Twin-Screw Extrusion Processing of Feed Blends Containing Distillers Dried Grains with Solubles (DDGS)

Nehru Chevanan,[1] Kurt A. Rosentrater,[2,3] and Kasiviswanathan Muthukumarappan[4]

ABSTRACT

Cereal Chem. 84(5):428–436

Extrusion trials were conducted with varying levels of distillers dried grains with solubles (DDGS) along with soy flour, corn flour, fish meal, vitamin mix, mineral mix, and net protein content adjusted to 28% using a Wenger TX-52 twin-screw extruder. The properties of extrudates were studied in experiments conducted using a full-factorial design with three levels of DDGS content, two levels of moisture content, and two levels of screw speed. Increasing the DDGS content from 20 to 60% resulted in a 36.7% decrease in the radial expansion, leading to a 159 and 61.4% increase in the unit density and bulk density of the extrudates, respectively. Increasing the DDGS content resulted in a significant increase in the water absorption index (WAI) but a significant decrease in the water solubility index (WSI) of the extrudates. Changing the screw speed and moisture content had no significant effect on the radial expansion ratio but resulted in a significant difference in the bulk density of the extrudates, which may be due to the occurrence of longitudinal expansion. Even though changing the moisture content and screw speed had no significant effect on the WSI of the extrudates, significant differences in the WAI of the extrudates were observed. The ingredient components in the blend and moisture content had an influence on the color changes of the extrudates, as did the biochemical changes occurring inside the barrel during processing. Overall, it was determined that DDGS could be included at a rate of up to 60% using twin-screw extrusion, and that viable pelleted floating feeds can be produced.

Distillers dried grains with solubles (DDGS), a coproduct of ethanol production, contains high amounts of protein and fiber, and a low amount of starch. It is mostly used as animal feed. The protein conversion efficiency of fish is very high compared with other animals. Hence DDGS has great potential to be used as an alternative protein source for aquaculture feed production. Research by Wu et al (1994, 1996) indicated that tilapia fish can be grown with DDGS, which can improve the economic viability of aquaculture farms. Because starch is converted to ethanol during the fermentation process, micronutrients are available in concentrated quantities compared with whole corn. This potentially makes DDGS a better base material for aquaculture feed than whole corn. But the main problems with the production of extruded aquaculture feed with DDGS are its low starch content and high fiber content (Chin et al 1989). As the starch content is decreased, expansion obtained during extrusion is reduced, which subsequently affects the physical characteristics of the extrudates. As the fiber content is increased, extrudate mechanical strength and durability decrease due to its noninteracting nature with other components in the ingredient mix. Whey, a by-product of cheesemaking, has excellent binding properties. Whey has actually been used as a binder for production of aquaculture feed by the pelleting process (Lovell 1988). But inclusion of whey during extrusion processing of cereal foods has been found to decrease expansion, increase unit density, water absorption index, and breaking strength (Martinez-Serna and Villota 1992; Matthey and Hanna 1997). The exact mechanism of interaction of whey with other components is not well understood (Cumming et al 1973). We have found that whey can be successfully used as a binder with DDGS-based aquaculture feed blends (Chevanan et al 2005, 2006).

In the aquaculture industry, depending on the fish species, floating feeds are often preferred to sinking feeds, to prevent over feeding, and to maintain the cleanliness in the pond (Lovell 1988; Chang and Wang 1999). Twin-screw extruders have various advantages over single-screw extruders for production of floating feeds. Twin-screw extruders can handle viscous, oily, sticky, or wet ingredients with varying levels of protein, starch, fat, and fiber over a wide range of particle sizes and can achieve wide variations in the extrudate properties (Riaz 2000). The main difference between single-screw and twin-screw extruders is the transport mechanism (Mercier et al 1989). Single-screw extruders transport material by friction between the screw and the product, as well as the barrel and the product. With single-screw extruders, it is very difficult to achieve the required operating conditions for the production of floating feeds from ingredient mixes containing DDGS due to the low starch content. For example, extrudates (containing up to 40% DDGS and other ingredients including soy flour, corn flour, fish meal, mineral mix, vitamin mix, and whey, with a net protein content adjusted to 28%, db) obtained using a Brabender laboratory-scale single-screw extruder did not float (Chevanan et al 2006). In twin-screw extrusion, however, material within the barrel is transported positively, irrespective of the friction between the screw, barrel surface, and material (Zuilichem and Stolp 1984). Proper operating conditions can be easily achieved by varying parameters such as screw speed, feeding rate, ingredient moisture content, and temperature. Because of differences between the single-screw and twin-screw processes, it may be possible to use twin-screw extruders for ingredient mixes containing low starch materials such as DDGS, to produce floating feeds, although no studies have yet been reported that have accomplished this.

Aquaculture feeds typically require 26–50% protein, depending on the species to be fed (Lovell 1988), so the formulated ingredient mix will often contain a high amount of both starch and protein. During extrusion processing of the starch-based products, a relatively elastic melt is formed inside the barrel which results in a more expanded and crispy product (Ibanoglu et al 1996; Ilo et al 1996; Alves et al 1999; Thomas et al 1999). During extrusion processing of protein-based products, on the other hand, a plastic melt is formed inside the barrel and a more porous and textured product is formed (Gwiazda et al 1987; Harris et al 1988; Singh et al 1991; Sandra and Jose 1993). Extrudate floatability can be achieved through expansion during extrusion processing. Very little information is available on the effect of ingredient mixes containing high proportions of both starch and protein on expansion and other physical properties of extrudates by twin-screw extrusion. Thus the objective of this study was to examine the effect of varying levels of screw speed, blend moisture content, and DDGS content

---

[1] Graduate research assistant, South Dakota State University, Brookings, SD 57007.

[2] Bioprocess engineer, North Central Agricultural Research Laboratory, USDA – ARS Brookings, SD 57006.

[3] Corresponding author. Phone: (605) 693-3241. Fax: (605) 693-5240. E-mail address: krosentr@ngirl.ars.usda.gov

[4] Professor, South Dakota State University, Brookings, SD 57007.

doi:10.1094/CCHEM-84-5-0428

(in an ingredient blend containing soy flour, corn flour, Menhaden fish meal, mineral mix, vitamin mix, and whey) on physical properties of extrudates (such as extrudate expansion ratio, moisture content, durability, particle density, unit density, bulk density, water absorption index, water solubility index, and color) produced by twin-screw extrusion.

METHODS AND MATERIALS

Feed Blend Formulation

Three isocaloric ingredient blends were formulated to ≈28% protein with an energy content of ≈350 kcal/100 g. The ingredients in feed blends included DDGS, soy flour, corn flour, Menhaden fish meal, whey, vitamin mix, and mineral mix (Table I). The DDGS was provided by Dakota Ethanol LLC (Wentworth, SD) and was ground to a particle size of ≈100 μm using a laboratory grinder (S500 Disc Mill, Genmills, Clifton, NJ). 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 (Vitapak, Land O'Lakes Feed, St. Paul, MN), fish meal (Consumer Supply Distribution Company, Sioux City, IA) and whey (Bongards Creameries, Perham, MN) were incorporated into the ingredients blends at levels of 1, 2, 5, and 5%, respectively, on a wet (as-is) basis. The ingredients were mixed in a laboratory-scale mixer (N50 Mixer, Hobart Corporation, Troy, OH) for 30 min, and then stored at room temperature for two days until the experiments commenced.

