

**EFFECT OF STARCH SOURCE, SCREW CONFIGURATION
AND STEAM INJECTION ON PHYSICAL QUALITY AND
COLOR DEVELOPMENT OF EXTRUDED FISH FEED**

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Abstract

The study investigated the effect of starch sources, screw configuration, and steam injection on physical quality of extruded fish feed. Pellet durability, water stability and hardness were measured and analyzed. Four starch sources; pregelatinized potato starch, potato starch, whole wheat and wheat starch were used in the production of extruded fish feed. Three screw configuration types; polygon-2L, polygon-LR, and LRLR were used during production of feed. Similarly, steam injection and no steam injection in barrel were used for the production of the extruded fish feed. Contrast, brightness and entropy were used to analyze differences in color. Four diets; Pregelatinized potato starch, potato starch, whole wheat and wheat starch showed varied results. In general, potato starch diets gave the highest pellet durability, hardness and water stability than the wheat starch diets. Pregelatinized potato gave highest pellet durability and water stability. Hardness (length, width and breaking force) was also higher for pregelatinized potato starch. Potato starch gave the highest hardness. Screw configuration LRLR gave the highest pellet durability and Polygon-LR had the lowest. Screw configuration Polygon-2L and LRLR gave higher breaking force than Polygon-LR. There was no difference in the effect of screw configuration on length and diameter. No steam injection in the barrel gave a higher pellet durability and water stability. Steam pressure did not affect length, diameter and breaking force of the pellet.

Wheat diets gave brighter pellets than the potato starch diet, indicating that extent of maillard and caremelization differed for different diet compositions. Screw configuration and steam pressure did not show significant effect on contrast and entropy of the picture. Whole wheat gave lowest contrast and highest entropy. Wheat starch gave highest contrast and lowest entropy. Overall results indicated that starch source and processing parameters affected physical quality of pellets. Color analysis showed that starch sources used in feed production had influenced color development during extrusion, indicating possible nonenzymatic browning of the feed.

Key words: extrusion, starch sources, physical quality of feed, color development

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1. Introduction

Manufacture and formulation of fish diet evolved tremendously during 20th century. Fish feed production was confined to extensive system, the concept developed was based on helping natural production of food on pond by fertilization and supplying extra food if needed. It's easy to formulate a diet fulfilling all the nutritional requirement of fish. However, when technical or physical quality of fish feed is considered, the processing becomes complicated. Critical considerations have to be made about ingredient price and availability, pelletability, pellet handling requirement and storage. Hence, fish diet production is a compromise between the nutritional quality and physical quality of fish feed (Hardy and Barrows, 2002). Today fish feed is commonly produced by extrusion systems. Extrusion is a process by which moistened, expansible, starch and/or proteinous materials are plasticized and cooked in a tube by a combination of moisture, pressure, temperature and mechanical shear (Kearns, 1999). Extrusion of feed is a combination of heat; shearing and pressure build up in the presence of water. The superiority of extrusion over conventional steam conditioning and ring die pelleting include (Kiang, 1999): better formulation flexibility, higher versatility in physical size and shape, greater water stability and physical integrity, and greater fat absorption capacity. According to (Smith, 1971) principal advantages of extrusion over traditional food and feed production methods includes the following; adaptability; product characteristics; energy efficiency; low cost; new foods; high productivity and automated control; high product quality; no effluent; process scale-up and use as a continuous reactor. Extrusion process enhances the feed efficiency and prolongs the shelf life of the feed.

Image processing is getting popular for food quality inspection (Du and Sun, 2004). Smolarz, Van Hecke, and Bouvier, (1989) reported the use of image processing techniques to characterize the internal structure of expanded products. Several authors have used color as a quality control indicator of processes because brown pigments increases as browning and caramelization reaction proceeds (Moss and Otten, 1989). Dan et al., (2007) used image texture analysis methods to characterize the spatiotemporal stress distribution during food fracture.

1.1 General description of Extrusion line for fish feed production

Series of steps follows an Extrusion process. General steps in extrusion are briefly described as follows (ForTek, UMB) : a. Receiving of ingredients b. Storage c. Batching system d. grinding e. Mixing and conditioning f. Extrusion g. Drying h. vacuum coating i. cooling and packaging

A. Receiving of ingredients: Raw materials are received through different means according to the available transportation system. Ships, buses, animal driven carts can be used. Sizing of

receiving units is important for improved safety and for cost and logistic reason. It should be large enough to handle all deliveries in a fixed time. Advanced feed manufacturing industries have well developed receiving units.

