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Extrusion Studies of Aquaculture Feed using Distillers Dried Grains with Solubles and Whey

Nehru Chevanan • Kasiviswanathan Muthukumarappan • Kurt A. Rosentrater

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Abstract Three isocaloric (3.5 kcal/g) ingredient blends containing 20, 30, and 40% distiller-dried grains with solubles (DDGS) along with 5% whey were prepared with a net protein content adjusted to 28% (wet basis [wb]). Other ingredients in the blends included soy flour, corn flour, fish meal, vitamin, and mineral mix. These blends were extruded in a single-screw extruder at 15, 20, and 25% (wb) moisture content and at 130 and 160 rpm screw speeds. Compared to previous research, the durability and unit density of the extrudates in this study were found to increase substantially by the addition of whey to the blends. Increasing the DDGS content from 20 to 40% resulted in a 5.8 and 16.8% increase in extrudate moisture content and redness, respectively, but produced a decrease of 11.2% in brightness and 3.6% in yellowness of the extrudates. Increasing the moisture content of the ingredient blends from 15 to 25% resulted in an increase of 16.1, 8.7, and 9.3% in moisture content, durability, and redness, respectively, but a decrease of 9.8 and 5.6%, respectively, in brightness and yellowness of the extrudates. Neither DDGS level nor screw speed significantly affected extrudate

N. Chevanan Biosystems Engineering and Soil Science, University of Tennessee, Knoxville, TN 37996, USA

K. Muthukumarappan Department of Agricultural and Biosystems Engineering, South Dakota State University, Brookings, SD 57007, USA

K. A. Rosentrater (⊠)
North Central Agricultural Research Laboratory, ARS, USDA, 2923 Medary Ave.,
Brookings, SD 57006, USA
e-mail: kurt.rosentrater@ars.usda.gov

durability or unit density. In fact, changing the screw speed had no significant effect on many of the properties of the extrudates studied, except for moisture content, redness, and yellowness. As demonstrated in this study, ingredient moisture content and screw speed are critical considerations when producing extrudates with feed blends containing DDGS; further work is needed to optimize processing conditions and to produce floating feeds.

Keywords DDGS · Distillers grains · Extrusion ·

Pellet durability index \cdot Single-screw extruder \cdot Unit density \cdot Water activity \cdot Whey

Introduction

Distillers dried grains with solubles (known as DDGS), is a coproduct of ethanol manufacturing and generally contains relatively high levels of protein, fat, and fiber but low starch. It is mostly used as livestock feed-primarily beef and dairy but also swine and poultry. Fish feed also requires high quantities of protein. Moreover, the protein conversion efficiency of fish is very high compared to these other, land-based animals. Hence, DDGS has great potential to be used as an alternative protein source for aquaculture feed production vis-à-vis fish meal. Initial feeding studies have been conducted on tilapia (Wu et al. 1994, 1996; Coyle et al. 2004), on trout (Cheng et al. 2003; Cheng and Hardy 2004a, b), and on catfish (Webster et al. 1993) and have indicated that fish can be successfully grown with DDGS as a protein source. In fact, the use of DDGS may improve the economic viability of aquaculture farms. Because starch is converted to ethanol during the fermentation process, micronutrients in DDGS are available in concentrated quantities compared to whole corn. Depending on the fish

species and the age/size, aquaculture feeds typically require 26 to 50% protein.

Extrusion cooking is widely used in the food and feed industries because of versatility during processing and the ability to produce various finished textural properties (Mercier et al. 1989). For aquaculture rations, floatability, durability, and water stability are key physical properties that impact the quality of the feeds (Bandypadhyay and Rout 2001; Rolfe et al. 2001). The extent of expansion obtained is very important for producing floating feeds. Additionally, the ingredient chemical composition, extent of gelatinization during processing, and protein denaturation will all affect the durability and water stability, as well as other physical properties (Case et al. 1992; Ibanoglu et al. 1996; Thomas et al. 1999). At commercial aquaculture feed production plants, fat is often added to the extrudates by enrobing to improve water stability and energy content (Chang and Wang 1998).

