Properties of Extrusion Processed Corn and Corn Coproducts

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Abstract

As the world population continues to grow, the demand for human food and animal feed grows exponentially. Aquaculture is the food sector which has been growing at the greatest rate for several years. Because of the expense of fishmeal in aquaculture fees, an inexpensive protein source could be corn-based proteins. Although many studies have focused on the effects of extruding corn-based blends along with other supplement ingredients, few studies have focused on the extrusion of individual corn-based ingredients. This study examined physical effects of extrusion on distillers dried grains with soluble (DDGS) and corn. Specific objectives included determining moisture content, water activity, color, unit density, durability, water stability, floatability, and bulk density for each corn-based extrudate. Blends were prepared with three levels of moisture (15, 25, and 35% db), and extrusion conditions included three screw speeds (50, 75, and 100 rpm) and three barrel temperatures (100, 125, and 150 °C). Results showed that as the moisture content increased, the water activity increased in the raw ingredients, and the moisture content of the extrudates increased. As the screw speed increased, the floatability of the extrudates increased, while the bulk density decreased. The amount of protein and starch content in the corn products affected the physical quality of the pellets, which is important in aquaculture feed development.

Keywords: extrusion, corn, DDGS, aquaculture, feed

1. Introduction

As the world's population continues to grow exponentially, the need for efficient food and feed production increases each day. Snack foods and breakfast cereals are among many food industries that have grown in the United States, and many of these are produced through extrusion (Razzaq et al., 2012).

Along with the human food industry, aquaculture is one of the fastest growing food production activities globally. It plays a major role in many third world countries because it produces relatively higher income, better nutrition, and better employment opportunities (Kannadhason et al., 2008). Nutritional feed for aquaculture is costly, and the feed cost is generally 30% to 60% of total operational costs (Kannadhason et al., 2009, 2010), both due to raw ingredient costs as well as processing costs. Because feed is so expensive, it is difficult to provide the fish appropriate nutrients they need to thrive.

Extrusion has become a popular processing technique in the feed, cereal, and snack food industries (Delgado-Nieblas et al., 2012). Extrusion is one of the most versatile unit operations (Forsido and Ramaswamy, 2011). Extrusion systems consist of barrel housing with one or two rotating screws, a preconditioner, and an accompanying machine control system (Sorensen, 2012). Extrusion cooking uses high temperatures in a relatively short time to move the ingredients through the barrel and exit through the die. The goals of the extrusion process include cooking, sterilization, expansion, texturization, and product shaping. These factors are especially important in aquaculture feeds because porous pellets that float in water are required. Moreover, these are all important factors for human foods as well (Liu and Rosentrater, 2011).

During the extrusion process, starch is gelatinized, which plays an important role in the final extrudate properties (Chevanan et al., 2008a). Gelatinization is crucial because it affects feed digestibility, expansion, and contributes to water solubility and particle binding (Rosentrater et al., 2009a,b). Along with starch gelatinization, protein denaturation and destruction of microbes and other toxic compounds are other reactions that may also occur

during cooking (Kannadhason et al., 2008). The sterilization process allows for the termination of pathogens, which is important in food products for humans, as well as fish (Liu and Rosentrater, 2011).

In the Midwest U.S., corn and soybean crops are the most prevalent for food production and processing. Both corn and soy are relatively high in protein, which is important for aquaculture feeds. According to a study performed at North Dakota State University, corn contains approximately 55-60% escape or bypass protein (Lardy, 2002). The United Soybean Board found soybeans contain 35-38% protein (The United Soybean Board [USB], 2012). Many types of concentrated and isolated meals have been produced for livestock feed. Using the high-protein feed produced for livestock has potential to be extruded for use in aquaculture feed.

Changes of the extrusion conditions, especially barrel temperature, screw speed, and moisture content can have significant effects on the resulting physical properties of the extrudates (Singh et al., 2012). Physical properties are important because they impact the quality of the extrudates. Moisture content is a determining factor for cohesiveness (Ayadi et al., 2011a). Water activity values indicate shelf life (Ayadi et al., 2011b). Because spoilage microorganisms thrive in water activity levels greater than 0.5, a value less than 0.5 is preferred. The greater the values of water activity, the more likely the microorganisms are to thrive, so a lower water activity value is preferred. Simons found that as extruder screw speed increases, water activity decreases (Simons et al., 2012). Color of the extrudate indicates, to some extent, the nutritional quality of the product (Kannadhason et al., 2010), as darker colors can indicate protein degradation. The mass flow rate quantifies the processing performance of the extruder (Ayadi et al., 2011c). Rosentrater found that an increase in both moisture content and temperature result in a decrease in die pressure (Rosentrater et al., 2009a,b). Chevanan found that increasing the moisture content resulted in an increase in pellet durability (Chevanan et al., 2007a). They also found that increasing the temperature resulted in a decrease in apparent viscosity and pellet durability. Chevanan also found that changes in screw speed had a significant effect on all extrusion parameters except for the temperature of the die (Chevanan et al., 2008b). They also found that increasing the screw speed resulted in a decrease in bulk density because of greater expansion (Chevanan et al., 2007b).