- B. Storage: Different raw materials are stored in different silos. The capacity of silo generally varies. Main ingredients are stored in bigger silos than the minor ingredients.
- C. Batching system: Capacity of batching system varies from plant to plant. Batching system consists of scales for both major and minor ingredients. Typically, batching system has series of ingredients bins, feeder screws and load cells.
- D. Grinding: Hammer mill and roller mill are used to reduce particle size. Particle size reduction improves mixability, increases surface area, which can enhance protein denaturation and gelatinization contributing to pellet quality.
- E. Mixing: Three basic types of mixers have been used in animal feed manufacturing; vertical screws, rotary drums, and horizontal. Now, twin shaft mixers are widely used in feed manufacturing purposes.
- F. Conditioning: It includes a preconditioning chamber when feed is to be extruded. In this chamber feed ingredients are uniformly moistened and heated by contact with water or steam under constant mixing, before entering the extruder. The purpose of preconditioning is to add moisture and heat to soften feed particles which helps in gelatinization of starches and protein denaturation. Temperature of 90-95°C may occur in the preconditioner due to the addition of steam and water.
- G. Extrusion: Extruder barrel consists of extruder screws, heads and shear locks. It consists of die at the discharge end of the extruder. There are three principal different regions on the screw: feed zone, kneading zone and final cooking zone. Retention time in the extruder in general is 15-20 seconds. Higher temperature and pressure is developed inside the barrel and forces the mash through the die holes at the end of the extruder barrel. Combination of heat (125-150°C), moisture (20-24%) and pressure causes gelatinization of starch and protein denaturation transforming particles into dough before pellets are shaped in the die.
- H. Drying: To decrease susceptibility to deterioration during storage, pellets have to be dried to a moisture level below 10%. Higher moisture level increases the risk of bacterial and mould growth. Hot air drying is common method in feed production industry.
- I. Vacuum coating: Vacuum coating is used to increase fat content.
- J. Cooling and packaging: Extrudate is cooled prior to packaging. During packaging there should not be temperature difference of more than 10°C between ambient environment and feed, to avoid condensation inside the bag.

1.2 Classification of extruders

In the feed manufacturing technology, single and twin-screw extrusion is commonly used and will therefore be described. Because extruders are used for different purposes, there is different classification. Classification schemes based on shear, heat generation and numbers of screws are explained below.

Classification based on shear (Harper, 1981)

- Cold forming extruders: It is used to produce moist aquatic feeds. It usually requires binders to hold the final shaped pellet together. It is a low-shear machine with smooth barrels, deep flights, and low screw speeds.
- High-pressure forming extruders: It is a low-shear machine with grooved barrels and compressing screws, typically used to extrude pre-gelatinized cereal.
- Low-shear cooking extruders: It is a moderate-shear machine with high compression screws and grooved barrels which enhances mixing.
- Collet extruders: It is high-shear machines with grooved barrels and screws with multiple shallow flights that are used for manufacturing puffed snacks from defatted corn grits.
- High shear cooking extruders: It is a high-shear machine, with screws for changing flight depth and/or screw pitch, that have the ability to achieve high compression ratios, high temperatures, and various degrees of puffing.

Classification based on heat generation (Harper, 1981):

- Adiabatic (autogenous) extruders produce heat by friction (viscous dissipation of mechanical energy input).
- Isothermal extruders work at a constant product temperature throughout the length of the barrel and are used mainly for forming.
- Polytropic extruders have provisions of adding or removing heat as required during the process.

Classification based on number of screws:

Twin screw extruders; they are used in wet extrusion. They have limited use in animal feed and pet food industry. They are more used for aquatic feed production, when specialized feed is required (Enterline et al., 2005).

- **Counterrotating twin-screw extruders:** They are not widely used in food industry but they work as excellent conveyors. They are good in processing non-viscous materials which requires low speeds and long residence time.

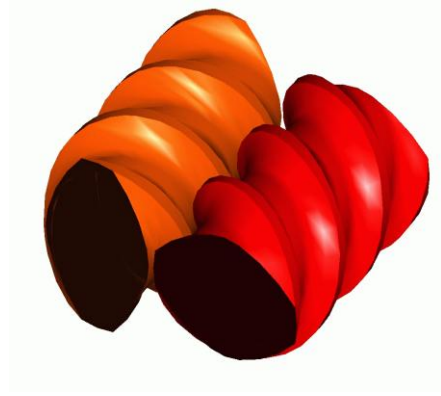


Fig. 1. Geometry of the intermeshing screw in counter rotating twin screw extruder

- **Corotating twin screw extruders:** There are two types of extruders. One is corotating intermeshing screw and another is corotating nonintermeshing screw. In intermeshing screw type the flight of one screw engages or penetrates the channels of the other screw. It has a positive pumping action, efficient mixing, and self-cleaning characteristics. While in nonintermeshing screw type the screws do not engage each other's threads, allowing one screw to turn without interfering with the other. They have a higher capacity than the counterrotating type.