During extrusion processing of starch-based products, an elastic melt is formed inside the barrel, which results in a more expanded final product (Ilo et al. 1996; Alves et al. 1999; Lin et al. 2000). However, during extrusion processing of protein-based products, a plastic melt is formed, which ultimately results in a more porous and textured final product (Cumming et al. 1973; Gwiazda et al. 1987; Singh et al. 1991; Sandra and Jose 1993). Moisture content (MC) of the mix as well as temperature gradient in the barrel are two important factors that affect the development of proper viscosity of the melt and also impact the final product characteristics (Kokini et al. 1992; Chevanan et al. 2007a, c). Aquaculture feeds require high proportions of both protein and starch, but very little information is currently available on extrusion processing of these types of ingredient blends.

Because aquaculture feeds require high amounts of protein (often 26 to 50%), these formulated feeds will typically contain high quantities of both protein and starch. Although DDGS contains a fairly high proportion of protein, the amount of DDGS incorporated into the feed blend is often limited because of its low starch and high fiber contents (Chin et al. 1989). As the starch content is decreased, the expansion obtained during extrusion is typically reduced, which negatively affects the resulting floatability of the extrudates. On the other hand, as the fiber content increases, the mechanical strength will decrease because of its noninteracting nature with other chemical components in the ingredient blend, and it tends to produce ingredients that do not readily bind together. If DDGS is going to be successfully used for aquaculture feeds, these challenges must be overcome. Binders are often used to overcome issues with cohesiveness.

Whey is a byproduct of cheese making and has excellent binding properties. In addition, whey can be used as a binder for aquaculture feed (Lovell 1988). However, the inclusion of whey during extrusion processing of cereal foods has been found to produce challenges during processing, such as decreasing the expansion and increasing the unit density and water absorption index (Martinez-Serna and Villota 1992; Matthey and Hanna 1997). The exact mechanisms of the interactions between whey and other components are not yet well understood. Even so, whey may be an appropriate binder for DDGS-based aquaculture feeds.

To viably incorporate DDGS into aquaculture feeds, the impact of addition of whey as a binder on extrudate quality, especially strength and cohesiveness, as well as other physical properties, must be determined. Consequently, the objective of this study was to use single-screw extrusion processing to examine the effects of various levels of DDGS, along with soy flour, corn flour, fish meal, vitamin, and mineral mix, as well as whey as a binder, on resulting extrudate properties.

Materials and Methods

Feed Blend Formulation

Three nutritionally balanced isocaloric (3.5 kcal/g) ingredient feed blends were formulated to an isonitrogenous net target protein of 28% wet basis (wb), using three levels of DDGS: 20, 30, and 40% DDGS. Additional ingredients used in each of the blends included soy flour, corn flour, Menhaden fish meal, whey, vitamin mix, and mineral mix (Table 1). The corn flour was provided by Cargill Dry Ingredients (Paris, IL), and soy flour was provided by Cargill Soy Protein Solutions (Cedar Rapids, IA). The vitamin mix, mineral mix, fish meal, and whey were obtained locally and were mixed at 1, 2, 6, and 5%, respectively, on an as-is basis (i.e., wb). DDGS was obtained from Dakota Ethanol LLC (Wentworth, SD) and

Table 1 Ingredient components in the prepared feed blends

Feed ingredients	Mass of ingredients (g/100g)							
	Blend I	Blend II	Blend III					
DDGS	20	30	40					
Soy flour	31	27	23					
Corn flour	35	29	23					
Fish meal	6	6	6					
Mineral mix	1	1	1					
Vitamin mix	2	2	2					
Whey	5	5	5					
Total	100	100	100					

was ground to a particles size of approximately $100 \ \mu m$ using a laboratory grinder (Model S500 disc mill, Genmills, Clifton, NJ). The ingredients were then mixed with appropriate quantities of water in a laboratory-scale mixer (Hobart Corporation, Troy, OH) for 10 min and stored overnight at refrigerated conditions to achieve moisture stabilization.