Experimental Design and Extrusion Processing

Experimental extrusions were conducted using a co-rotating, fully intermeshing, self-wiping, twin-screw extruder (Wenger TX-52, Sabetha, KS), with a feed hopper and preconditioner. The ingredient blends were metered from the feed hopper into the preconditioner, which was fitted with steam injection and water injection ports to condition the ingredients to specific temperature and moisture contents. The conditioned ingredients were dropped into the extruder at a feeder speed of 14 rpm and an average feed rate of 49 kg/hr. The extruder had 52-mm diameter twin screws, and the barrel had a length-to-diameter ratio of 25.5:1. The screw had 25 individual sections and the configuration (down the length of the barrel from the feeding section to the die section) used in our experiments consisted of four conveying screws, three shear locks, one conveying screw, one conveying screw backward, three conveying screws, one conveying screw backwards, four conveying screws, one shear lock, one interrupted flight conveying screw, one conveying screw, one interrupted flight conveying screw, one shear lock, and at the end, one final (cone) screw. The two die plates each had circular openings of 3.175 mm diameter. The cutter was positioned off center at the end of the die, had three blades, and the rotational speed was adjusted to 750 rpm to produce pelleted extrudates.

Experiments were conducted using a full-factorial design, with three levels of DDGS (20, 40, and 60%), two levels of moisture content (15 and 19%, wb) achieved through the preconditioner before extrusion, and two levels of screw speed (350 rpm, 36.7 rad/sec and 420 rpm, 44.0 rad/sec) and was implemented using a completely randomized design.

The barrel had six temperature zones from the feeding section to the die section, which were maintained at 60, 80, 80, 80, 100, and 110°C throughout the experiments.

Measurement of Physical Properties

Expansion ratio. Radial expansion ratio was measured as the ratio of diameter of the extrudates to the diameter of the die (Conway and Anderson 1973). The diameter of the six extrudates for each treatment was measured using a digital caliper (Digimatic Series No. 293, Mitutoyo Co., Tokyo, Japan) and the average of six readings was used for analysis.

Moisture content of the extrudates, as well as that of the preconditioned ingredient blends, was determined following Method S352.2 (ASAE 2004) using a forced-convection laboratory oven (Thelco Precision, Jovan, Winchester, VA) at 103°C for 72 hr.

Extrudate durability was determined by Method S269.4 (ASAE 2004). Extrudates (200 g) were tumbled inside a pellet durabilty tester for 10 min and sieved through a No. 6 (3.4 mm) screen. Durability was then calculated as

$$\text{Durability}\,(\%) = \frac{\text{Mass of pellets after tumbling}}{\text{Mass of pellets before tumbling}} \times 100 \quad (1)$$

Particle density of the extrudates was determined with a multivolume pycnometer (model No. 1305, Micromeritics, Atlanta, GA) using the volume displacement method with helium gas (Chang 1988). Extrudates were placed into 150-cm^3 cups and the true volume was determined. The particle density was calculated as the ratio of the mass of the extrudates to the true volume (Rahman 1995). Calibration of the equipment was performed with a metal ball provided by the manufacturer.

Unit density. The extrudate samples were cut to 2-cm lengths, weighed on an electronic balance (model A-250, Denver Instrument, Arvada, CO), and measured for diameter using digital caliper (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) (Jamin and Flores 1998).

Bulk density was measured as the ratio of the mass of extrudates to the bulk volume that the extrudates occupied in a given volume, and was determined using a standard bushel tester (Seedburo Equipment Company, Chicago, IL) following the method prescribed by USDA (1999).

Water absorption index (WAI) was determined by the method of Jones et al (2000). WAI is defined as the mass of gel (g) obtained per mass (g) of solids. To determine the water absorption index, 2.5 g of finely ground sample was suspended in 30 mL of distilled water at 30°C in a 50-mL tarred centrifuge tube. The content was stirred intermittently over a period of 30 min and then centrifuged at 3,000 × *g* for 10 min. The supernatant water was transferred carefully into tarred aluminum cups. The mass of the remaining gel was weighed and WAI was then calculated as the ratio of the mass of gel to the mass of the sample.

Water solubility index (WSI) was determined as the water-soluble fraction in the supernatant, expressed as a percent of dry sample (Jones et al 2000). The WSI was determined from the amount of dried solids recovered by evaporating the supernatant in an oven at 135°C for 2 hr.

Color of the whole extrudates was determined using a calibrated spectrophotometer (portable model CM 2500d, Minolta Corporation, Ramsey, NJ) using the Hunterlab color space where L^* quantified brightness/darkness of the extrudates, a^* quantified redness/greenness of the extrudates, and b^* quantified yellowness/blueness of the samples.

TABLE I
Ingredient Components in Experimental Feed Blends

	Weight of Ingredients (g/100 g)		
Feed Ingredients	**Blend I**	**Blend II**	**Blend III**
DDGS	20.0	40.0	60.0
Soy flour	32.9	24.8	16.5
Corn flour	34.1	22.2	10.5
Fish meal	5.0	5.0	5.0
Whey	5.0	5.0	5.0
Mineral mix	1.0	1.0	1.0
Vitamin mix	2.0	2.0	2.0

Nutrient Analysis

Extrudates were air-dried and protein, fiber, fat, and ash content were determined following official Methods 990.03, 978.10, 920.39, and 920.48, respectively (AOAC 2003). Nitrogen-free extract (NFE) was determined after deducting the moisture, protein, fat, fiber, and ash contents. The protein, fat, ash, and NFE were measured in duplicate ($n = 2$) for all raw ingredient blends.

Statistical Analyses

Three replicates were measured for all physical properties studied except expansion ratio, where six measurements were taken. The data were then analyzed with the Proc GLM procedure (v.8, SAS Institute, Cary, NC) to identify the significant main and interaction effects at an α level (Type I error rate) of 0.05. Mean values associated with significant effects were compared using least significant differences (LSD).

RESULTS AND DISCUSSION

Changing the level of DDGS had significant effects on all the parameters studied (Table II). Changing the levels of blend moisture content also had significant effects on the extrudate properties studied, except for expansion ratio, unit density, particle density, and extrudate brightness (L^*). Changing the screw speed had a significant effect only on durability, particle density, bulk density, and WSI. None of the interaction effects were significant for expansion ratio, unit density, or yellowness of extrudates. All the interaction effects were significant for extudate moisture content, durability, and bulk density. The interaction effect of blend moisture content with screw speed was not significant (Table III).

Expansion Ratio

The extent of radial expansion attained during extrusion processing affects the floatability of extrudates (Oliveira et al 1992). In our experiments, the extrudates obtained from the 12 treatment combinations floated for more than 16 hr and maintained cohesive structure during this time. Changing the levels of DDGS from 20 to 60% had a significant effect on the expansion ratio of the extrudates and resulted in a 36.7% reduction in their expansion (Table II). But changing the moisture content of the ingredient mix from 15 to 19%, and the screw speed from 350 rpm to 420 rpm, had no significant effects on the expansion ratio of the extrudates. All extrudates floated, even though the average radial expansion ratio achieved in the extrudates containing 60% DDGS was only 1.05. This indicated that for extrudates containing DDGS, perhaps the longitudinal expansion was greater compared with the radial expansion. Generally shrinkage occurs during the drying process and the higher the moisture content of the biological materials before drying, the more shrinkage typically results. Postextrusion shrinkage of 20–50% was observed by Park (1976). The moisture content of extrudates immediately after extrusion processing was highest for the extrudates containing 60% DDGS. The expansion ratio was measured only for the dried extrudates. Hence, the expansion ratio of the wet extrudates containing 60% DDGS immediately after extrusion might have been higher than 1.05, thus resulting in a more porous structure. In twin-screw extrusion, screw speed is generally believed to have little effect on the extrudate expansion (Martin-Cabrejas et al 1999; Ding et al 2005) and in our experiment also we observed that screw speed had no significant effect on the expansion of the extrudates.