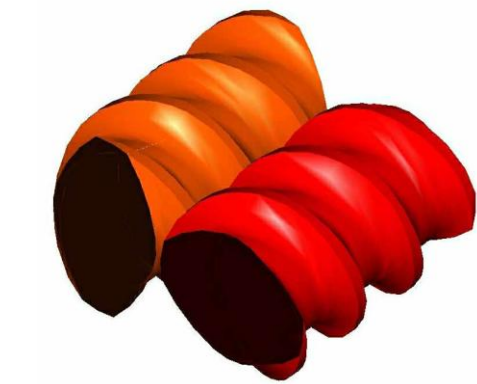


Fig. 2. Geometry of the intermeshing screw in co-rotating extruder

- **Single screw extruders;**
Single-screw extruders are based on single screw in the barrel of the extruder. It transports and shapes ingredients into a uniform food product by forcing the ingredient mix through a shaped die to produce the uniform shape. Single-screw extruders consist of three zones:

feeding zone, kneading zone, cooling zone. As compared to twin-screw extruders, single-screw extruders have poor mixing ability and therefore materials should be pre-mixed or pre-conditioned (foodtechcompare.com).

1.3 Conveying and transformation in the barrel

Extrusion process is a combined unit operations which includes feeding, mixing, cooking, kneading, shearing, shaping and forming. Extruder barrel consists of three main zones (Bishop Andrew, 2005). Even though extruders are classified according to method of operation i. e. cold or hot extruders and method of construction i. e. single and twin-screw extruders, the basic operation principles are similar for all. Raw materials are fed into the barrel and the screw conveys and transforms the mash into dough. Feeding zone is the area where low-density discrete particles are transported but the flow channel of the screw is not filled completely. There is only minimal compression action in this area. Water can be added into this zone. Added water enhances viscosity, texture and heat transfer. As the food moves inside the barrel, smaller flights restrict the increase in volume and consequently the food movement is restricted. This restricted movement fills the barrel and the spaces between the screw flights so compression begins. Further down the barrel kneading process begins and converts the food into semi-solid, plasticized mass. When the feed comes into kneading zone, compression increases and also the flow channel of the screw also gets higher degree of fill. Because of higher degree of fill, pressure increases. Finally, when the flow channel fill with loose granular material and begins to compress, shear start to work. The temperature also increases due to conduction, viscous energy dissipation and discrete particles are transformed into dough mass. At the end of kneading section, feed gets compacted. Frictional heat produced by contact surface, shear and pressure as well as additional heat added will cause an increase in temperature. Now the food is passed to the section of the barrel (cooking zone) with the smallest flights, where pressure and shearing is further increased. The final cooking zone is the section with highest temperature and pressure where melting and texturizing occur. This region has also the highest shear and compression rate. And eventually it is forced through the restricted opening i. e. dies. When the food passes through the dies, expansion takes place. Various shapes like, rods, spheres, doughnuts, tubes, strips, squirls or shell can be formed depending on the die configuration (Fellows, 2000).

1.4 Nutrient requirement of fish feed

Nutrient requirement in fish changes through life-cycle. Different types of feed are used accordingly. It is essential to know the nutrient requirement of fish so complete balanced diet can be formulated according to the need of fish and resource availability. Generally, as a rule of thumb dietary requirement of a fish can be established for energy, protein and amino acids,

lipids, minerals, and vitamins. Balanced diet is essential for fish growth, reproduction and health.

It is desirable to have a balanced protein and energy in a diet. An excess or a deficit of Digestible energy (DE) can reduce growth in a fish. The diet low in energy in relation to protein, protein may be used for energy instead of growth. Energy requirement of a fish depends upon the water temperature; in higher water temperature energy demand is higher. Lipids are used to supply the diets with energy and to meet the requirement of essential fatty acids. Sources of oils includes; fish oils, plant oils, such as soybean, corn and cotton seed; animal fats and poultry fats (Takeuchi and Watanabe, 1982 in Hardy and Barrows, 2002). A recommended optimal dietary fat level is below 12% in the diets of many cyprinids. Although diets with 20% high quality lipid sources like fish oil did not produce any negative effects on the growth of common carp (Zeitler et al, 1983). The energy content in the feed varies among species. For Atlantic salmon up to 40% oil is used in the diet, while other species may have 12-18% in the feed.

Protein is required by fish for growth and reproduction. Most fish digest protein well, and the energy in them is available to the fish. Quantitative dietary protein requirement of fish varies from 25-50% of dry matter basis. Digestibility of protein from normally used ingredients is high in all fish, which mean amino acid availabilities is also high with significant effect on fish (Plakas and Katayama).

Protein ingredients can be categorized into three groups based on protein content of the ingredients. First group with 20-30% protein includes by-product of brewing and distilling industries, wheat germ meal and corn gluten feed. Next group with 30-50% includes ingredients such as oilseed meals, crab meal and dried milk products. And the final group with more than 50% protein includes the ingredients such as fish meal, blood meal, feather meal, meat and bone meal, yeast products, shrimp meal, poultry by-product meal, soy protein concentrates, wheat gluten, corn gluten meal and casein (Hardy and Barrows, 2002). Vitamin premixes are added at a level ranging from 0.5% to 4% of the diet (Gabaudan and Hardy, 2000 in Hardy and Barrows, 2002). Feed should be supplemented with elements such as P if the ingredients contain low levels and lysine and methionine should be added if plant source is used.