The ingredients used in feed extrusion must generally be high in protein, because aquaculture feeds typically required 26 to 50% protein, depending on the fish type, species, and age (Chevanan et al., 2007c). Because of the recent biofuel revolution, the production of ethanol has grown immensely across the United States. A coproduct of ethanol manufacturing is distillers dried grains with solubles (DDGS), which contains moderate levels of protein, fat, and fairly high levels of fiber. Because of the nutritional value provided by DDGS, it has become a key ingredient in feeds for many species, including ruminants and non-ruminants. Research on using DDGS in fish feeds is also promising (Liu and Rosentrater, 2011).

The purpose of this study was to evaluate the effects of extrusion conditions on DDGS, focusing on the effects of ingredient moisture content, extruder screw speed, and barrel temperature. In order to find optimal extrudate properties, extensive physical property testing was conducted after the extrusion process, and results were compared to corn – based extrudate properties.

2. Materials and Methods

2.1 Sample Preparation

Corn ingredients, including DDGS and corn (for comparison purposes), were ground to <1.0 mm particle size using a laboratory mill (Model 4, THOMAS-Wiley, Swedesboro, NJ, USA). The ingredients were then adjusted to three target moisture contents (15, 25, and 35% db) by adding appropriate amounts of water. The ingredients and water were mixed for approximately 20 min, using a small rotary mixer (Model 043206, Type B, Kobalt, Surrey, BC, Canada). The water was added slowly using a spray bottle to prevent agglomeration. The blends were stored overnight to allow for the distribution and equilibration of moisture at room temperature (25 ± 1 °C).

2.2 Extrusion Processing

A single-screw extruder (Model PL2000, Type 680143, C. W. Brabender, South Hackensack, NJ, USA) was used for the cooking process. The screw compression ratio was 1:1, and the diameter of the die was 3.05 mm. The temperature of the feeding zone was room temperature $(25 \pm 1 \text{ C})$, the metering zone was adjusted to 75 °C, and the die zone was adjusted to one of three temperatures (100, 125, and 150 °C) throughout the process. The extruder was capable of a screw speed ranging from 0-100 rpm, so the screw speed was adjusted to three speeds (50, 75, and 100 rpm) for the appropriate treatments. Once the processing was completed, the extrudates were stored for approximately 72 h at room temperature (25±1 °C), allowing them to air dry.

2.3 Measurement of Extrudate Properties

The extrudates were subjected to extensive physical property analyses, including moisture content (% db), water

activity (a_w , -), color (L*, a*, b*), unit density (g/cm³), pellet durability (%), water stability (s), floatability (s), and bulk density (kg/m³). The mass flow rate (g/min) and the temperature of the feeding, metering, and die zones (°C) were recorded during the extrusion process.

2.3.1 Moisture Content (MC)

Moisture content was determined using approved American Association of Cereal Chemists [AACC] method 44-19 (2000) with a laboratory oven (HERAtherm, Thermo Scientific, Waltham, MA, USA). The samples were prepared and heated for 2 h at 135 °C. Moisture content was determined for both the raw ingredients, as well as the extrudates.

2.3.2 Water Activity (a_w)

The water activity was determined for both the raw ingredients, as well as the extrudates using a water activity meter (Series 3TE, AquaLab, Pullman, WA, USA). The meter was calibrated and a sample cup was filled with each sample and placed inside the reading chamber.

2.3.3 Color

Color (L*, a*, and b*) of the raw ingredients and extrudates was determined using a spectrophotometer (LabScan XE, HunterLab, Reston, VA, USA). L* is the brightness/darkness, a* is the redness/greenness, and b* is the yellowness/blueness.

2.3.4 Unit Density (UD)

Extrudates were cut using a razor blade into pieces approximately 2 cm long. Each extrudate was weighed on an electric balance (Model AV114, OHAUS Adventurer Pro, Pine Brook, NJ, USA), and the length and diameter were determined using a digital calliper (Model 14-648-17, Fisher Scientific, Pittsburgh, PA, USA). The unit density was defined as the ratio between the mass compared to the volume, which was based on the cylindrical shape of the extrudate.