Moisture Content

We observed that changing DDGS content and moisture content of the ingredient blends had a significant effect on the moisture content of the resulting extrudates immediately after extrusion. The moisture content of extrudates is a very important parameter that influences extrudate properties such as durability, WAI, and WSI (Rolfe et al 2001). As expected, changing the screw speed had no significant effect on the moisture content of extrudates. Increasing the DDGS content from 20 to 60%, however, resulted in nearly a 100% increase in the moisture content of the extrudates (Table II). In twin-screw extruders, the required processing temperature can be achieved by three means: 1) application of heat through stream during preconditioning and extrusion; 2) heat

TABLE II
Main Effects Due to DDGS, Screw Speed (SS), and Moisture Content (MC) on Physical Properties of Extrudates[a]

	Expansion Ratio (-)	MC of Extrudates (% wb)	Durability (%)	Particle Density (g/cm³)	Unit Density (g/cm³)
DDGS (%)					
20	1.66a (0.02)	12.54a (1.28)	97.76a (0.45)	1.397a (0.003)	0.235c (0.006)
40	1.36b (0.01)	19.11b (0.90)	97.60a (0.19)	1.383b (0.001)	0.342b (0.003)
60	1.05c (0.01)	25.48a (0.65)	96.85b (0.41)	1.366c (0.001)	0.609a (0.007)
SS (rpm)					
350	1.37 (0.01)	21.90 (1.51)	97.31 (0.40)	1.379b (0.003)	0.407 (0.001)
420	1.35 0.08	22.85 (0.85)	97.48 (0.36)	1.385a (0.003)	0.384 (0.001)
MC (%)					
15	1.35 (0.08)	20.79b (0.83)	98.20a (0.34)	1.382 (0.006)	0.389 (0.009)
19	1.37 (0.07)	23.97a (1.37)	96.61b (0.26)	1.382 (0.003)	0.401 (0.011)

[a] Means with different letters in a column, within each main effect, were significantly different ($P < 0.05$). Values in parentheses are standard error.

TABLE II *(continued)*
Main Effects Due to DDGS, Screw Speed (SS), and Moisture Content (MC) on Physical Properties of Extrudates[a]

	Bulk Density (g/cm³)	WSI (%)	WAI (-)	L^*	a^*	b^*
DDGS (%)						
20	0.236c (0.006)	15.51c (0.51)	3.73a (0.06)	48.26a (0.58)	10.02b (0.17)	37.61c (0.45)
40	0.300b (0.003)	16.32b (0.27)	3.34b (0.03)	38.28b (1.13)	10.48b (0.22)	46.48b (0.96)
60	0.381a (0.003)	16.96a (0.28)	2.77c (0.28)	32.91c (0.59)	12.50a (0.33)	52.51a (0.59)
SS (rpm)						
350	0.309a (0.001)	16.20 (0.30)	3.34a (0.14)	39.40 (2.17)	11.12 (0.43)	45.85 (1.90)
420	0.302b (0.001)	16.32 (0.49)	3.22b (0.11)	40.24 (1.90)	10.89 (0.37)	45.21 (1.97)
MC (%)						
15	0.301b (0.010)	16.60 (0.20)	3.35a (0.12)	40.03 (2.16)	11.27a (0.48)	44.60b (1.82)
19	0.310a (0.009)	15.92 (0.52)	3.21b (0.13)	39.61 (1.93)	10.73b (0.48)	46.47a (2.02)

[a] Means with different letters in a column, within each main effect, were significantly different ($P < 0.05$). Values in parentheses are standard error.

developed by shearing action (friction); and 3) application of heat through electrical heaters in various sections of the barrel. Water is held in biological materials by molecular and capillary absorption corresponding to the bound water and free water, respectively (Mohsenin 1986). In extrusion processing, the micropore structure in the extrudates results from the expansion occurring due to flashing of water because of the sudden drop in pressure at the die exit, and as such, expansion, texture, and final moisture content will depend on the ability of the matrix to stretch and to retain the evaporating water. At higher DDGS content, the applied energy and expansion that occurred may have resulted in the molecular and micropore structural changes in such a way that it retained more water and resulted in a higher moisture content of the extrudates. The ingredient moisture content at 15%, wb, resulted in a moisture content of 20.79%, wb, in the extrudates, while an ingredient moisture content of 19%, wb, resulted in an extrudate moisture content of 23.97%, wb (Table II). The moisture content of the dry ingredient mix containing 20, 30, and 40% DDGS before preconditioning were observed to be 7.9, 8.2, and 8.4%, respectively. The higher moisture content observed in the extrudates compared with the raw material was due to moisture added through steam in the preconditioner and extruder barrel. High temperatures, shear stresses, and shear strains produced during extrusion processing affect the complex interactions between water and the other chemical constituents, and alter the resulting internal cellular structures that occur during water evaporation upon die exit (Miller 1985). These alterations are ultimately reflected in the expansion of the material as it passes through the extruder die and can be quantified through the expansion ratio (Moore et al 1990). Expansion and resulting extrudate structure are highly dependent on the composition of the extruded material, especially starch content, which is key to product expansion through starch gelatinization (Nielsen 1976). Products high in starch generally exhibit high expansion; those low in starch (e.g., high in protein or fiber) generally exhibit limited expansion.

Durability

The mechanical strength of extrudates is often described by durability, which is a very important quality of feed materials (Rosentrater et al 2005). Strength of the extrudates depends on several factors including the extent of heat treatment and the relative degree of starch transformation inside the barrel. If the raw ingredients contain highly altered materials that do not easily interact, the extrudates become noncohesive, resulting in poor durability (Colonna et al 1989). We observed that changing the levels of DDGS and preconditioned blend moisture content did have significant effects on durability ($P < 0.05$), whereas changing the screw speed had no significant effect on extrudate durability. Increasing the DDGS content from 20 to 40% did not result in significant changes in the durability. But increasing further from 40 to 60% resulted in a significant decrease in the durability. Even though the durability of the extrudates obtained with twin-screw extrusion was very high (95.41–99.50%), the reduced durability with 60% DDGS content was probably due to the low starch and high fiber contents in the ingredient blend. Significant increases in the mechanical strength of extrudates have been observed by many researchers when the moisture content of the ingredient mix entering the extruder was increased for various ingredients (Bandyopadhyay and Rout 2001; Rolfe et al 2001; Shukla et al 2005). In our experiments also, changing the moisture content of the ingredients had a significant effect on the durability, and increasing the moisture content from 15 to 19% resulted in a 1.65% increase in extrudate durability.