Fiber is a nonnutritive portion of feed ingredients. It is generally recommended that diet should not exceed 8-12% fiber for a fish. Excessive fiber content results in a dilution of nutrients and energy in the diet. Excessive fiber also negatively affects the nutrient digestibility. Fiber is added to semi purified diets to enhance binding and increase digestion efficiency (Buhler and Halver, 1961 in Hardy and Barrows, 2002).

Fish feed produced must withstand normal handling and shipping without deforming. It should also be water stable. Commonly used binders includes regular feed ingredients,

pregelatinized potato starch, bentonite, lignin sulfonate and hemicellulose extract (Hardy and barrows, 2002).

Fish feed can also be supplemented with probiotics, hormones, antimicrobial agents, antioxidants, flavorings and palatability enhancers to prevent diseases or to improve the growth performance.

1.5 Methods to analyze physical quality of pellets and extruded fish feed

Physical quality of the feed can be defined as capability of processed pellets to resist handling without creating excessive amount of fines. Different physical properties of feed are used for various species of animal. This indicates different quality standards are used. Horse feed are more fibrous in nature so their pellet quality criteria is different than fish feed. Fish feed characteristics such as sinking velocity, water absorption, and water solubility are important (Thomas and Van der Poel, 1996). Physical quality of feed pellet must have certain integrity and should not produce fines due to stress of various nature like transportation and handling (Skoch et al., 1983 in Thomas and ver der Poel, 1996). Physical characteristics of fish feed are measured in terms of hardness, durability, sinking velocity, bulk density, fat absorption capacity, starch gelatinization and expansion rate (Sorensen, 2003). Since I have used only hardness, pellet durability and water stability test in my master theses, I have explained only these.

Hardness

Hardness is the force required to crush a pellet (Pfof, 1963 in Thomas and van der Poel, 1996). Pellet breakage might occur during handling, transportation and in storage silos. Hardness can be measured by different manual and automatic hardness tester. Stress, strain, elasticity are the terminologies required to explain hardness.

Durability

It is highly recommended that the structural integrity of the particles is retained till the feed is consumed by the animal (Crampton, 1985 in Aarseth, 2004). Durability is defined as the amount of fines obtained from pellets caused from pneumatic or mechanical agitation (Pfof, 1963). Fines or dust represents direct loss of feed and also causes inferior working environment and animal welfare (Kertz et al. 1981 in Aarseth, 2004). There are several durability testers available in the market. Some of the popular devices are; Holmen tester, tumbling box tester, ligno tester, tube tester, seedburo pellet durability tester. Pellet durability index is expressed as PDI%, which is calculated by dividing weight of pellet after test by weight of pellet before test and multiplied by 100.

Water stability

Water stability is a measure for the time it takes for a pellet to dissolve in water. Some species require a high water stability (for shrimp that eat slowly), while other species eat the feed immediately (typical of a species in cage culture systems). It can also be used to mirror the degradation pattern of feed in the stomach of fish. So, special focus needs to be taken into consideration depending upon the type of fish and water quality. Usually, water stability should be high to ensure that pellets remains intact and not dissolve quickly. Water stability is measured as loss of dry matter during a certain time in the shaking water bath.

Image analysis

An image analysis method was used to determine the color change of rice-glucose-lysine blend during extrusion (Lei et al., 2007). Thermal processing changes food color which can be used to predict the extent of quality deterioration of food. During food processing nonenzymatic browning reaction occurs which can be either desirable or undesirable (Baisier and Labuza, 1992 in Lei et al., 2007). Differences in the color of the extrudates can be studied by using various software programmes, for example, imageJ.

1.6 Effects of processing on physical quality of feed

Water and heat addition will affect raw materials like starch and protein in the feed mash and affects the binding properties of pellet (Thomas and van der Poel, 1996). Change in protein (denaturation) and starch (gelatinization) can be manipulated, if residence time is taken into consideration. Sorensen et al. (2010) found that the injection of steam into the extruder barrel affected physical quality of feed significantly, but most of the variation in physical quality was explained by starch source and screw configuration. Skoch et al. (1981) reported that more steam during the production improved pellet durability. Sorensen et al. (2010) found that depending on screw configuration, the holmen durability was in the range (37-76%), and hardness in the range (22-30N). Likewise, Aarseth et al. (2006) reported that physical quality of extruded pellets is affected by the extruder temperature. Reduction in viscosity and increase in steam pressure of the melt is the result of the elevated temperature. So consequently, reduction in viscosity and increase in steam pressure caused the expansion of the product.

Starch gelatinization is affected by steam pressure. At low steam pressure, more water, relative to heat is added to the diet, which enhances starch gelatinization. Conversely, high steam pressure is used where low amounts of water and higher temperature is required (Kinsella, 1979). According to Biliaderis et al. (1986), gelatinization is also affected by temperature. Cai et. al. (1995) showed that starch degradation occurs only in the cooking zone, rapidly in the beginning and then slowly till the die. In this process, there was a significant degradation of large amylopectin molecules in the cooking zone. Thus it confirms that thermal

energy enhances the degree of gelatinization. Mohamed (1990) reported that increasing the soya protein content from 0 to 25% resulted in decreased expansion of extruded corn starch. This indicates that proteins do not expand as well as starch. High shear, pressure, and temperature during extrusion, disrupts and alters protein structure (Harper, 1981 in Akdogan, 1999).