2.3.5 Pellet Durability Index (PDI)

Approximately 200 g of extrudates were broken into pieces approximately 2 cm long, then placed in a pellet durability tester (Model PDT-110, Gamet Automatic Sampling Equipment, St. Paul, MN, USA) for a 10-min tumbling period. After tumbling, the samples were then sieved using a no. 8 sieve (2.36 mm) (Model 122622665, Fisher Scientific, Pittsburgh, PA, USA). The pellet durability index was calculated using the following equation:

$$PDI = (M_{at}/M_{bt}) \times 100 \tag{1}$$

Where,

PDI = Pellet durability index (%)

 M_{at} = Mass of the pellets after tumbling (g)

 M_{bt} = Mass of the pellets before tumbling (g)

Bulk Density (BD): Using the method described by the USDA (1999), bulk density was measured using a standard bushel tester (Model 151, Seedburo Equipment Co, Chicago, IL, USA) and a test weight 1-L cup and leveling stick (Model 103, Seedburo Equipment Co, Chicago, IL, USA).

2.3.6 Mass Flow Rate

The mass flow rate was determined by collecting extrudates at the die for 30-s intervals, and then weighing them on an electronic balance (Model EX10201, Ohaus Explorer, Pine Brook, NJ, USA). The extrudates were then dried to determine the moisture content, which gave the dry-basis mass flow rate.

2.3.7 Statistical Analysis

Raw ingredients were adjusted to three moisture content levels (15, 25, and 35% db). The blends were extruded at three die temperatures (100, 125, and 150°C) and three screw speeds (50, 75, and 100 rpm). Each of the factors included a high point and a low point with a central composite point, which resulted in 9 treatments (i.e. 2x2x2=8, plus 1 center point). The experimental protocol is shown in Table 1. Most of the physical properties were measured in triplicate (n=3), except for mass flow rate, which was measured in duplicate (n=2). The results were analyzed via Statistical Analysis System (SAS) software, which determined the means and standard deviations for each treatment, as well as the main effects, interactions, and treatment combination effects, using an $\alpha=0.05$.

Treatment	Screw Speed (rpm)	Moisture Content (% db)	Temperature (°C)
1	50	15	100
2	50	15	150
3	50	35	100
4	50	35	150
5	100	15	100
6	100	15	150
7	100	35	100
8	100	35	150
9 (Center Point)	75	25	125

Table 1. Experimental design*

*The run value describes the order each sample was extruded. Experimental design was a $2 \times 2 \times 2 + 1$ center point = 9 treatment conditions. The Center Point was defined as Treatment 9.

3. Results and Discussion

With the extrusion conditions used in the study, neither the corn nor the DDGS extrudates were of exceptionally high quality, unfortunately. As shown in Figure 1, the DDGS extrudates did not exhibit extensive particle binding during processing, which led to pellets which were not highly cohesive. (Images of corn extrudates are not provided, but exhibited similar behavior.) It is likely that increasing the moisture of the raw blends could have improved particle binding, and thus pellet quality. It should be noted that in subsequent tables that some of the extrusion conditions did not produce results – this occurred due to starch gelatinization within the extruder barrel, which caused the screw to foul, and thus extrusion had to be discontinued.



Figure 1. Bulk and cross-sectional (100x) images of the DDGS extrudates for each treatment (1-9). Image analysis indicates that the quality of the extrudates was low due to incomplete particle binding which occurred during all extrusion conditions used in this study.

3.1 Raw Ingredient Properties

Table 2 and Table 3 provide the results of the effects of moisture content on the water activity and color (L*, a^* , b^*) of the DDGS and corn, respectively.

Treatment	Moisture Content	Water Activity		Color	
Heatment	(db, %)	(a _w)	L*	a*	b*
1	15.20	0.61	58.92	14.82	48.48
1	(1.11)	(0.00)	(0.95)	(0.28)	(0.08)
2	15.20	0.60	58.92	14.82	48.48
2	(1.11)	(0.00)	(0.95)	(0.28)	(0.08)
3	33.78	0.83	58.92	14.82	48.48
3	(0.71)	(0.00)	(0.95)	(0.28)	(0.08)
4	33.78	0.83	58.92	14.82	48.48
4	(0.71)	(0.00)	(0.95)	(0.28)	(0.08)
5	15.20	0.60	58.92	14.82	48.48
5	(1.11)	(0.00)	(0.95)	(0.28)	(0.08)
6	15.20	0.60	58.92	14.82	48.48
0	(1.11)	(0.00)	(0.95)	(0.28)	(0.08)
7	33.78	0.83	58.92	14.82	48.48
,	(0.71)	(0.00)	(0.95)	(0.28)	(0.08)
0	33.78	0.83	58.92	14.82	48.48
0	(0.71)	(0.00)	(0.95)	(0.28)	(0.08)
0	23.95	0.75	58.92	14.82	48.48
9	(0.61)	(0.00)	(0.95)	(0.28)	(0.08)

Table 2. Raw DDGS properties*

* Averages and standard deviations (shown in parentheses) for various physical properties of raw DDGS samples.