Particle Density

Particle density depends to a large extent on physical attributes of the raw materials such as particle size, density, mechanical properties, rheology of different components (single or in combination), in addition to the process of extrusion itself (Smith 1992). In our experiments, even though the particle density values of the extrudates were >1.0, the extrudates floated with the shape intact for >16 hr. In extrusion processing, closed and open pores are formed in the extrudates during the expansion process. The retention of the entrapped air in the extrudates depends on the water absorption characteristics of the extruded products. When extrudates absorb water, the structure collapses, and entrapped air is released. The gelatinization of starch and denaturation of protein occurring during processing might have resulted in retaining extrudate structure, and thus entrapped the air for a longer period and resulted in floatability of the extruded products up to 16 hr in our experiment. Changing the levels of DDGS had a significant effect on the particle density of the extrudates ($P < 0.05$) and increasing the DDGS content from 20 to 60% in the ingredient mix resulted in a 2.2% decrease in the particle density (Table II). To attain a net protein content of 28%, various raw material combinations were used, and thus the chemical constituents varied, which ultimately influenced the changes in the particle density of the extrudates. Increasing the screw speed from 350 rpm (36.7 rad/sec) to 420 rpm (44.0 rad/sec) resulted in a significant increase in the particle density of

TABLE III
Interaction Results for DDGS, Moisture Content (MC), and Screw Speed (SS) on Physical Properties of Extrudates (*P* values)

	Expansion Ratio (-)	MC of Extrudates (% wb)	Durability (%)	Particle Density (g/cm^3)	Unit Density (g/cm^3)
DDGS	<0.001	<0.001	<0.001	<0.001	<0.001
MC	0.370	<0.001	<0.001	0.742	0.578
SS	0.339	<0.332	0.295	<0.001	0.242
DDGS*MC	0.725	0.005	<0.001	<0.001	0.511
DDGS*SS	0.956	<0.001	<0.001	0.495	0.163
MC*SS	0.952	<0.001	0.004	0.002	0.989
DDGS*MC*SS	0.222	<0.001	0.003	0.007	0.965

TABLE III (*continued*)
Interaction Results for DDGS, Moisture Content (MC), and Screw Speed (SS) on Physical Properties of Extrudates (*P* values)

	Bulk Density (g/cm^3)	WSI (-)	WAI (%)	L^*	a^*	b^*
DDGS	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
MC	<0.001	0.003	<0.001	0.512	0.041	0.016
SS	<0.001	0.536	<0.001	0.196	0.012	0.184
DDGS*MC	<0.001	<0.001	0.050	<0.001	0.363	0.381
DDGS*SS	<0.001	0.002	0.007	0.194	0.565	0.265
MC*SS	<0.001	0.355	0.778	<0.001	0.823	0.721
DDGS*MC*SS	<0.001	<0.001	<0.001	0.082	0.340	0.324

extrudates; this might be due to structural changes associated with denaturation of protein and gelatinization of limited starch occurring in the melt inside the barrel. Changing the moisture content did not significantly affect the particle density. On the other hand, significant interactions between moisture content and DDGS content, and moisture content and screw speed (Table III) were observed. These interactions may have resulted in changes in the particle density of extrudates and may have been due, in part, to simultaneously changing the moisture content of the ingredient mix, which altered the chemical constituents, and thus matrix behavior when subjected to varying rates of shear.

Unit Density

Unit density of extrudates is directly related to the degree of expansion obtained during extrusion processing (Colonna et al 1989). The highest expansion ratio of 1.66 and the lowest unit density of 0.236 g/cm^3 was observed for extrudates containing 20% DDGS (Table II). The lowest expansion ratio of 1.05 was observed for extrudates containing 60% DDGS, which produced the highest unit density of 0.609 g/cm^3. Increasing DDGS content from 20 to 60% resulted in a 159% increase in the unit density of extrudates. Changing screw speed and moisture content, however, did not result in significant changes in the expansion ratio of the extrudates, and conversely resulted in no significant changes in the unit density of these extrudates.

Bulk Density

Bulk density of the extrudates is affected by the volume of pores formed inside the extrudates as expansion occurs during extrusion processing, as well as void spaces formed during filling of the irregular-shaped extrudates into containers of a specific size during testing. Bulk density is very important because it determines the space required to store the extruded feed materials, both at production sites and on farms. All three independent factors (level of DDGS, screw speed, and moisture content) significantly affected the bulk density of the extrudates ($P < 0.05$) (Table III). Increasing the DDGS content from 20 to 60% resulted in a 61.4% increase in bulk density (Table II). This was expected because as the DDGS content increased, the expansion ratio decreased and the extrudates became lighter. Increasing the screw speed from 350 rpm (36.7 rad/sec) to 420 rpm (44.0 rad/sec) resulted in a 2.3% decrease in bulk density, while increasing the moisture content of the ingredient mix from 15 to 19% resulted in a 3% increase in bulk density. Even though no significant difference in the radial expansion of the extrudates was observed by changing either the moisture content of the ingredient mix or the screw speed, significant differences in the bulk density were observed. There may have been differences in the longitudinal expansion occurring, which in turn may have influenced the bulk density of the extrudates.

WAI and WSI

WAI, a measure of volume occupied by the starch after swelling in excess water, is a portion of the starch that maintained integrity during extrusion processing (Mason and Hoseney 1986). WSI, on the other hand, is a measure of the degree of starch conversion during extrusion processing and is an indicator of the degradation of molecular components (Kirby et al 1988). In general, an increase in WSI corresponds to decrease in the WAI of extrudates and has been observed by many authors (Kirby et al 1988; Ng et al 1999). In our experiments, increasing the DDGS level produced a higher WSI and a decreasing trend in WAI of the extrudates.

Changing the levels of DDGS, screw speed, and moisture content had a significant effect on the WAI of the extrudates as well (Table II). Increasing the DDGS content from 20 to 60% resulted in a 25.7% decrease in the WAI of the extrudates. Increasing the screw speed from 350 rpm (36.7 rad/sec) to 420 rpm (44.0 rad/sec) resulted in a 3.6% decrease in the WAI of the extrudates, while increasing the moisture content of the ingredient mix from 15 to 19% resulted in a 4.2% decrease in WAI.

The levels of DDGS content resulted in significant changes in the WSI of the extrudates. But changing the screw speed and moisture content of the ingredient mix did not result in significant changes in the WSI of the extrudates (Table II). The WSI depends on various factors. For example, starch sources from cereal and potato result in different levels of WSI, depending on the amount of amylase and amylopectin present in the starch. The extent of dextrination occurring in the extrudates due to high shear and high temperature also influences the WSI (Mercier and Feillet 1975; Gomez and Aguilera 1984; Govindasamy et al 1995). Interaction between the starch and protein influences the WSI of the extrudates as well (Paton and Spratt 1984). In our experiments, increasing the DDGS content from 20 to 60% resulted in a 9.3% increase in the WSI. However, there were no significant changes in WSI due to changing the moisture content of the ingredient mix and screw speed (Table II).