Gogoi et al. (1996) reported that the reverse screw and kneading element combination increased the specific mechanical energy, expansion ratio and water solubility index but decreased shear stress and bulk density. Olivira (1990) reported increased barrel temperature gave increased expansion ration mainly because of increased gelatinization of starch and denaturation.

1.7 Effects of ingredients on physical quality of feed

Physical quality of pellets (pellet durability and hardness) is greatly influenced by the ingredient composition of the diet (Wilson, 1994 in Thomas et al., 1998). Wood, (1987) reported that the degree of starch gelatinization and protein denaturation greatly influences durability and hardness of pellet. Generally, higher rate of starch gelatinization and protein denaturation gave higher durability and hardness of the pellet. Wood, (1987) obtained highest pellet durability (93%) with mixtures containing raw soya protein and pre-gelatinized tapioca starch but low durability was obtained with denatured soya protein and native tapioca starch. Also the higher specific output and output rate was obtained with higher pre-gelatinized starch content.

High moisture content during extrusion gives complete starch gelatinization and affects rheological properties of feed. Increased moisture content causes low melt viscosity and consequently result low pressure build-up, which in turn gives low expansion at the die. The processing condition and ingredients used have an effect on extrudate expansion. A low starch content of the recipe and low degree of gelatinization result in low expansion. Similarly, lower viscosity will impact the expansion (Akdogan, 1999). Sorensen et. al. (2010) reported that types of starch sources have different impact on physical pellet quality. They obtained highest Holmen durability (79%) with pre-gelatinized potato starch and lowest value (44%) with wheat starch. Snack-like products obtained by extrusion cooking of chest nut (rich in sugar)-rice flour limited the gelatinization and the expansion of the products (Sacchetti et al. 2004).

Diets processed with the inclusion of yeast cells improved tensile strength of pellets significantly (Aarseth et. al. 2006). Likewise, (Areas, 1992 in Gogoi et al. 1996) reported that addition of proteins to starch- rich flours gave less expansion and the product is harder and resistant to water dispersion.

Lipids also affect the extrudate expansion. Bhattacharya and Hanna, (1988) indicated that an increase in fish solids content in the formulation caused an increase in the lipid as well as the protein causing reduced expansion. Similarly, Bhattacharya and Hanna, (1987 in Singh and Smith, 1997) reported reduced expansion with increasing temperature for waxy maize starch. Singh and Smith, (1997) reported a decrease in expansion of oat flour extrudates with increasing moisture at 125 °C.

1.8 Aim of research

The objective of this research was to investigate the effect of starch source and processing parameters on the physical quality of the feed and to investigate if image analysis can be used to describe physical quality of pellets.

2. Materials and methods

Five feeds were produced at Fortek, UMB. Feeds were based on fishmeal mixed with four different starch sources. The starch sources used in my experiment were pre-gelatinized potato starch, potato starch, whole wheat and wheat starch. Three screw configuration types; polygon-2L, polygon-LR, and LRLR were used during production of feed. Similarly, steam injection and no steam injection in barrel were used for the production of the extruded fish feed. I have not explained the production process (extrusion) and the formulation ratio here, as this thesis is a continuation of the experiment conducted by Sørensen et al. (2010). A description of formulation, production parameters and chemical analysis is found in Sørensen et al. (2010). In this report, I am explaining the material and methods part, which I have conducted by myself.

A physical quality of the pellets which includes hardness, pellet durability and water stability was measured. Image analysis was also used to evaluate changes in color, and if these changes were correlated with physical quality of pellets.

2.1 Measurement of pellet durability

Durability was measured using Holmen pellet tester (Borregaard Lignotech, Hull, UK). Surface attrition was measured as the pellets were conveyed at high air velocity with reference to time. Attrition of surface occurs when pellets hit pipe walls, bends, and other pellets. 100g pellets were taken and the Holmen was run for 60s and 30s respectively. Broken pellets and the dust were collected after sieving. Pellet durability index (percentage) was calculated using the following formula. Three replications were taken for measurement.

$$\text{PDI (\%)} = \frac{\text{weight of intact pellets after Holmen (g)}}{\text{weight of pellets before Holmen (g)}} \times (100)$$

2.2 Measurement of Hardness

Hardness was measured using manual Kahl pellet tester. The pressure/force was applied until any visible sign of breakage of pellet was observed. The length and width of pellets were maintained to some degree of similarity, but was not possible all the time because of irregular and diverse size and shape of pellets. Each sample (30 pellets) was divided into three groups (10 each group) and average was calculated. The value was recorded as length (mm), width (mm) and force (kg). Result was presented as mean and the standard deviation.

