) of different experimental fish feeds

The mean values with different alphabetical superscript within a row for parameters are significantly different ( $p<0.05$ ).



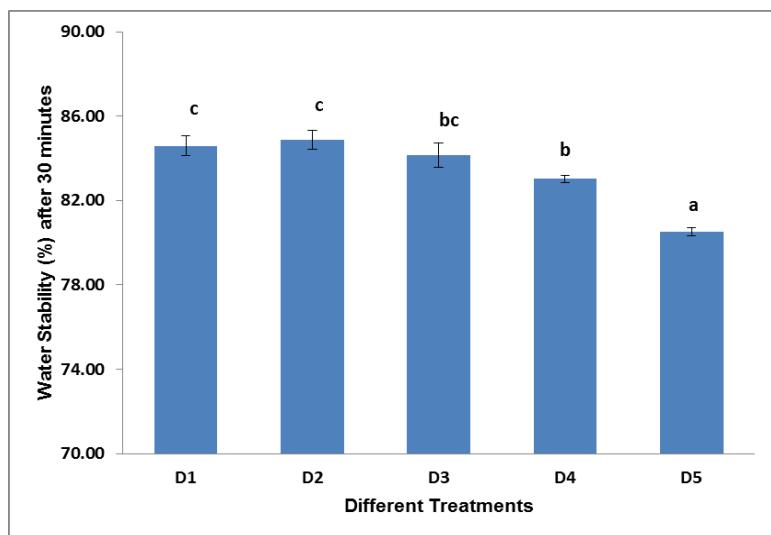
**Fig 2:** Mean sinking velocity ( $\pm 1\text{SE}$ ) of different experimental fish feeds

The mean values with different alphabetical superscript within a row for parameters are significantly different ( $p<0.05$ ).



**Fig 3:** Mean water stability ( $\pm 1\text{SE}$ ) of different experimental fish feeds after 15 minute exposure to water

The mean values with different alphabetical superscript within a row for parameters are significantly different ( $p<0.05$ ).



**Fig 4:** Mean water stability ( $\pm 1\text{SE}$ ) of different experimental fish feeds after 30 minute exposure to water  
The mean values with different alphabetical superscript within a row for parameters are significantly different ( $p<0.05$ ).

**Table 1:** Proximate composition of feed ingredients on dry matter basis

Feed Ingredient	Ash (%)	Lipid (%)	Protein (%)	Fibre (%)	NFE (%)
Fish Meal	$28.06 \pm 0.33$	$5.77 \pm 0.06$	$54.86 \pm 0.1$	$0.55 \pm 0.08$	$10.76 \pm 0.50$
Mustard Oil Cake	$9.92 \pm 0.05$	$12.76 \pm 0.03$	$31.44 \pm 0.21$	$7.70 \pm 0.02$	$38.81 \pm 0.14$
Wolffia Meal	$13.59 \pm 0.07$	$2.54 \pm 0.08$	$29.03 \pm 0.16$	$11.76 \pm 0.31$	$43.08 \pm 0.29$
Soybean Meal	$6.66 \pm 0.02$	$0.20 \pm 0.01$	$46.97 \pm 0.19$	$4.26 \pm 0.17$	$41.91 \pm 0.28$
Rice Bran	$10.24 \pm 0.37$	$3.44 \pm 0.18$	$7.09 \pm 0.11$	$21.01 \pm 0.33$	$58.22 \pm 0.71$
Broken Corn	$1.44 \pm 0.04$	$2.30 \pm 0.03$	$8.35 \pm 0.16$	$2.34 \pm 0.27$	$85.57 \pm 0.31$
Tapioca Flour	$2.84 \pm 0.05$	$0.78 \pm 0.04$	$3.03 \pm 0.03$	$5.12 \pm 0.22$	$88.23 \pm 0.32$

Data shown in the table are Mean  $\pm$  SE, n = 3.

**Table 2:** Ingredient composition (%) of the different experimental fish feeds

Feed Ingredient	D1	D2	D3	D4	D5
Fish Meal	20.0	20.0	20.0	20.0	20.0
Mustard Oil Cake	15.0	15.0	15.0	15.0	15.0
Wolffia Meal	10.0	10.0	10.0	10.0	10.0
Soybean Meal	15.0	15.5	16.5	17.5	18.0
Rice Bran	6.0	5.5	4.5	3.5	3.0
Broken Corn	30.0	22.5	15.0	7.5	0.0
Tapioca Flour	0.0	7.5	15.0	22.5	30.0
Vegetable Oil	2.0	2.0	2.0	2.0	2.0
Vitamins & Minerals mixture	2.0	2.0	2.0	2.0	2.0