Color

In extrusion processing, Maillard reactions can be a cause of color changes in extrudates. Color, per se, probably is not an important factor in aquaculture feed by itself, but changes in color due to high temperatures and other reactions during processing can be an indication of alteration or loss of lysine, which is an important amino acid needed in aquaculture diets. For example, lysine losses ≤40% have been observed during extrusion processing, due primarily to high temperatures (Bjorck and Asp 1983). In this study, as DDGS content increased from 20 to 60%, a significant decrease of 31.8% in L^* was observed (Table II). DDGS is typically dark compared with soy flour and corn flour. Thus, as the DDGS content was increased, the feed brightness decreased as well. Increasing the moisture content of the feed ingredient mix from 15 to 19% and screw speed from 350 to 420 rpm did not significantly change L^* value. Increasing the DDGS content from 20 to 60% increased redness of the extrudates by 24.8%, which might be due, in part, to the difference in the color of the raw ingredients used before extrusion. Increasing the moisture content from 15 to 19% decreased the redness of the extrudates by 5.3%, but no significant changes in the a^* value were observed by changing the screw speed from 350 to 420 rpm. Increasing the DDGS content from 20 to 60% resulted in a 39.6% increase in yellowness of the extrudates. This was expected because DDGS was yellowish in color. Increasing the DDGS content thus resulted in a significant increase in the yellowness of the resulting extrudates. Changing the level of screw speed did not result in significant changes in the yellowness of the extrudates but changing the moisture content from 15 to 19% did result in a 4.2% increase in the yellowness of the extrudates. This indicated that the color differences in the extrudates were highly influenced by changes in the moisture content of the ingredient blend before processing, as well as biochemical changes inside the extruder. Although not determined, the overall differences could also be influenced by differences in the color of the raw ingredient blends themselves.

Treatment Combination Effects

The effect of DDGS on different properties of extrudates can be well understood through examination of the treatment combination effects (Figs. 1–4). As the level of DDGS content was changed, significant changes in the expansion ratio were observed. However, there was no significant effect on expansion ratio due to screw speed and moisture content of the ingredient blend as DDGS varied. In general, as DDGS content increased, the moisture content of the extrudates increased. The moisture content of the extrudates decreased significantly as the DDGS content increased. At specific DDGS levels, both screw speed and moisture content of the ingredient blend had significant effects on various physical properties. Moisture content and screw speed had significant effects on extrudate durability, in addition to the levels of DDGS present in the ingredient mix (Fig. 1). In general, at lower DDGS levels, bulk density and unit density were lower, but particle density was higher.

The higher particle density at lower DDGS levels might be due to the particle density difference between the components in the ingredient mixes themselves. In general, WAI was higher at lower DDGS levels, but there was no significant effect on WAI due to moisture content of the ingredient blend or screw speed at 20 and 40% DDGS levels.

However, at the 60% DDGS level, significant effects due to moisture content of the ingredient blend were observed (Fig. 3). The color (brightness, redness, and yellowness) of the extrudates was mostly due to color of the ingredient components in the blend, and there was no effect due to either moisture content on screw speed at 20 and 40% levels. But at 60% moisture content, changing the moisture content of the ingredient blend had a significant effect on the color of the extrudates (Fig. 4).

Correlations

Correlation analysis of the multivariate data provides valuable information about the relationships between the different properties in extrusion studies. A higher correlation between some un-

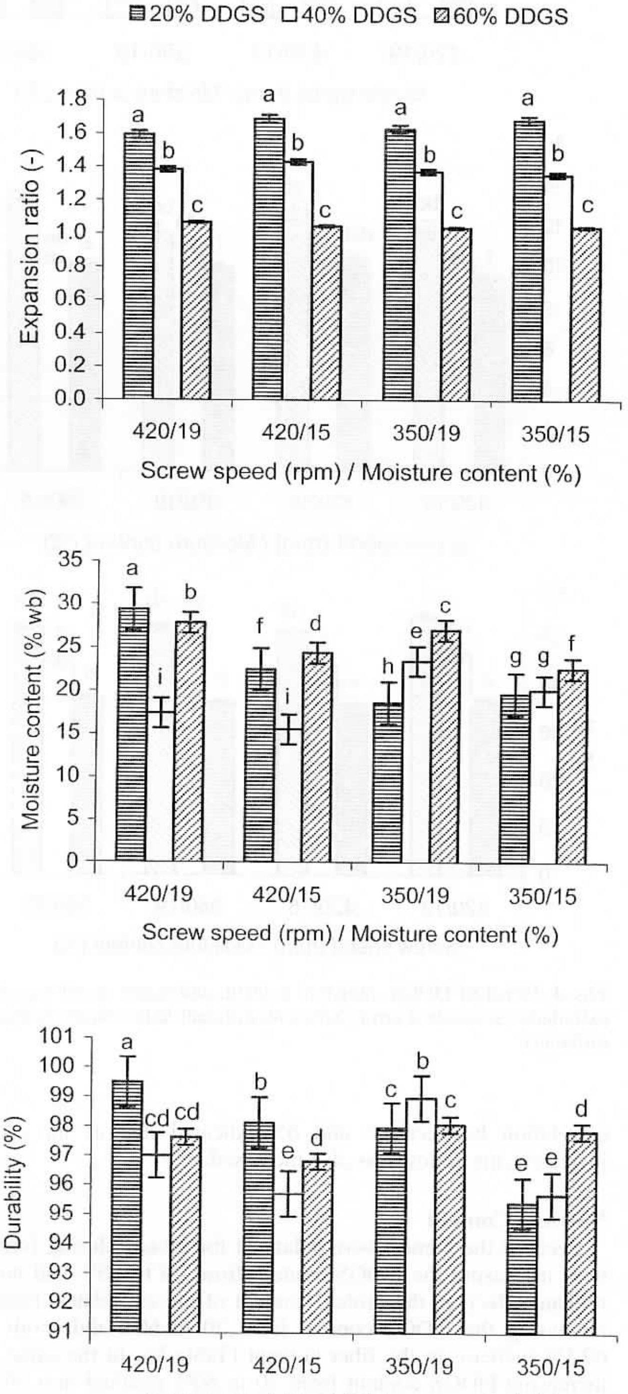

Fig. 1. Effect of DDGS, moisture content, and screw speed on expansion ratio, moisture content, and durability of extrudates (± standard error; bars with different letters were significantly different).

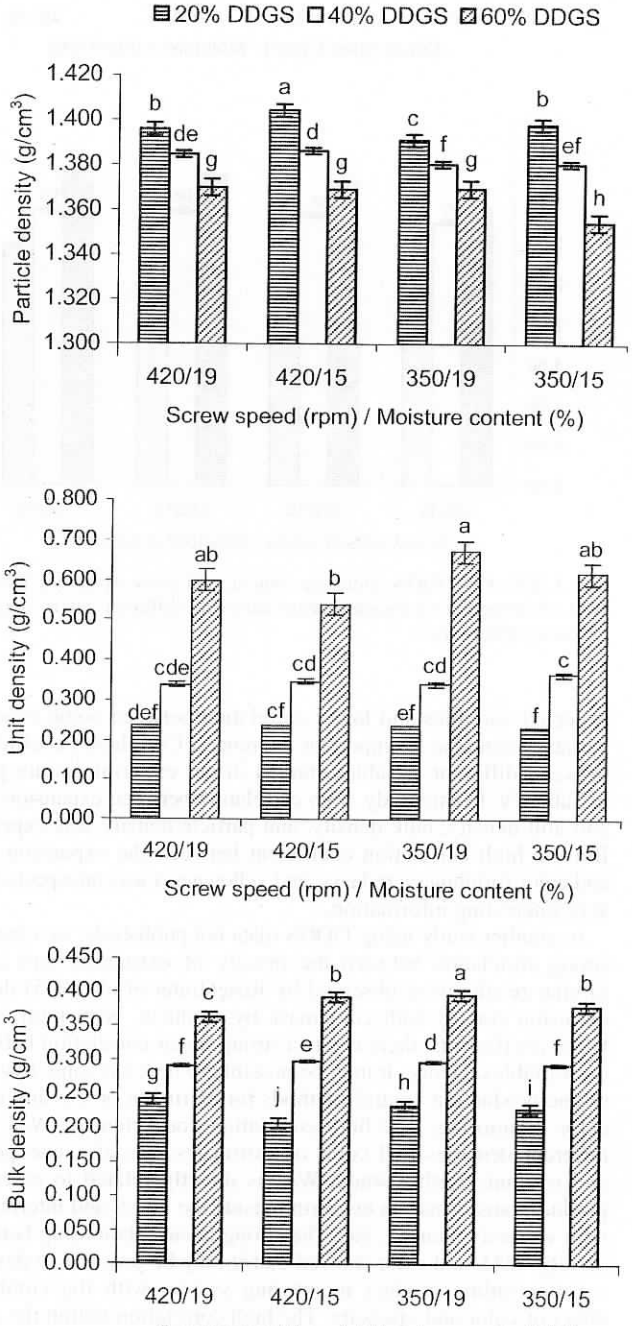

Fig. 2. Effect of DDGS, moisture content, and screw speed on particle density, unit density, and bulk density of extrudates (± standard error; bars with different letters were significantly different).