Experimental Design and Extrusion Processing

The extrusion studies were carried out using a singlescrew extruder (Model PL 2000, Brabender, South Hackensack, NJ), which had a barrel length of 317.5 mm and a length-to-diameter ratio of 20:1. The die assembly had an internal conical section and had a length of 101.6 mm. A screw with a uniform pitch of 9.05 mm was used in the experiments. The screw had variable flute depth, with a depth at the feed end of 9.05 mm and that near the die of 3.81 mm. The compression ratio achieved inside the barrel was 3:1. The speed of the screw and the temperature inside the barrel were controlled by an external computer. The extruder barrel had band heaters, so that the temperature in the feed zone, transition zone in the barrel, and the die section could be controlled. Compressed air cooling was provided in the barrel section but not the die section. The extruder had a 7.5-hp motor, and the computer could control the speed of the screw from 0 to 210 rpm (0 to 21.99 rad/s).

Experiments were conducted using a full-factorial design with three levels of DDGS (20, 30, and 40%), three levels of ingredient MC (15, 20, and 25% wb), and two levels of screw speed (130 and 160 rpm [13.61 to 16.76 rad/s]). During all experiments, the temperature of the feed zone was maintained at 90°C, while that of the transition zone and the die section were maintained at 120°C.

Measurement of Physical Properties

Moisture Content

The extrudates were cooled and air dried at room temperature (i.e., ambient conditions—approximately $25\pm$ 1°C) for 5 days. MC of the dried extrudates was then determined following ASAE (2004) Method S352.2, using a forced-convection laboratory oven (Thelco Precision, Jovan, Wincester, VA) at 103°C for 72 h. Two measurements were made for each experimental run.

Water Activity

Water activity was measured using a calibrated water activity meter (AW Sprint TH 500, Novasina, Talstrasse, Switzerland). Four measurements were made for each experimental run.

Durability

Pellet durability index (PDI, %) was determined per ASAE (2004) Method S269.4. Approximately 200 g of the extrudates were tumbled inside a pellet durability tester for 10 min and then sieved via a no. 6 (3.4-mm) screen. The resulting pellet durability index was then calculated as:

$$PDI = \left(\frac{M_{a}}{M_{b}}\right) \times 100 \tag{1}$$

where M_a is the mass of the extrudates retained on the screen after tumbling (g) and M_b is the mass of extrudates retained on the screen before tumbling (g). Two measurements were made for each experimental run.

Unit Density

Extrudate samples were cut to 2-cm lengths, weighed on an electronic balance (Model A-250, Denver Instrument, Arvada, CO), and their diameter was measured using a digital calipers (Digimatic Series no. 293, Mitutoyo, Tokyo, Japan). Unit density was calculated as the ratio of the mass of each 2-cm extrudate piece to the calculated volume of that piece (assuming cylindrical shapes for each extrudate sample) following Jamin and Flores (1998). Four measurements were made for each experimental run.

Color

Color of the extrudates was determined using a spectrophotocolorimeter (Model CM 2500d, Minolta, Japan) using the Hunterlab color space. L^* value quantified the brightness/darkness of the extrudates, a^* value depicted the redness/greenness, and b^* value denoted the yellowness/ blueness. Four measurements were made for each experimental run.

Nutrient Analysis

The extrudates were dried, and then protein, fiber, fat, and ash contents were determined in duplicate (for each treatment combination) following official Method 990.03, 978.10, 920.39, and 920.48, respectively (AOAC 2003). The nitrogen-free extract (NFE) was then determined by difference.

Statistical Analysis

The collected data were analyzed with two-way analysis of variance by general linear models using the Proc GLM procedure in SAS (2004) version 8 (SAS Institute, Cary, NC), with a type I error rate (α) of 0.05, to determine the main and interaction effects and least significant differences between treatment combinations.