Table 3. Raw corn properties *

Traatmant	Moisture Content	Water Activity		<u>Color</u>	
meanneint	(db, %)	(a_w)	L*	a*	b*
1	16.16	-	80.66	5.20	32.41
1	(0.77)	-	(1.18)	(0.20)	(1.49)
2	16.16	-	80.66	5.20	32.41
2	(0.77)	-	(1.18)	(0.20)	(1.49)
3	34.26	0.97	80.66	5.20	32.41
5	(0.66)	(0.00)	(1.18)	(0.20)	(1.49)
4	34.26	-	80.66	5.20	32.41
	(0.66)	-	(1.18)	(0.20)	(1.49)
5	16.16	0.75	80.66	5.20	32.41
	(0.77)	(0.00)	(1.18)	(0.20)	(1.49)
6	16.16	0.76	80.66	5.20	32.41
	(0.77)	(0.00)	(1.18)	(0.20)	(1.49)
7	34.26	0.98	80.66	5.20	32.41
/	(0.66)	(0.00)	(1.18)	(0.20)	(1.49)
0	34.26	0.98	80.66	5.20	32.41
8	(0.66)	(0.00)	(1.18)	(0.20)	(1.49)
0	23.68	-	80.66	5.20	32.41
9	(0.47)	-	(1.18)	(0.20)	(1.49)

*Averages and standard deviations (shown in parentheses) for various physical properties of raw corn samples.

3.1.1 Moisture Content

According to Table 2, the exact moisture contents were relatively close to the desired moisture content of the raw DDGS. The exact moisture content values were off by 1.34, 4.40, and 3.62% for the targets of 15, 25, and 35%, db, respectively. Because of the low variance, the extrusion process was performed without further adjustment.

3.1.2 Water Activity

The water activity level of the raw ingredients is important because the greater the value, the greater the chance for microbial metabolism and growth. The greater the microbial growth rate, the more likely the product is to spoil. According to Table 2, as the moisture content increased, the water activity level increased in the raw DDGS blends. Similar results are seen in Table 3 for the raw corn blends.

3.2 Extrudate Properties

Table 4 and Table 5 provide the results for the treatment effects of moisture content, screw speed, and temperature on the moisture content, water activity, color (L*, a*, b*), unit density, pellet durability, water stability, floatability, and bulk density of the DDGS and corn extrudates, respectively. The tables display the

averages for each property, as well as the standard deviations, which are shown in parentheses. Table 6 displays the means and standard deviations for the main effects on the dependent variables of the DDGS extrudates. Table 7 displays the interaction effects.

Traatmant	Moisture Content	Water Activity		Color		Unit Density	Pellet Durability	Water	Floatability	Bulk Density
Heatment	(db %)	(a _w)	L*	a*	b*	(g/cm^3)	Index (%)	Stability (s)	(s)	(kg/m^3)
1	15.52cd	0.63c	52.56abc	12.57b	41.10bc	0.72ab	38.83bc	4.00de	2.67abc	378.80bc
1	(0.29)	(0.01)	(1.72)	(0.18)	(0.67)	(0.05)	(2.27)	(0.0)	(0.58)	(0.60)
2	15.32cd	0.63c	50.45c	14.02a	41.13bc	0.75ab	34.87cd	8.00b	3.33a	372.00c
2	(0.81)	(0.0)	(1.36)	(0.33)	(0.13)	(0.11)	(1.60)	(1.00)	(0.58)	(4.93)
2	18.80b	0.70a	52.34bc	12.70b	40.03c	0.78ab	40.60bc	6.00c	2.00c	399.00a
3	(0.37)	(0.0)	(0.93)	(0.50)	(0.95)	(0.08)	(7.59)	(1.00)	(0.0)	(2.16)
4	16.03c	0.62c	53.49ab	14.23a	44.45a	0.85a	-	1.67fg	3.33a	-
4	(0.33)	(0.01)	(0.94)	(0.39)	(1.24)	(0.04)	-	(0.58)	(0.58)	-
5	15.28cd	0.65b	52.72abc	12.92b	41.96b	0.62b	36.53cd	3.67de	2.33bc	355.00d
5	(0.29)	(0.02)	(1.21)	(0.16)	(0.52)	(0.03)	(1.12)	(0.58)	(0.58)	(7.34)
6	14.58d	0.63c	52.20bc	12.77b	40.25c	0.86a	47.90a	5.00cd	3.00ab	324.73e
0	(0.32)	(0.01)	(2.59)	(0.16)	(0.20)	(0.31)	(1.71)	(1.00)	(0.0)	(6.82)
7	20.53a	0.68a	55.35a	13.82a	43.81a	0.69ab	30.63d	3.00ef	2.67abc	381.80b
,	(2.15)	(0.03)	(2.11)	(0.29)	(0.94)	(0.09)	(4.68)	(1.00)	(0.58)	(0.20)
0	18.82b	0.69a	55.06ab	13.89a	44.32a	0.84ab	17.33e	0.40g	2.67abc	385.53b
0	(0.28)	(0.01)	(1.96)	(0.15)	(1.48)	(0.13)	(6.05)	(0.0)	(0.58)	(1.29)
0	16.28c	0.64bc	52.43bc	12.64b	40.77bc	0.75ab	43.83ab	9.67a	2.00c	359.33d
9	(0.17)	(0.01)	(1.65)	(0.28)	(0.18)	(0.11)	(2.68)	(1.15)	(0.0)	(0.99)

Table 4. Treatment effects on properties of DDGS extrudates*

Averages and standard deviations (shown in parentheses) for various physical properties of DDGS extrudates. The treatment effect relationships are shown below. Differing letters indicate significant differences among treatments ($\alpha = 0.05$) for a given dependent variable.