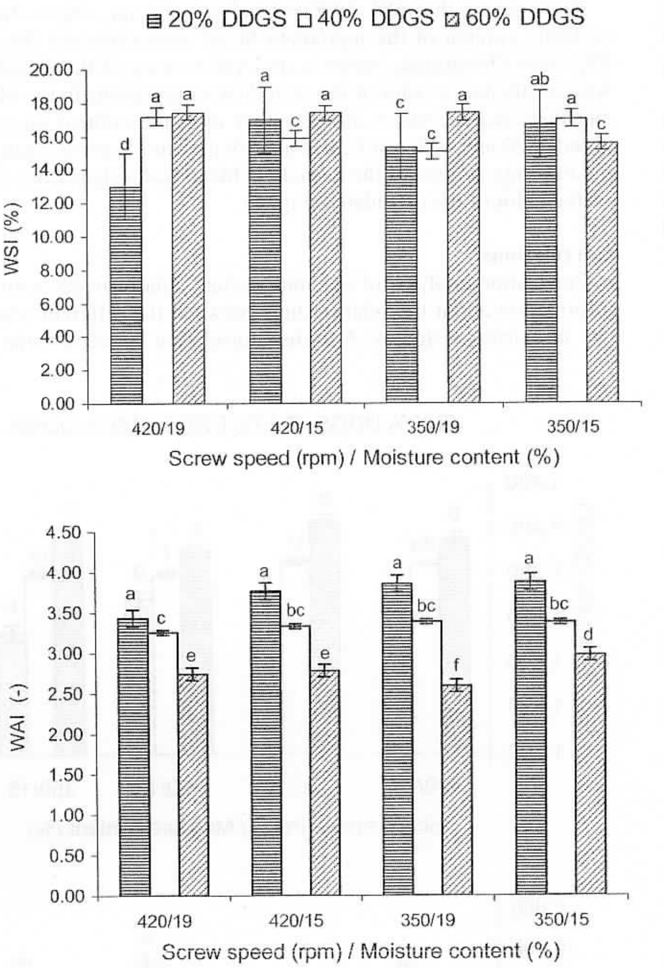

Fig. 3. Effect of DDGS, moisture content, and screw speed on WSI and WAI of extrudates (± standard error; bars with different letters were significantly different).

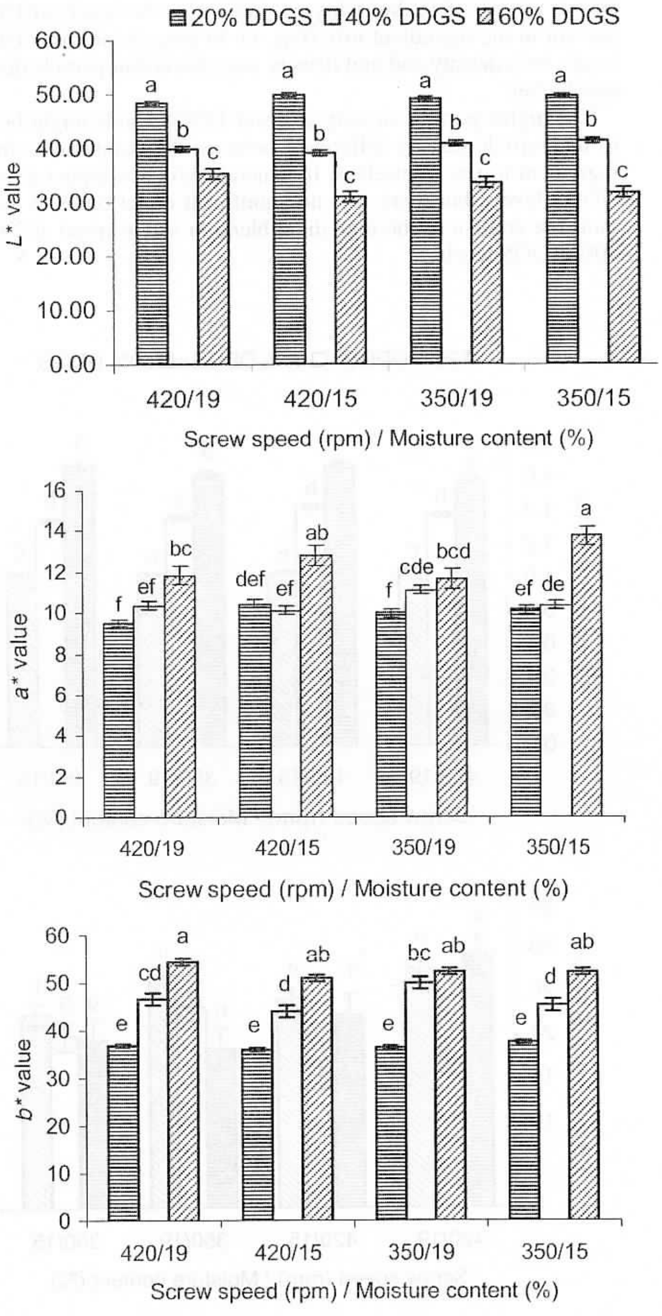

Fig. 4. Effect of DDGS, moisture content, and screw speed on color of extrudates (± standard error; bars with different letters were significantly different).

expected variables and lower correlation between some expected variables can lead to important inferences. Correlation coefficients between different variables studied in our experiments are given in Table IV. In this study, high correlation between expansion ratio and unit density, bulk density, and particle density was expected. But the high correlation coefficient between the expansion ratio and color (brightness, redness, and yellowness) was unexpected and very interesting information.

In another study using DDGS (data not published), we observed strong correlation between the density of extrudates and color. Similar results were observed by Rosentrater et al (2005) during extrusion studies with corn masa by-products. With high correlation coefficients, there exists a strong linear correlation between the variables. Hence, it may be possible to develop some low-cost online production control methods for extrusion processing using color monitoring. The high correlation coefficient of WAI with different densities and color of extrudates was also unexpected and warrants further study. WAI is directly related to extent of product transformation occurring inside the barrel and interrelated with viscosity, density, etc. The strong linear relationship between density, WAI, and color showed that it may be possible to develop a better online product monitoring system with the combined effect of color and viscosity. The high correlation within the color components (brightness, redness, and yellowness) of extrudates was expected. Negative correlation of L^* with a^* and b^* indicated that, as the brightness of the extrudates decreased, the redness and yellowness of the extrudates increased. However, the positive correlation between a^* and b^* indicated that as the redness increased, the yellowness also increased.