Results and Discussion

Considering main effects (i.e., each independent variable) only, changing the incorporation levels of DDGS had a significant effect on all the properties studied except durability and unit density (Table 2). Likewise, changing the MC had a significant effect on all the properties studied. Changing the screw speed, on the other hand, had a significant effect only on the final extrudate MC, Hunter a, and Hunter b, but not on the other properties. Furthermore, the interaction effects because of the levels of DDGS, MC, and screw speed were found to be significant for most of the properties studied (Table 3). The nature and extent of expansion occurring because of water vapor depends on the constituents present in the ingredient mix, on the resulting melt viscosity, and on the biochemical changes that occur in the extruder, in other words, interactive effects. Thus, the simultaneous alterations in the treatment combinations influenced the changes that occurred in the matrix during processing. Further discussion regarding the interactions (i.e., treatment combination effects) follows.

Moisture Content

Ingredient MC has a strong influence on final extrudate properties, as it impacts processing behavior (Rolfe et al. 2001). Furthermore, the resulting MC of the extrudates is very important as it affects the shelf life of the manufactured feeds. The various thermomechanical and biochemical changes occurring inside the barrel will result in changes in the nature of bound vs unbound water, which will be present in the extrudates, as well as the total moisture present. The maximum (10.02% wb) and minimum (7.09% wb) MCs of the dried extrudates were achieved at 30% DDGS, 20% MC, and 130 rpm and 40% DDGS, 20% MC, and 160 rpm, respectively (Table 4). The MC of the original DDGS used in preparing the blends was 8.8% (wb), which was the highest of all the components in the ingredient mix. Increasing the DDGS content from 20 to 40% resulted in a 5.8% increase in the final MC of the extrudates (Table 2). Increasing the screw speed from 130 to 160 rpm resulted in a 2.6% reduction in MC of the extrudates. Furthermore, increasing the MC of the ingredient blends from 15 to 25% resulted in a 16.1% increase in the MC of the extrudates. The high temperature and shear conditions inside the extruder affect the complex interactions between water and the other chemical constituents and alter the cellular structures that result at the die exit when the water flashes into steam (Miller 1985).

Water Activity

Water activity is a measure of the free water present in materials. The higher the water activity, the greater the chance of rapid microbial spoilage, which will result in reduced storage stability. The maximum (0.58) and minimum (0.42) water activity was achieved at 30% DDGS, 20% MC, and 130 rpm and 40% DDGS, 20% MC, and 160 rpm, respectively (Table 4). These are reflective of the behavior found with extrudate final MC as well. Increasing

Table 2 Main effects of DDGS, screw speed, and moisture content on the properties of extrudates⁺

	Moisture content (% wb)	SD	Water activity	SD	Unit density (g/cm ³)	SD	PDI (%)	SD	Color L* value	SD	Color a* value	SD	Color b* value	SD
DDGS	(% wb)													
20	7.78 ^a	0.70	0.47^{a}	0.04	1.05	0.06	94.04	1.13	48.54 ^a	3.88	4.28 ^a	0.57	15.13 ^a	0.76
30	8.61 ^b	0.97	0.50°	0.05	1.07	0.05	94.02	1.48	47.25 ^b	5.66	4.43 ^b	0.75	14.50 ^b	0.47
40	8.23 ^c	0.87	0.48 ^b	0.04	1.06	0.05	93.52	2.27	43.09 ^c	4.40	5.00 ^c	0.49	14.59 ^b	0.97
Screw	speed (rpm))												
130	8.31 ^a	0.99	0.48	0.05	1.07	0.06	93.59	2.09	46.10	6.44	4.49 ^a	0.81	14.32 ^a	0.56
160	8.09 ^b	0.80	0.48	0.04	1.06	0.05	94.14	1.08	46.49	3.64	4.65 ^b	0.52	15.16 ^b	0.79
Moistu	ire content (% wb)												
15	7.22 ^a	2.31	0.47 ^a	0.04	1.05 ^a	0.05	86.21 ^a	1.93	49.75 ^a	3.73	4.28 ^a	0.69	15.24 ^a	0.79
20	7.77 ^b	2.59	0.49 ^c	0.06	1.09 ^b	0.06	87.24 ^a	1.61	44.28 ^b	5.24	4.75 ^b	0.57	14.59 ^b	0.80
25	8.38 ^b	0.52	0.48 ^b	0.03	1.05 ^a	0.04	93.68 ^b	1.34	44.86 ^b	4.81	4.68 ^b	0.70	14.39 ^c	0.56

⁺ Values with the same letter for a given property, within each independent variable, are not significantly different (p < 0.05) for that dependent variable; SD denotes 1 standard deviation.