2.3 Measurement of Water stability

10g of feed was put into wire mesh cylinder. The weight of the cylinder was measured before the sample was placed in the bath. Cylinder with the feed was transferred to bottle (800ml) with water. The bottle was again transferred to shake incubator and the samples were incubated at constant temperature and rotation for specific period of time. The incubation temperature, rotation and time were 24 °C, 1400 rpm and 120 minutes respectively. After the procedure was finished, weight of sample was taken and the samples were dried in oven drier for 24 hours at 105 °C. Dry matter of the feed was calculated. The difference in initial weight of the sample and the weight of the sample after water treatment was measured and expressed as percentage.

2.4. Color analysis

Macro picture of the pellets were taken. Pellets were put in the petridish and camera fixed in a tripod stand with constant height and light intensity was used to take the picture. The camera used was a digital 12 megapixel Nikon, D200 model (Nikon Corporation, Tokyo Japan). Image j software was used to determine the brightness, contrast and entropy values of the picture. Software was Java programming language (National institutes of Health, Maryland USA). Using the color analysis program of the image j, the values were easily obtained.

Brightness gives the grey level of the picture and was calculated by using the following equation; $\text{grey level} = 0.3 \cdot R + 0.6 \cdot G + 0.11 \cdot B$ where, R, G, B denotes red, green and blue respectively.

Contrast is the local variation from the mean in a smaller area. It can also be called as local standard deviation.

Entropy is the measure of information content. It measures the randomness of intensity distribution. Entropy is higher when all the entries are of similar magnitude and smaller when the entries are unequal.

The visible difference in the color of the pellets was also observed by visual inspection. So it was also used as materials for result and discussion.

2.5 Statistical analysis

Excel (2010) was used to calculate mean and the standard deviation for durability, hardness and water stability. Results are presented as percentage value. SAS computer software (SAS 1990) was used to perform analysis of variance (ANOVA) for brightness, contrast and entropy. The experiment set up was 3_x 4_x2 factorial design with three screw configuration (Polygon-2L, Polygon-LR, and LRLR), four carbohydrate sources (Pregelatinized potato starch, potato starch, whole wheat and wheat starch) and either with steam injection and without steam injection into the barrel. Duncan's multiple range test was done to find the significant ($P \leq 0.05$) differences among means.

2. Results

Pre-gelatinized potato starch had significantly higher pellet durability than whole wheat and wheat starch diet. Potato starch also had higher pellet durability compared to whole wheat and wheat starch diet but lower than pre-gelatinized potato starch (Table 1). Similarly, pre-gelatinized potato starch and potato starch had higher water stability compared to other diets (Table 1). Pellet hardness breaking force and diameter was highest in potato starch diet. Pellet length was highest for potato starch and whole wheat (Table 1).

Screw configuration LRLR gave the highest pellet durability and Polygon-LR had the lowest. Screw configuration LRLR also gave higher water stability than the Polygon-2L but the numerical difference was small. Screw configuration Polygon-2L and LRLR gave higher breaking force than Polygon-LR. There was no difference in the effect of screw configuration on length and diameter (Table 1).

No Steam injection in a barrel gave a higher pellet durability and water stability. Steam pressure did not affect length, diameter and breaking force of the pellet (Table 1).

Table 1 Physical quality of pellets produced with different starch sources, thermal treatment and screw configuration presented as mean values and standard deviation.

	Pellet Durability (%)	Water Stability (%)	Length (mm)	Diameter (mm)	Force (Kg)
Starch Source					
Whole wheat	65.65±18.01	75.73±2.09	7.22±0.35	5.5±0.08	2.13±0.81
Potato starch	82.94±15.36	81.87±5.48	7.02±0.4	5.64±0.18	2.71±0.78
Pre-gelatinized potato starch	92.73±2.71	81.25±1.91	6.5±0.1	5.6±0.08	2.58±0.48
Wheat starch	67.23±13.12	79.08±1.2	6.64±0.09	5.53±0.06	1.66±0.51
Screw configuration					
Polygon-2L	79.47±14.79	78.9±4.05	6.94±0.41	5.58±0.15	2.31±0.64
Polygon-LR	67.38±20.54		6.93±0.42	5.53±0.11	1.74±0.75
LRLR	84.56±12.02	80.07±3.59	6.67±0.33	5.59±0.09	2.76±0.53
Steam pressure					
No steam	80.96±19.34	80.78±3.02	6.9±0.4	5.53±0.11	2.32±0.84
Steam	73.31±14.38	78.18±4.14	6.79±0.4	5.6±0.13	2.23±0.67

Contrast and entropy was significantly ($P<0.05$) affected by starch source. Wheat starch had the highest contrast but lowest entropy. Whole wheat gave the lowest contrast but highest entropy. Potato starch also had a higher contrast than whole wheat and pre-gelatinized potato starch (Table 2). Screw configuration and steam pressure did not give significant differences in contrast and entropy (Table 2).