Nutrient Content

Because the blends were balanced for protein during formulation, increasing the DDGS content from 20 to 60% had no significant effect on the protein content of the extrudates. However increasing the DDGS content from 20 to 60% did result in a 69.3% increase in the fiber content (Table V). In the same way, increasing DDGS content from 20 to 60% resulted in a 493.0% increase in the fat content of the extrudates. The fiber content and fat content of the DDGS was higher than the corn flour and soy flour and, hence, increasing the DDGS content from 20 to 60% resulted in significant increases in fiber content and fat content of

TABLE IV
Correlation Coefficients for All Multivariate Extrudate Data[a]

	ER	ED	UD	BD	TD	WAI	WSI	MCE	L*	a*	b*
ER	1										
PD	0.256	1									
UD	**–0.899**	–0.192	1								
BD	**–0.967**	–0.238	**0.922**	1							
TD	**0.921**	0.307	**–0.877**	**–0.940**	1						
WAI	**0.922**	0.070	**–0.862**	**–0.941**	**0.819**	1					
WSI	–0.446	–0.334	0.365	0.317	–0.239	–0.343	1				
MCE	–0.293	0.341	0.418	0.361	–0.248	–0.489	–0.177	1			
L*	**0.886**	0.236	**–0.817**	**–0.898**	**0.857**	**0.788**	–0.305	–0.208	1		
a*	**–0.717**	–0.400	**0.735**	**0.743**	**–0.767**	–0.612	0.257	0.294	**–0.725**	1	
b*	**–0.921**	–0.187	**0.811**	**0.910**	**–0.861**	**–0.855**	0.365	0.309	**–0.908**	**0.739**	1

[a] Values in bold letters had significant ($P < 0.05$) correlation coefficients. ER, expansion ratio; PD, extrudate durability; UD, unit density; BD, bulk density; TD, particle density; WAI, water absorption index; WSI, water solubility index; MCE, moisture content of extrudates; L*, brightness; a*, redness; b*, yellowness.

TABLE V
Nutrient Content (% db) of Extrudates with Different Levels of Distillers Dried Grains with Solubles (DDGS) ($n = 2$)[a]

% DDGS	Protein	Fiber	Fat	Ash	NFE[b]
20	30.53 (0.15)	3.62c (0.38)	1.15c (0.09)	5.93c (0.06)	56.76a (0.39)
40	30.85 (0.14)	4.50b (0.27)	2.98b (0.17)	6.05b (0.45)	55.15b (0.26)
60	30.95 (0.22)	6.13a (0.38)	6.82a (0.15)	6.58a (0.07)	52.60c (0.51)

[a] Means with different letters in a column were significantly different ($P < 0.05$). Values in parentheses are standard error.
[b] Nitrogen-free extract (NFE).

the extrudates as well. Increasing the DDGS content from 20 to 60% resulted in a 7.3% decrease in the nitrogen-free extract (NFE) of the extrudates. During ethanol production, most of the starch was converted to ethanol. Due to this, the ingredient mix containing lower amounts of DDGS had increased quantities of starch. This might result in a higher amount of NFE for the extrudates with the ingredient mix containing 20% DDGS. Changing the levels of moisture content and screw speed had no significant effect on most of the nutrient components studied in our experiments.

CONCLUSIONS

The floatability of the extrudates is a key factor for aquaculture feeds. All extrudates produced with experiments conducted using a full-factorial design with three levels of DDGS, two levels of blend moisture content, and two levels of screw speed were observed to float. Moreover, all extrudates exhibited excellent floating capabilities. Changing the level of DDGS had significant effects on all the properties studied, such as extrudate expansion ratio, final moisture content, durability, particle density, unit density, bulk density, WSI, WAI, and color. Changing the moisture content of the ingredient mix had significant effects on extrudate moisture content, durability, bulk density, WSI, WAI, and yellowness. Changing the screw speed had a significant effect on particle density, bulk density, and WAI of the extrudates. Results from this industrial-scale twin-screw extrusion study indicate that floating aquaculture feeds can be successfully produced using ingredient mixes containing ≤60% DDGS for the process conditions studied. This level of DDGS inclusion has not been achieved in any prior studies, and thus follow-up feeding trials are warranted to determine whether DDGS at this level can effectively be used as a primary alternative protein source for aquaculture feeds.

ACKNOWLEDGMENTS

We thankfully acknowledge the financial support provided by the Agricultural Experiment Station, South Dakota State University, Brookings, SD, and the North Central Agricultural Research Laboratory, USDA-ARS, Brookings, SD, for carrying out this project.

LITERATURE CITED

Alves, R. M. L., Grossmann, M. V. E., and Silva, R. S. S. F. 1999. Gelling properties of extruded yam (*Dioscorea alota*) starch. Food Chem. 67:123-127.

AOAC. 2003. Official Methods of Analysis of AOAC International, 17th Ed. AOAC International: Gaithersburg, MA.

ASAE. 2004. American Society of Agricultural Engineers Standards, Engineering Practices and Data. The Society: St. Joseph, MI.

Banyopadhyay, S., and Rout, R. K. 2001. Aquafeed extrudate flow rate and pellet characteristics from low cost single screw extruder. J. Aquatic Food Product Technol. 10:3-14.

Bjorck, I., and Asp, N. G. 1983. The effects of extrusion cooking on nutritional value. A literature review. J. Food Eng. 2:281-308.

Chang, C. S. 1988. Measuring density and porosity of grain kernels using a gas pycnometer. J. Dairy Res. 65:13-15.

Chang, Y. K., and Wang, S. S. 1999. Advances in extrusion technology, aquaculture/animal feeds and foods. Technomic Publishing: Lancaster, PA.

Chevanan, N., Muthukumarappan, K., and Rosentrater, K. 2005. Effect of whey protein as a binder during the extrusion of fish feed pellets. ASABE Paper No. SD05-100. ASABE: St. Joseph, MI.

Chevanan, N., Muthukumarappan, K., and Rosentrater, K. 2007. Effect of die dimensions on extrusion processing parameters and properties of DDGS-based aquaculture feeds. Cereal Chem. 84:389-398.

Chin, H. K., Joseph, A. M., and Jeffery, T. M. 1989. Properties of extruded dried distillers grains (DDG) and flour blends. J. Food Process. Preserv. 13:219-231.

Colonna, P., Tayeb, J., and Mercier, C. 1989. Extrusion cooking of starch and starchy products. Pages 247-320 in: Extrusion Cooking. C. Mercier, P. Linko, and J. M. Harper, eds. AACC International: St. Paul, MN.

Conway, H. F., and Anderson, R. A. 1973. Protein fortified extruded food products. Cereal Sci. Today 18:94-97.

Cumming, D. B., Stanley, D. W., and Deman, J. M. 1973. Fate of water soluble soy protein during thermoplastic extrusion. J. Food Sci. 38:320.

Ding, Q. B., Ainsworth, P., Tucker, G., and Marson, H. 2005. The effect of extrusion conditions on the physiochemical properties and sensory characteristics of rice based expanded snacks. J. Food Eng. 66:283-289.

Gomez, M. H., and Aguilera, J. M. 1984. A physicochemical model for extrusion of corn starch. J. Food Sci. 49:40-43.