Variable	df	Moisture content (% wb)	Water activity	PDI (%)	Unit density (g/cm ³)	Color L* value	Color a* value	Color b* value
DDGS	2	0.0001	0.0001	0.3362	0.3526	0.0001	0.0001	0.0001
MC	2	0.0001	0.0001	0.0256	0.0211	0.0001	0.0001	0.0001
SS	1	0.0252	0.9048	0.0963	0.3211	0.1182	0.0001	0.0001
DDGS×MC	4	0.0004	0.0001	0.0030	0.3923	0.0001	0.0001	0.0007
DDGS×SS	2	0.0029	0.0001	0.0029	0.7321	0.0001	0.0002	0.0001
MC×SS	2	0.0001	0.0001	0.0128	0.2846	0.0001	0.0001	0.0001
DDGS×MC×SS	4	0.0001	0.0001	0.0015	0.7493	0.0001	0.0001	0.0001

Table 3 Interaction results (p values) for DDGS, moisture content and screw speed on the properties of the extrudates^a

^aA significance level of α =0.05 was used.

the DDGS content from 20 to 30% resulted in a 6.4% increase in water activity, but increasing the DDGS from 30 to 40% resulted in a 4.0% reduction in the water activity of the extrudates (Table 2). On the other hand, increasing the screw speed from 130 to 160 rpm did not significantly affect the water activity of the extrudates. Increasing the MC of the ingredient blends from 15 to 20% resulted in a 4.3% increase in the water activity of the extrudates, but increasing the MC further from 20 to 25% corresponded to a 0.2% decrease in the water activity of the extrudates (Table 2). Chevanan et al. (2007b) also found that water activity of extrudates can be influenced by both DDGS level and feed ingredient MC. These factors will impact the macroscopic and microscopic structure of the extrudates and will influence the proportion of bound vs free water in the resulting extrudates.

Durability

Mechanical strength and toughness dictate the stability of extrudates during transportation, storage, and feeding. The strength of extrudates depends to a large degree on the extent of heat treatment, the relative degree of starch transformation that occurs inside the barrel during this heating, and ultimately on the resulting cohesiveness of the chemical constituents in the extrudates (Colonna et al. 1989). The mechanical strength of pelleted feed materials can be indirectly assessed by PDI (Rosentrater et al. 2005), which is an important quality parameter of aquaculture and other livestock feeds. For the ingredient blends used in this study, changing the level of MC had a significant effect on extrudate PDI (Table 2). Increasing the MC from 15 to 25% resulted in a 8.7% increase in extrudate PDI. This behavior

Table 4 Treatment combination effects due to DDGS, screw speed, and mositure content levels on extrudate properties⁺