Table 2. Effect of starch sources, Screw configuration and Steam Pressure on Contrast and Entropy

	Contrast	SE(±)	Entropy	SE(±)
Starch source				
Whole wheat	12.86	0.31	6.9	0.02
Potato starch	13.81	0.33	6.86	0.02
Pre-gelatinized potato starch	12.98	0.42	6.83	0.02
Wheat starch	14.08	0.38	6.79	0.11
Screw configuration				
Polygon-2L	13.02	0.26	6.85	0.02
Polygon-LR	13.85	0.39	6.88	0.02
LRLR	13.42	0.3	6.81	0.08
Steam Pressure				
No steam	13.7	0.28	6.9	0.02
Steam	13.16	0.24	6.8	0.05

Starch source tended to affect brightness values ($P=0.07$). Screw configuration and steam pressure did not affect brightness. Wheat sources, in particular wheat starch, gave the highest brightness value. The lowest brightness value was obtained for potato starch (Table 3). Screw configuration (Polygon-LR) had the highest brightness while screw configuration Polygon-2L and LRLR gave the similar brightness value (Table 3). Steam produced higher brightness than diet without steam (Table 3).

Table 3. Effect of starch sources, screw configuration and steam pressure on brightness

	Brightness	SE(\pm)
Starch source		
Whole wheat	60.25	1.01
Potato starch	57.3	1.06
Pre-gelatinized potato starch	59.16	1.15
Wheat starch	61.17	1.38
Screw configuration		
Polygon-2L	58.55	1.14
Polygon-LR	60.96	0.78
LRLR	58.89	1.09
Steam Pressure		
No steam	58.62	0.89
Steam	60.31	0.77

Wheat starch* Polygon-LR*No steam injection combination gave a highest contrast (15.22) over all combinations. The lowest contrast (11.16) was observed for whole wheat*LRLR*steam injection combination (Table 4).

The interaction SCF*Steam ($P=0.002$) as well as Starch*SCF* Steam ($P=0.04$) had a significant effect on contrast. Brightness was also affected by the Starch*Steam interaction ($P=0.03$). Highest contrast was obtained for Polygon-LR*No steam injection combination. Lowest contrast was observed for Polygon-2L*No steam injection combination.

Highest brightness (61.52) was observed for whole wheat *No steam injection combination. The lowest was observed for potato starch * No steam injection combination (Table 4).

Table 4. Effect of interactions between starch sources, screw-configuration and steam pressure on contrast and brightness. Only significant interactions are presented.

Starch*SCF*Steam				
Starch	SCF	Steam	Contrast	SE(±)
3	c	+	11.16	0.44
4	a	-	13.53	0.2
4	a	+	13.2	0.91
4	b	-	15.22	1.03
4	b	+	14.13	1.18
4	c	-	13.67	1.36
4	c	+	14.72	0.87

SCF*Steam			
SCF	Steam	Contrast	SE(±)
a	-	12.65	0.39
a	+	13.38	0.33
b	-	14.86	0.49
b	+	12.84	0.45
c	-	13.58	0.36
c	+	13.27	0.49

Starch*Steam			
Starch	Steam	Brightness	SE(±)
1	-	57.67	1.31
1	+	60.64	1.42
2	-	54.17	1.41
2	+	60.43	0.59
3	-	61.52	1.71
3	+	58.98	1.5
4	-	61.12	1.62
4	+	61.21	2.34

Starch 1, 2, 3 and 4 indicates Pre-gelatinized potato, Potato, Whole wheat and Wheat starch respectively.

Screw configuration a, b and c indicates Polygon-2L, Polygon-LR and LRLR respectively.

Steam - and + indicates no steam injection and steam injection respectively.

4. Discussion

A. Effect of starch sources

To a large extent diets prepared with potato starch gave higher durability, water stability, hardness, length and diameter of the pellets than the diets prepared with wheat starch (Table 1). The results suggest that the starch sources had different functional properties, affecting the functional properties. Swinkels (1985) reported that starch functionality is affected by starch structure and size of the granules and morphology. Each starch source has a distinct amylose: amylopectin ratio. This ratio affects the intermolecular bonding and water absorption. Potato has 20:80 and wheat 25:75 ratio of amylose: amylopectin content (Thomas and Atwell, 1999). Native potato starch has weak intermolecular bonding between the amylopectin molecules and can therefore absorb more water. The swelling power of wheat is low and may explain lower water absorption (Swinkels, 1985). In the presence of heat and moisture, bonds are disrupted leading to swelling of the potato starch higher viscosity pastes (Bemiller and Whistler, 1996). Thomas and van der Poel, (1996) suggested that ingredients with high viscous properties will stick particles together, improving the binding strength and eventually improving the physical quality of pellet. Furthermore, Sørensen et al. (2010) reported that types of starch sources have different impact on physical pellet quality. They observed highest Holmen durability (79%) with pre-gelatinized potato starch and lowest value (44%) with wheat starch.

Potato starch has larger grain size than wheat starch (Charley, 1982). Bouvier, (1996) demonstrated that large starch grain size gave higher expansion. Desrumaux et al., (1998) also found that large particle size gave harder extrudates. Pre-gelatinized starch is pre-cooked and is completely gelatinized. Granular structure are broken down during pre-gelatinization process, increasing water absorption and water solubility (Colonna et al., 1984) giving highest durability, water stability and hardness as in our case. Gelatinization of potato starts at lower temperature than wheat starch so usually there is higher degree of gelatinization in potato, giving higher water stability (Charley, 1982).