Tractmont	Moisture Content	Water Activity		Color		Unit Density	Pellet Durability	Water	Floatability	Bulk Density
Treatment	(db, %)	(a _w)	L*	a*	b*	(g/cm^3)	Index(%)	Stability (s)	(s)	(kg/m^3)
1	-	-	-	-	-	-	-	-	-	-
1	-	-	-	-	-	-	-	-	-	-
2	-	-	-	-	-	-	-	-	-	-
2	-	-	-	-	-	-	-	-	-	-
2	11.39	0.65	46.66	10.81	40.93	1.32	99.80	1800.00	<1	620.33
3	(0.96)	(0.01)	(1.96)	(0.89)	(1.13)	(0.03)	(0.10)	(0.0)	(0.0)	(1.70)
4	-	-	-	-	-	-	-	-	-	-
-	-	-	-	-	-	-	-	-	-	-
5	12.69	0.65	62.54	4.90	35.06	1.14	97.73	1729.00	<1	490.07
5	(0.13)	(0.0)	(2.31)	(0.41)	(1.28)	(0.08)	(0.15)	(67.77)	(0.0)	(5.90)
C	12.89	0.63	66.39	3.57	31.37	0.78	-	468.00	902.67	-
0	(0.05)	(0.0)	(1.81)	(0.32)	(0.95)	(0.05)	-	(44.00)	(85.80)	-
7	15.42	0.72	52.47	8.99	40.78	1.31	99.40	1800.00	<1	599.20
/	(5.35)	(0.04)	(0.90)	(0.33)	(1.22)	(0.09)	(0.70)	(0.0)	(0.0)	(3.14)
0	14.41	0.65	57.99	6.16	31.40	1.12	98.90	1800.00	0.33	446.87
0	(0.24)	(0.01)	(2.04)	(0.88)	(1.62)	(0.08)	(0.10)	(0.0)	(0.0)	(1.68)
9	-	-	-	-	-	-	-	-	-	-
	-	-	-	-	-	-	-	-	-	-

Table 5. Treatment effects on properties of corn extrudates*

* Averages and standard deviations (shown in parentheses) for various physical properties of corn extrudates.

Table 6. Main effects on properties of DDGS extrudates*

Parameter	Lavals	Moisture Content	Water Activity		Color		Unit Density	Pellet Durability	Water Stability	Floatability	Bulk Density
Taraneter	Levels	(% db)	(a _w)	L*	a*	b*	(g/cm ³)	Index (%)	(s)	(s)	(kg/m ³)
	15	15.18a	0.64a	51.99a	13.07a	41.11a	0.74a	39.53a	5.17a	2.83a	357.63a
Moisture	15	(0.55)	(0.01)	(1.80)	(0.62)	(0.74)	(0.17)	(5.46)	(1.90)	(0.58)	(22.32)
Content (%	25	16.28b	0.64a	52.43ab	12.64b	40.77a	0.75a	43.83a	9.67b	2.00b	359.33a
	23	(0.17)	(0.01)	(1.65)	(0.28)	(0.18)	(0.11)	(2.68)	(1.15)	(0.0)	(0.99)
db)	25	18.55c	0.68b	54.06b	13.66c	43.15b	0.79a	29.52b	2.77c	2.67a	388.78b
	33	(1.94)	(0.03)	(1.86)	(0.67)	(2.15)	(0.10)	(11.45)	(2.28)	(0.65)	(7.94)
	100	17.54a	0.67a	53.24a	13.00a	41.72a	0.70a	36.65a	4.17a	2.42a	378.65a
Temperature (°C)	100	(2.50)	(0.03)	(1.85)	(0.57)	(1.60)	(0.08)	(5.57)	(1.34)	(0.51)	(16.70)
	125	16.27b	0.64b	52.43a	12.64a	40.77a	0.75ab	43.83b	9.67b	2.00a	359.33b
	125	(0.17)	(0.01)	(1.65)	(0.28)	(0.18)	(0.11)	(2.68)	(1.15)	(0.0)	(0.99)
	150	16.19b	0.64b	52.80a	13.73b	42.54b	0.82b	33.37a	3.77a	3.08b	360.76b
	150	(1.73)	(0.03)	(2.35)	(0.64)	(2.13)	(0.16)	(13.67)	(3.17)	(0.51)	(27.97)
	50	16.42a	0.65a	52.21a	13.38a	41.68a	0.77a	38.10a	4.92a	2.83a	383.27a
	50	(1.52)	(0.03)	(1.59)	(0.84)	(1.88)	(0.08)	(4.78)	(2.54)	(0.72)	(12.46)
Screw Speed (rpm)	75	16.28ab	0.64a	52.43ab	12.64b	40.77a	0.75a	43.83a	9.67b	2.00b	359.33b
	15	(0.17)	(0.01)	(1.65)	(0.28)	(0.18)	(0.11)	(2.68)	(1.15)	(0.0)	(0.99)
	100	17.31b	0.67b	53.84b	13.35a	42.58b	0.75a	33.10b	3.02c	2.67a	361.77b
	100	(2.74)	(0.03)	(2.26)	(0.56)	(1.85)	(0.18)	(11.99)	(1.88)	(0.49)	(25.86)