Property	DDGS content (%wb)	15% MC (wb)		20% MC (wb)		25% MC (wb)		
		130 rpm	160 rpm	130 rpm	160 rpm	130 rpm	160 rpm	
Moisture content	20	7.19 ^{j-k} (0.02)	7.16 ^{j-k} (0.12)	8.20 ^{d-g} (0.09)	7.35 ^{i-k} (0.01)	7.76 ^{f-i} (0.36)	$9.00^{b-c} (0.03)$	
(% wb)	30	7.45^{h-k} (0.04)	9.48^{a-b} (0.09)	10.02^{a} (0.05)	8.09^{d-g} (0.11)	8.43^{d-e} (1.04)	8.17^{d-g} (0.06)	
	40	7.70 ^{g-j} (0.01)	7.92^{e-h} (0.09)	$9.75^{a}(0.02)$	$7.09^{k} (0.03)$	8.31 ^{d-f} (0.04)	8.59 ^{c-d} (0.06)	
Water activity	20	0.44^{j-k} (0.01)	0.45^{h-j} (0.01)	0.48^{e-g} (0.01)	0.46^{f-i} (0.04)	0.46^{f-i} (0.02)	$0.53^{\rm c}$ (0.01)	
-	30	0.46 ^{g-j} (0.01)	0.56^{a-b} (0.01)	0.58^{a} (0.01)	0.47 ^{e-h} (0.01)	0.45^{h-j} (0.01)	0.47^{e-h} (0.03)	
	40	0.44^{i-k} (0.01)	$0.48^{d-f} (0.01)$	0.55^{b-c} (0.02)	0.42^{k} (0.01)	0.49^{d-e} (0.01)	$0.50^{d} (0.01)$	
Color L*	20	54.11 ^a (0.11)	47.63 ^e (1.19)	41.87 ^g (2.22)	49.67 ^{b-c} (1.41)	48.75 ^{c-e} (1.54)	49.24^{b-d} (0.67)	
	30	55.08 ^a (1.52)	48.07 ^{d-e} (0.89)	38.82^{h-i} (1.20)	49.65^{b-c} (0.59)	50.30 ^b (1.00)	41.61 ^g (0.50)	
	40	48.26^{c-e} (0.83)	45.37 ^f (0.35)	37.69 ⁱ (0.50)	47.97 ^{d-e} (0.98)	40.03 ^h (0.57)	39.25 ^h (0.62)	
Color a*	20	3.50 ^j (0.03)	4.97 ^e (0.21)	5.02 ^{d-e} (0.20)	4.04^{h-i} (0.09)	4.09 ^{g-h} (0.20)	4.07 ^{g-i} (0.05)	
	30	3.40 ^j (0.04)	4.35 ^f (0.12)	$5.50^{a} (0.18)$	4.25^{f-g} (0.15)	3.88 ⁱ (0.13)	$5.20^{c-d} (0.17)$	
	40	$4.29^{\rm f}$ (0.05)	5.17 ^{c-d} (0.07)	5.28^{b-c} (0.08)	4.43 ^f (0.10)	5.45 ^{a-b} (0.15)	5.41 ^{a-b} (0.13)	
Color b*	20	14.89 ^{d-e} (0.11)	16.47 ^a (0.39)	14.21^{h-i} (0.33)	15.19 ^{c-d} (0.33)	14.82^{d-f} (0.48)	15.19 ^{c-d} (0.23)	
	30	14.82^{d-f} (0.24)	14.76 ^{d-g} (0.37)	14.35 ^{g-i} (0.60)	14.82^{d-f} (0.18)	13.91 ⁱ (0.33)	14.37^{f-h} (0.32)	
	40	14.46^{e-h} (0.18)	16.02^{a-b} (0.17)	13.39 ^j (0.18)	15.60^{b-c} (0.36)	14.06^{h-i} (0.32)	14.01^{h-i} (0.34)	

⁺ Values with the same letter for a given property are not significantly different (p < 0.05) for that dependent variable among the treatment combinations; values inside the parenthesis are 1 standard deviation.

because of moisture has been noted by others as well (Bandyopadhyay and Rout 2001; Rolfe et al. 2001; Shukla et al. 2005). However, changing the levels of DDGS and screw speed did not significantly impact the PDI values. The pellet durability of the extrudates containing DDGS was not affected by the screw speed in twin-screw extrusion processing either (Chevanan et al. 2007b). For the current experiment, the PDI values of the extrudates ranged from 89.74 to 96.37%, with an average of 93.86% (Fig. 1). The maximum and minimum PDI values were achieved at 40% DDGS, 20% MC, and 130 rpm and 40% DDGS, 15% MC, and 130 rpm, respectively (in another experiment with the same formulations, using the same experimental conditions but without whey incorporation, the resulting PDI values ranged from 0.37 to 0.97, with an average of 0.70 [this data is not presented in this paper-see Chevanan et al. 2005 for more information]). Thus, the current results indicate that the addition of whey did improve the binding properties of the ingredient blends and substantially improved extrudate cohesion and, ultimately, durability.