B. Effect of screw configuration

LRLR gave highest durability, water stability and hardness. Screw configuration affects the SME (specific mechanical energy) and retention time on extruder and thereby affects the quality of pellets. Screw configuration setting might influence retention time in the barrel and mechanical energy input. Screw speed also plays a role in physical quality of pellets. Higher speed increases the mechanical energy, which in turn reduce the retention time and fill in the barrel.

C. Effect of steam addition

Pellet durability, water stability and the hardness was higher with no steam injection in the barrel (Table 1). One explanation may be that steam injection increased temperature, solubility and reduced friction in the barrel resulting in less durable pellets with lower water stability and hardness.

4.1 Discussions of color analysis

A. Effect of starch sources, screw configuration and steam pressure on contrast and entropy

Contrast and entropy appeared to be associated with the process in the extruder. The granular structure, packing of amylose and amylopectin within the granule has a major role for gelatinization and functionality affecting the contrast and entropy of a picture. Whole wheat starch is tightly packed, giving the lowest contrast and highest entropy. While wheat starch, which was extracted from the whole wheat, had highest contrast and lowest entropy. But on the other hand, potato starch having a stable or similar contrast and entropy could be due to the higher swelling property and water absorption quality and extent of gelatinization. The higher and lower values for contrast and entropy could not be explained and also not supported by relevant literature. Other experiments (Dan et al. 2007) showed that contrast and homogeneity was associated with crispiness/crunchiness of food products like, ritz crackers and premium crackers. With regard to animal feed it can be argued that contrast and entropy could have impact on dust formation. If extruded fish feed pellets are brittle or crisp due to starch source, processing conditions and processing parameters than during transportation and handling it could create dust.

Screw configuration and steam pressure did not show significant effect on contrast and entropy of the picture. Most likely, the physical characteristics or color changes was not big enough to be affected by the image. If more extreme screw configuration and steam pressure was used, this would more likely have effect on contrast and entropy.

B. Effect of starch sources, Screw configuration and Steam Pressure on brightness

Feed prepared from potato starch sources tended to have lower brightness compared to wheat sources. Effect of starch sources on brightness was not significant but showed only tendency (Table 3). It can be explained that potato starch in the presence of heat and water, undergoes nonenzymatic browning reaction, making the final product much dark colored. Manufacturing of potato powder/meal can also risk undergoing oxidation, giving brown color. Peeled potato surface are highly reactive as a consequence of thermal and mechanical damage during peeling (Sarper et. al. 1995). Wheat starches are less susceptible for nonenzymatic browning reaction, contributing to higher brightness. Air temperature and puffing time were major factors that affected volume expansion ratio and nonenzymatic browning of potato cubes (Mohini, 2007). Mandarin juice concentrates heated at 98°C caused carotene loss and

color was increased, probably due to maillard reaction (Ibarz et al., 2011). Baisier and Labuza, (1992 in Lei et al., 2006) stated that nonenzymatic browning reaction are significant phenomena that happens during food processing and storage. Nonenzymatic reaction is dependent on temperature and water activity of the food (Driscoll and Madamba, 1994 in Lei et al 2006). Screw configuration and steam pressure did not show significant effect on brightness as because brightness is the product of chemical reaction not the mechanical configuration.

C. Effect of interactions between starch sources, screw-configuration and steam pressure on contrast and brightness

The highest contrast for Wheat starch*polygon-LR*no steam injection could not be explained or supported by literature (Table 4). Likewise, lowest contrast was observed for whole wheat*LRLR*with steam injection (Table 4) but again it could not be explained.

Highest brightness observed for starch whole wheat*no steam injection can be explained by low nonenzymatic browning (caramelization and maillard reaction) in wheat. The lowest brightness was observed for potato starch*no steam injection (Table 4). Extent of caramelization and maillard reaction is higher in potato than in wheat causing the final product to be much darker than the wheat based diet. Moss and Otten (1989) stated that many researchers use color as a quality control indicator of processing condition because brown pigments increase as browning and caramelization reaction progress. With high temperature, high pressure but low moisture content of the feed, extrusion process gives colored products even though the residence time is short (Lie et al., 2006).

5. Conclusions

Physical quality of feed was affected by starch source, screw configuration and steam injection into the barrel. Potato starch gave the highest pellet durability, water stability and hardness. Screw configuration LRLR gave the highest durability, water stability and hardness. Likewise, no steam injection in the barrel gave the highest durability, water stability and hardness. The results also showed that image analysis discriminated among the feeds. Contrast and brightness changed with starch source, but was not affected by other processing parameters such as screw configuration and steam injection. Image analysis alone cannot be used to describe the physical quality such as durability or hardness but it can help explain the process undergone during extrusion, which might explain in part about the physical quality.

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