#### 4. Discussion

Carbohydrates represent the cheapest source of dietary energy, and hence, hence are included in diets matching to the maximum tolerance level of the target fish so as to spare more expensive dietary proteins from being utilized for energy production. Accordingly, dietary carbohydrates are usually quantitatively the largest constituent of diet (40-55%; W/W) particularly in feeds for carps, the most dominant group of fishes with respect to not only world aquaculture but also Indian Aquaculture. The rice bran, corn, and wheat are the conventional carbohydrate sources in India. None of these items are produced locally in sufficient quantity. The tapioca has been selected as one of the local energy sources primarily because it is endemic to the region, is pretty hardy, predominantly grown in the whole north eastern region without much care and have high yield potential. In fact, the tapioca plant has been known to have the third highest yield of carbohydrates per cultivated area among crop plants, after

sugarcane and sugar beets [9]. Besides, tapioca roots contain significant amounts of calcium, phosphorus and vitamin C. Further, the tapioca tubers have been reported to have favourable impacts on nutrient retention, fish growth, feed utilization and economic utilization indices in African countries [11]. It has been reported that tapioca flour could completely replace yellow maize in the fish feed for African catfish [1]. Due to these reasons, despite of low crude protein levels and short shelf-life in fresh conditions may have practical utility for sustainable aquaculture development and/or productivity enhancement.

It is also important to note that the chosen method of fish feed manufacturing *viz.* extrusion is the most versatile fish feed manufacturing method considering multiple positive impacts on feed and its utilization efficiency. Various reactions take place during extrusion including thermal treatment, gelatinization, protein denaturation, hydration, texture alteration, partial dehydration, and destruction of microorganisms and other toxic compounds during extrusion cooking [14]. In addition, extrusion has been known to impart greater positive impact on water stability and digestibility due to gelatinization than other forms of processing [5, 7, 20]. The extrusion is also considered environmentally friendly as it improves conservation and the compactness of feed in the aquatic environment [4].

The physical properties of fish feeds are the net results of feed ingredients and manufacturing methods and conditions, and have important implications for feed consumption and utilization efficiency [21]. The significant attributes which affects the quality of fish feed include bulk density, water absorption and solubility, hardness or softness, colour, pellet size, shape, resiliency, buoyancy and chewiness [15]. In the

present study, the physical properties of extruded fish feed containing varying levels of replacements of corn with tapioca as a local starch source in supplementary carp feed. BD, SV and WS showed significant differences ( $p<0.05$ ) among treatments. The BD affects transportation and storage costs. In the present study, the highest BD was observed for D1 (541.5 g L<sup>-1</sup>) while the lowest bulk density observed in D5 (454.8 g L<sup>-1</sup>). SV is an important extrudate property which determines the stability of the extrudates in water and is closely related to the absorption of water throughout feeding at the water surface [6]. In the present study, SV also significantly varied ( $p<0.05$ ) among treatments having highest for T2 (0.07 m s<sup>-1</sup>) while lowest for T5 (0.04 m s<sup>-1</sup>). However, there was no significant difference ( $p>0.05$ ) between D1, D3 and D4 with regards to SV of experimental fish feeds. The WS of fish feed determines the overall performance of the feed [3]. The disintegration of feed pellets due to nutrient leaching leads to non-availability to the animals. The water stability of pellets can be greatly improved through proper selection of feed ingredients, processing techniques and use of proper processing equipment [16]. In the present study, WS of fish feed increased up to 50% replacement of corn starch with tapioca starch. The comparative work on binding capacity of some synthetic and natural binders reported tapioca as a better feed binder than corn starch [13]. Our finding is also corroborated by the findings of earlier works reporting increased water stability for feeds made using tapioca starch compared to feeds using corn starch [23]. However, a decrease in water stability as noticed in this experiment at higher inclusion levels could be due to loss of mixing property, lesser amylose-lipid interaction necessary for pasting of ingredients in feed, lowered gluten content of feed [18]. The similar findings had also been reported as the level of tapioca starch was progressively increased and they attributed the same to early gelatinisation, it reduced retro-gradation (amylose-lipid interaction) of the ingredients leading to somewhat lower water stability [10].

## 5. Conclusions

The replacement of corn with tapioca starch as a feedstuff for had significant impacts on physical properties of extruded fish feeds viz. bulk density, sinking velocity and water stability. Nevertheless, up to 50% replacement level no or only mild differences in physical properties were noted. Even at 100% replacement level the differences though significant were not dramatic. Accordingly, our conclusion is that tapioca could completely replace corn at the inclusion level of 30% in the supplementary diet of *O. belangeri*.

## 6. Acknowledgements

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