Unit Density

Unit density is another important property of aquaculture feeds; it dictates whether an extrudate will float or sink. It is directly related to the expansion that occurs at the die exit (Colonna et al. 1989). The unit density of the extrudates containing 20% moisture was 3.8% greater than the extrudates containing 15% moisture, but then increasing the moisture to 25% resulted in a 3.7% decrease (Table 2).

On the other hand, changing the DDGS level and screw speed had no significant effect on the unit density of the extrudates. These results parallel those found by Chevanan et al. (2007b). The maximum and minimum unit density values were achieved at 30% DDGS, 20% MC, and 130 rpm and 20% DDGS, 25% MC, and 160 rpm, respectively. For the current experiment, considering all treatment combinations, the unit density values of the extrudates ranged from 1.02 to 1.13 g/cm³, with an average unit density of 1.06 g/cm³. Because the extrudates all had unit density values greater than 1.0 g/cm³, they actually did not float-which is necessary for aquaculture feeds. Thus, the treatment combinations used in this study did not produce extrudates with appropriate functionality for fish. Of course, these responses may change when scaling up to a commercial extruder (in another experiment with the same formulations, using the same experimental conditions but without whey incorporation, the resulting unit density values ranged from 0.81 to 1.05 g/cm^3 , with an average of 0.94 g/cm³; thus, many of those extrudates floated [this data is not presented in this paper-see Chevanan et al. 2005 for more information]). The minimum unit density observed at 20% DDGS was probably due to the interactions between the chemical constituents, such as starch and fiber contents, but also due to the whey acting as binder, which held the components together and interacted with the moisture in the blend. The higher unit density at the 30% DDGS content might have been due to the biochemical changes brought about by the starch content in the ingredient mix interacting with the whey, as well as whey binding with other chemical

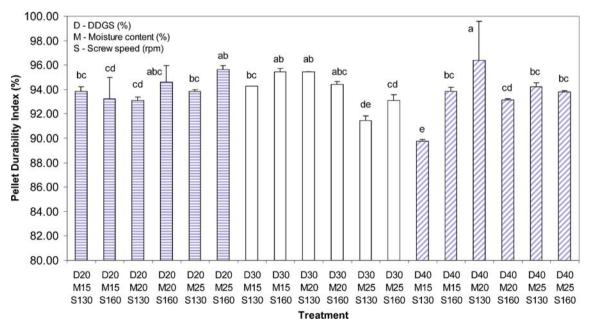


Fig. 1 Effect of DDGS, moisture content, and screw speed on pellet durability index of the extrudates (± 1 standard deviation). Bars with different letters were significantly different (p < 0.05). Columns are color coded according to the DDGS level

constituents. These results indicate that the whey contributed not only to binding but also toward the expansion and ultimately the unit density of the extrudates as well, which is in accordance with the results of Peri et al. (1983) and Martinez-Serna and Villota (1992) for materials containing whey. Hence, further studies are necessary to optimize the inclusion level of whey, which will result in higher PDI but hopefully without increasing the unit density of the extrudates.