*Averages and standard deviations (shown in parentheses) of the main effects of the DDGS extrudates. The relationships amongst the main effects are shown below. Differing letters indicate significant differences among levels of a specific independent variable ($\alpha = 0.05$) for a given dependent variable.

Table 7. Interaction effects for properties of DDGS extrudates*

Daramatar	Moisture Content	Water Activity		Color	
Palalletei	(% db)	(aw)	L*	a*	b*
Moisture Content	<.0001	<.0001	0.0076	0.0001	<.0001
Temperature	0.0007	0.0001	0.5319	<.0001	0.0285
Screw Speed	0.0152	0.0017	0.0301	0.7940	0.0163
Moisture Content*Temperature	0.0145	0.0173	0.2219	0.5380	0.0001
Moisture Content*Screw Speed	0.0006	0.2090	0.3458	0.0024	0.0154
Temperature*Screw Speed	0.6696	0.0043	0.9582	<.0001	0.0006
Moisture Content*Temperature*Screw Speed	0.2523	<.0001	0.2874	0.7624	0.1321
Darameter	Unit Danaity (a/am ³)	Pellet Durability	Water	Floatability	Bulk Density
	Unit Density (g/cm)	Index(%)	Stability (s)	(s)	(kg/m^3)
Moisture Content	0.3165	<.0001	<.0001	0.3979	<.0001
Temperature	0.0382	0.0352	0.2557	0.0028	0.0022
Screw Speed	0.6934	0.7084	<.0001	0.3979	<.0001
Moisture Content*Temperature	0.8308	<.0001	<.0001	1.0000	<.0001
Moisture Content*Screw Speed	0.6489	0.1258	0.5022	0.3979	0.1793
Temperature*Screw Speed	0.1904	0.0052	0.5022	0.1004	0.0001
Moisture Content*Temperature*Screw Speed	0.5479	-	0.0047	0.1004	-

* Relationships between the treatment interactions of the DDGS extrudates (p-values). Interactions were deemed significant if p < 0.05.

3.2.1 Moisture Content

The moisture content of the extrudates is important to consider because it determines the freshness of the pellets. Moisture also impacts the rate of spoilage of the extrudates. When developing a pellet for aquaculture feed, it is important to consider the amount of time the pellets will be stored, so the optimum amount of moisture present in the extrudates can be determined. According to Table 4, treatment 7, which had a screw speed of 100 rpm, raw moisture content of 35% db, and die temperature of 100 °C, had the greatest extrudate moisture content and was significantly different from the other eight treatments. Treatments 3 and 8 had similar moisture contents of 18.80% db and 18.82% db, respectively, and treatments 4 and 9 were similar as well, with moisture content of 15% db, and a die temperature of 150 °C, had the lowest extrudate moisture content of 15.52% db, and similar results to both treatments 4 and 9 and treatment 6, with extrudate moisture contents of 15.52% db, 15.32% db, respectively. According to Table 3, the greater the initial moisture content of the raw DDGS blends, the greater the moisture content in the extrudates. A similar relationship between the corn extrudates was determined as well. According to Table 6, as the temperature of the die increased, the moisture content of the DDGS extrudates decreased, due to increased evaporation. According to Table 6, as the screw

speed of the extruder increased from 50 to 100 rpm, the moisture content of the DDGS extrudates increased. According to Table 5, the amount of moisture present in the corn extrudates increased as the screw speed of the extruder increased; this was similar behavior as the DDGS.