Govindasamy, S., Campanella, O. H., and Oates, C. G. 1995. High moisture twin screw extrusion of sago starch: 1. Influence on granule morphology and structure. Carbohydr. Polym. 30:275-286.

Gwiazda, S., Noguchi, A., and Saio, K. 1987. Microstructural studies of texturized vegetable protein products: Effects of oil addition and transformation of raw materials in various sections of a twin screw extruder. Food Microstructure 6:57-61.

Harris, P. L., Cuppett, S. L., Rupnow, J. H., Walker, C. E., Mumm, R. F., and Hanna, M. A. 1988. Optimization of initial moisture, pH and extrusion temperature of an acetone extracted maize gluten meal and soy extrudate for use in pet foods. Cereal Chem. 65:260-270.

Ibanoglu, S., Paul, A., and Geroge, D. H. 1996. Extrusion of tarhana: Effect of operating variables on starch gelatinization. Food Chem. 57:541-544.

Ilo, S., Tomschik, E. U., Berghofer, E., and Mundigler, N. 1996. The effects of extrusion operating conditions on the apparent viscosity and properties of extrudates in twin screw extrusion cooking of maize grits. Lebensm. Wiss. Technol. 29:593-598.

Jamin, F. F., and Flores, R. A. 1998. Effect of separation and grinding of corn dry-milled streams on physical properties of single-screw low speed extruded products. Cereal Chem. 75:775-779.

Jones, D., Chinnaswamy, R., Tan, Y., and Hanna, M. A. 2000. Physiochemical properties of ready-to-eat breakfast cereals. Cereal Foods World 45:164-168.

Kirby, A. R., Ollett, A. L., Parket, R., and Smith, A. C. 1988. An experimental study of screw configuration effects in the twin screw extrusion cooking of maize grits. J. Food Eng. 8:247-272.

Lovell, T. 1988. Nutrition and feeding of fish. Van Nostrand Reinhold: New York.

Martin-Cabrejas, M. A., Jaime, L., Karajha, C., Downie, A. J., Parkar, M. L., Lopez-Andreu, F. J., Maina, G., Esteban, R. M., Smith, A. C., and Waldron, K. W. 1999. Modifications to physiochemical and nutritional properties of hard to cook beans (*Phaseolus vulgaris* L.) by extrusion cooking. J. Agric. Food Chem. 47:1174-1182.

Martinez-Serna, M. D., and Villota, R. 1992. Reactivity, functionality, and extrusion performance of native and chemically modified whey proteins. In: Food Extrusion Science and Technology. J. L. Kokini, C. T. Ho, and M. V. Karwe, eds. Marcel Dekker: New York.

Mason, W. R., and Hoseney, R. C. 1986. Factors affecting the viscosity of extrusion cooked wheat starch. Cereal Chem. 63:436-441.

Matthey, F. P., and Hanna, M. A. 1997. Physical and functional properties of twin screw extruded whey protein concentrate – corn starch blends. Lebensm. Wiss. Technol. 30:359-366.

Mercier, C., and Feillet, P. 1975. Modification of carbohydrate components by extrusion-cooking of cereal products. Cereal Chem. 3:283-297.

Mercier, C., Linko, P., and Harper, J. M. 1989. Extrusion Cooking. AACC International: St. Paul, MN.

Miller, R. C. 1985. Low moisture extrusion: Effects of cooking moisture on product characteristics. J. Food Sci. 50:249-253.

Mohsenin, N. N. 1986. Physical properties of plant and animal materials: Structure, physical characteristics and mechanical properties. Gordon and Breach: New York.

Moore, D., Sanei, A., Van Hecke, E., and Bouvier, J. M. 1990. Effect of ingredients on physical/structural properties of extrudates. J. Food Sci. 55:1383-1387.

Ng, A., Lecain, S., Parker, M. L., Smith, A. C., and Waldron, K. W. 1999. Modifications of cell wall polymers of onion waste. III. Effect of extrusion cooking on cell wall material of outer fleshy tissues. Carbohydr. Polym. 39:341-349.

Nielsen, E. 1976. Whole seed processing by extrusion cooking. J. AOCS 53:305-309.

Oliveira, M. A., Moller, H. S., Haaland, H., and Rosenlund, G. 1992. The effect of process parameters on expansion of extruded fish feed. In: Food Extrusion Science and Technology. J. L. Kokini, C. T. Ho, and M. V. Karwe, eds. Marcel Dekker: New York.

Park, K. B. 1976. Elucidation of extrusion puffing process. PhD thesis. University of Illinois at Urbana-Champaign: Urbana, IL.

Paton, D., and Spratt, W. A. 1984. Component interactions in the extrusion cooking process: Influence of process conditions on the functional viscosity of the wheat flour system. J. Food Sci. 49:1380-1385.

Rahman, S. 1995. Food Properties Handbook. CRC Press: New York.

Riaz, M. N. 2000. Extruder in Food Applications. Technomic Publishing: Lancaster, PA.

Rolfe, L. A., Huff, H. E., and Hsieh, F. 2001. Effects of particle size and processing variables on the properties of an extruded catfish feed. J. Aquatic Food Product Technol. 10:21-33.

Rosentrater, K. A., Richard, T. L., Bern, C. J., and Flores, R. A. 2005. Small scale extrusion of corn masa by-products. Cereal Chem. 82:436-446.

Sandra, H. P. F., and Jose, A. G. A. 1993. Effect of phospholipid on protein structure and solubility in the extrusion of lung proteins. Food Chem. 47:111-119.

Shukla, C. Y., Muthukumarappan, K., and Julson, J. L. 2005. Effect of single screw extruder die temperature, amount of distillers dried grains with solubles (DDGS), and initial moisture content on extrudates. Cereal Chem. 82:34-37.

Singh, R. K., Nielsen, S. S., and Chambers, J. V. 1991. Selected characteristics of extruded blends of milk protein raffinate or nonfat dry milk with corn flour. J. Food Process. Preserv. 15:285-302.

Smith, A. C. 1992. Studies on the physical structure of starch based materials in the extrusion cooking process. In: Food Extrusion Science and Technology. J. L. Kokini, C. T. Ho, and M. V., Karwe, eds. Marcel Dekker: New York.

Thomas, M., Paul, T. H., Ton, V. V., Dick, J. V. Z., and Antonius, F. B. V. P. 1999. Effect of process conditions during expander processing and pelleting on starch modification and pellet quality of tapioca. J. Sci. Food Agric. 79:1481-1494.

Wu, Y. V., Rostagi, R. R., Sessa, D. J., and Brown, P. B. 1994. Utilization of protein rich ethanol co-products from corn in tilapia feed. J. AOCS 71:1041-1043.

Wu, Y. V., Rostagi, R. R., Sessa, D. J., and Brown, P. B. 1996. Effects of diets containing various levels of protein and ethanol coproducts from corn on growth of tilapia fry. J. Agric. Food Chem. 440:1491-1493.

USDA. 1999. Practical Procedures for Grain Handlers: Inspecting Grain. United States Department of Agriculture—Grain Inspection, Packers, and Stockyards Administration. Available online at http://151.121.3.117/pubs/primer.pdf. USDA: Washington, DC.

Zuilichem, D. J. V., and Stolp, W. 1984. Engineering aspects of single and twin screw extrusion cooking of biopolymers. In: Extrusion Cooking Technology. R. Jowitt, ed. Elsevier Applied Sciences: London.

[Received January 5, 2007. Accepted May 8, 2007.]