Color

The color of extrudates is important because loss of color can be an indication of either a loss of lysine, which is an important amino acid that is required for aquaculture and other livestock feeds, or an indication of damage to the protein, which degrades digestibility. During extrusion processing, lysine losses up to 40% have been observed (Bjorck and Asp 1983). In this study (Table 4), the maximum (55.08) and minimum (37.69) brightness was achieved at 30% DDGS, 15% MC, and 130 rpm and 40% DDGS, 20% MC, and 130 rpm, respectively. The maximum (5.50) and minimum (3.40) redness/greenness was achieved at 30% DDGS, 20% MC, and 130 rpm, and 30% DDGS, 15% MC, and 130 rpm, respectively. The maximum (16.47) and minimum (13.39) yellowness/blueness was achieved at 20% DDGS, 15% MC, and 160 rpm and 40% DDGS, 20% MC, and 130 rpm, respectively. As the DDGS content was increased from 20 to 40%, the brightness of the extrudates reduced by 11.2%; redness increased by 16.8%, and the yellowness decreased by 3.6% (Table 2). This behavior was expected because DDGS was dark vellowish-brown in color, and increasing the DDGS content should result in darker, browner products. Increasing the screw speed did not significantly affect the brightness of the extrudates, but it did increase both the redness and the yellowness, by 3.6 and 5.9%, respectively. Increasing the MC of the ingredient mix from 15 to 25% resulted in a 9.8% decrease in brightness, a 9.3% increase in redness, and a 5.6% decrease in yellowness. Shukla et al. (2005) also observed a similar trend in color during extrusion processing of DDGS-based ingredient blends containing DDGS. Increasing the MC may have increased the temperature of the ingredient melt inside the barrel, which could have additionally altered extrudate color (via biological and compositional changes during processing because of the high temperatures and shear rates involved).

Nutrient Content

As anticipated, neither screw speed nor MC altered the proximate composition of the ingredient blends. Increasing the DDGS content from 20 to 40% had no significant effect on the protein content of the extrudates (Table 5) either, which was expected as all the blends were formulated to a constant protein level. Further examination revealed that increasing the DDGS content from 20 to 40% did not result in significant changes in extrudate fiber, fat, ash, or NFE content either, which was anticipated because of the mass balances used to formulate the blends. Although outside the bounds of this study, it would be interesting to investigate the potential interactions between whey and the other chemical constituents vis-à-vis binding properties.

Conclusions

Extrusion studies were conducted using a single-screw laboratory-scale extruder with three ingredient blends containing 20, 30, and 40% DDGS, each containing 5% whey as a binder. To provide a balanced ration for each blend, the other ingredients included soy flour, corn flour, fish meal, and mineral and vitamin mixes. The net protein content of each blend was adjusted to 28%. Quality parameters were studied on the resulting extrudates, including durability, unit density, MC, water activity, color, and nutrient content. The addition of whey resulted in increased durability and unit density of the extrudates (compared to equivalent blends without whey) because of its unique binding properties. Increasing the DDGS content resulted in an increase in extrudate MC, water activity, and redness, but a decrease in brightness and yellowness. Increasing the ingredient MC resulted in an increase in MC, water activity, and durability but a decrease in

Table 5 Nutrient contents of extrudates with different levels of DDGS $(n=2 \text{ for each treatment combination})^+$

DDGS level (% wb)	Protein (% db)	Fiber (% db)	Fat (% db)	Ash (% db)	NFE (% db)
20	29.60 (0.32)	4.36 (0.77)	4.13 (0.92)	7.27 (0.17)	54.63 (1.56)
30	29.71 (0.49)	4.25 (0.42)	3.62 (0.75)	7.20 (0.18)	55.21 (1.57)
40	29.59 (0.95)	4.71 (1.02)	4.18 (1.19)	7.15 (0.10)	54.38 (2.72)

⁺ Values with the same subscript for a given property are not significantly different (p < 0.05) among the blends; values inside the parenthesis are 1 standard deviation.

extrudate brightness and yellowness. Screw speed had no significant effect on the many of the properties studied, except for MC, redness, and yellowness. Overall, our results indicate that DDGS can be incorporated in aquaculture feed mixtures up to 40%, although these blends did not completely satisfactorily meet the needs of typical aquaculture feeds. Inclusion of 5% whey protein increased the pellet durability substantially compared to previous studies that did not use a binder, although the higher unit density observed for these extrudates is disadvantageous to production of fish feed, as the extrudates did not float. Further studies are necessary to simultaneously optimize the inclusion level of DDGS and whey to obtain extrudates, which are nutritionally appropriate, durable, floatable, and thus can be used as actual aquaculture feed. Additionally, optimization work must be conducted on the extrusionprocessing conditions because not only are floating, cohesive pellets required, but protein damage must also be minimized.

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