3.2.2 Water Activity

Water activity determines the amount of water available for microbial metabolism and growth and is important in the extrudates because it determines the rate of spoilage. When comparing moisture content to water activity, the water activity is more important in the biological sense of spoilage because it is the amount of water available for microbial growth. According to Table 4, treatments 3, 7, and 8 have the greatest water activity levels, with values of 0.70, 0.68, and 0.69, respectively. With a value of 0.65, treatment 5 has a slightly smaller water activity value and is significantly different than treatments 3, 7, and 8. Treatments 1, 2, 4, and 6 have the smallest values for water activity, and treatment 9 has a value of 0.64, which is not significantly different from treatments 1, 2, 4, 5, and 6. According to Table 6, as the moisture content of the raw DDGS ingredients increased, the water activity of the extrudates increased. A similar relationship as seen in the corn extrudates and is shown in Table 5. As the temperature of the die of the extruder increased, the water activity in the DDGS extrudates decreased and is displayed in Table 6. Similar results are shown in Table 5 for the corn extrudates. As the screw speed of the extruder increased, there was an increase in water activity levels from the 50 and 75 rpm to the 100 rpm treatments. According to Table 5, as the screw speed of the extruder increased, the water activity level of the corn extrudates increased.

3.2.3 Color

In order to have consumer acceptance, the appearance of extrudates is important. Color is one of the most important appearance attributes because consumers buy products based on expected color from previous experience. The color of the extrudates is measured based on three parameters: L*, a*, and b*. According to Table 4, the L* value for the DDGS was greatest in treatment 7, with a value of 55.35, while treatment 2 had the smallest L* value at 50.45. Treatments 2, 4, 7, and 8 have higher a* values that are significantly different from treatments 1, 3, 5, 6, and 9 (Table 4). The b* values for treatments 4, 7, and 8 were the greatest and were significantly different from the other treatments. Treatments 3 and 6 have the lowest b* values of 40.03 and 40.25, respectively. As the moisture content of the DDGS extrudates increased, the L* and b* values increased, although there was no relationship with a* values, according to Table 6. A similar relationship was true for the DDGS extrudates increased, the a* and b* values of the DDGS extrudates increased, the L* and b* values of the DDGS extrudates increased, the L* and b* values of the DDGS extrudates increased, the L* and b* values of the DDGS extrudates increased, the L* and b* values of the DDGS extrudates increased, the L* and b* values of the DDGS extrudates increased, the L* and b* values of the DDGS extrudates increased, the L* and b* values of the DDGS extrudates increased, the L* and b* values of the DDGS extrudates increased, but there was no relationship with the L* values. As the screw speed of the extruder increased, the L* and b* values of the DDGS extrudates increased, but there was not a significant relationship with the a* values.

3.2.4 Unit Density

Unit density is important in the extrudates because the amount of mass per unit volume should contain the proper nutrients, in the proper quantities but be able to float in water (i.e., if $UD < 1.0 \text{ g/cm}^3$). According to Table 4, treatment 4 and 6 contained the greatest mass per unit volume, with values of 0.85 and 0.86 g/cm³, respectively, while treatment 5 produced the smallest unit density with a value of 0.62 g/cm³. Treatments 1, 2, 3, 7, 8, and 9 were similar to treatments 4, 5, and 6. According to Table 6, there was no significant relationship between the unit density of the DDGS extrudates and the change in moisture content or screw speed. However, as the temperature of the die increased, the unit density of the DDGS extrudates increased. Because all treatments had UD values less than 1.0 g/cm³, they should have been able to float in water (if they could exhibit water stability, that is).

3.2.5 Pellet Durability Index

Pellet durability index is the amount of breakage the extrudate is able to withstand. It is important in aquaculture feeds because of storage and delivery processes. According to Table 4, the greatest value for the pellet durability index was treatment 6 at 47.90%. Treatments 1 and 3 were relatively similar to both treatment 9, as well as treatments 2 and 5. Treatment 7 was similar to treatments 2 and 5 as well. The lowest pellet durability index was treatment 8, at 17.33%. The pellet durability index for treatment 4 was unable to be determined due to the lack of cohesive of extrudates for this treatment, thus PDI could not be reported. According to Table 6, as the moisture content of the DDGS extrudates increased, the pellet durability index decreased. There was no significant relationship between the temperature of the DDGS extrudates and the pellet durability index values. As the screw speed of the extruder increased, the pellet durability index decreased in the DDGS extrudates.

3.2.6 Water Stability

Water stability is the amount of time the pellet holds together in water before it dissolves. It is important for aquaculture feeds because the fish need pellets to last approximately 30 min during feeding time. According to Table 4, the greatest value for water stability was treatment 9, which was the center point. Treatment 9 was followed by treatment 2, and then treatment 3, at 8 and 6 s, respectively. Treatment 6 was relatively similar to both treatment 3 and treatments 1 and 5. Treatments 1 and 5 were also relatively similar to treatment 3, and treatment 3 was relatively similar to treatment 4. Treatment 8 produced the lowest water stability with an average of 0.40 s. According to Table 6, the moisture content of the raw ingredients, temperature of the die, and screw speed of the extruder did not result in significant relationships for the water stability values of the DDGS extrudates. Because DDGS contains very little starch, the pellets lack a binder typically provided by gelatinization of starch. According to Table 5, the increase in moisture content caused an increase in water stability of the corn extrudates, but corn contains large starch content. An increase in both the screw speed of the extruder and the temperature of the die caused a decrease in the water stability of the corn extrudates. It is clear that in order to be effectively used in aquafeeds, DDGS cannot be used alone; instead it must be processed with other ingredients which, hopefully, will improve particle binding characteristics.

3.2.7 Floatability

Floatability is the amount of time the pellets float on top of the water. It is important in aquaculture feeds because some species only eat the feed if it is floating, and the pellets must float for approximately 30 min for appropriate feeding time. According to Table 4, the greatest floatability was produced by treatments 2 and 4 at 3.33 s, while the lowest floatability was produced by treatments 3 and 9 at 2 s. Treatments 1, 5, 6, 7, and 8 were all relatively similar to one another. There was no direct relationship between the moisture content and screw speed and the floatability of the DDGS extrudates, according to Table 6. As the temperature of the extruder increased, the floatability of the DDGS extrudates increased, due to greater expansion. According to Table 5, as the moisture content of the raw corn blends increased, the floatability of the corn extrudates decreased. As the screw speed and temperature of the extruder increased, the floatability of the corn extrudates decreased (Table 5).

3.2.8 Bulk Density

The bulk density quantifies the amount of mass a specific volume can hold. Bulk density is important in storage of aquaculture feeds because the pellets will be distributed in containers, and the containers need to hold as much mass as possible. According to Table 4, the greatest bulk density was produced by treatment 3 at 399.00 kg/m³. Treatments 7 and 8 followed treatment 3, having slightly lower bulk density. Treatment 1 was relatively similar to treatments 7 and 8, as well as treatment 2. Treatment 5 had a bulk density of 355.00 kg/m³, which was slightly lower than treatment 2. The smallest value for bulk density was produced by treatment 6, with a value of 324.73 kg/m³. According to Table 6, as the moisture content of the raw DDGS blend increased, the bulk density of the DDGS increased. The bulk density of the DDGS extrudates decreased as the temperature and screw speed of the extruder increased, due to greater expansion. According to Table 5, similar results were produced for the corn extrudates.

3.3 Implications

Figure 2 is a matrix plot of all independent and dependent variables, and it shows that the extruded DDGS had much different behavior than the extruded corn for all properties studied. This was true for all screw speeds, all ingredient moistures, and all extruder temperatures. This occurred due to the chemical composition being substantially different. As Liu and Rosentrater (2011) discussed, DDGS has approximately three times the levels of protein, fiber, fat, and minerals vis-àvis corn, whereas DDGS has substantially lower starch (because alcohol manufacturing utilizes only the starch component from the corn kernels). Use of DDGS vs. corn in aquafeed formulations definitely impacts extrudate quality, as shown in Figure 2.



Figure 2. Matrix plot of all independent and dependent variables shows that the extruded DDGS had much different behavior than the extruded corn for all properties studied.

4. Conclusion

In conclusion, the moisture content of the raw ingredients led to an increase in the water activity in the raw ingredients. It also led to an increase in moisture content, water activity level, L* values, b* values, and bulk density of the DDGS extrudates. An increase in moisture content led to a decrease in pellet durability index. There was no significant relationship between the moisture content of the raw ingredients and the a* values, unit density, water stability, or floatability of the DDGS extrudates. The increase in temperature of the extruder die led to an increase in the a* values, b* values, unit density, and floatability of the DDGS extrudates. An increase in the temperature of the die led to a decrease in the moisture content, water activity level, and bulk density of the DDGS extrudates. There was no significant relationship between the temperature of the die and the L* values, pellet durability index, or water stability of the DDGS extrudates. The increase in screw speed caused an increase in the moisture content, water activity level, L* values of the DDGS extrudates. An increase in the screw speed of the extruder caused a decrease in the pellet durability index and bulk density of the extrudates. There was no significant relationship between the screw speed of the extrudates, and increase in the screw speed of the extruder caused a decrease in the pellet durability index and bulk density of the DDGS extrudates. There was no significant relationship between the screw speed of the extruder and the a* values, unit density, water stability, and floatability of the DDGS extrudates.

Ultimately, the composition of the ingredients affects the resulting physical properties of the extrudates. The amount of starch and water present in the ingredients determines the quality of the pellets. Because starch gelatinizes and acts as a binding agent, the more starch present, the better the quality of the pellet. Along with starch content, the amount of fiber and protein in the ingredients affects the quality of the extrudates. It is important to have a high protein diet for aquaculture feed, so the fish reach substantial weight in a reasonable amount of time. The key is to find an appropriate balance between moisture content, screw speed, and temperature, as well as the starch, protein, and fiber content of the ingredients. It is clear that DDGS cannot be processed alone at the extrusion conditions used in this study. DDGS should be used in combination with other ingredients in order to balance nutrient levels as well as improve feed quality characteristics.

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