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# Twin-Screw Extrusion Processing of Distillers Dried Grains with Solubles (DDGS)-Based Yellow Perch (Perca flavescens) Feeds

Ferouz Y. Ayadi, *South Dakota State University*
Kurt A. Rosentrater, *United States Department of Agriculture*
Kasiviswanathan Muthukumarappan, *South Dakota State University*
Michael L. Brown, *South Dakota State University*

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ORIGINAL PAPER

# Twin-Screw Extrusion Processing of Distillers Dried Grains with Solubles (DDGS)-Based Yellow Perch (*Perca flavescens*) Feeds

**Ferouz Y. Ayadi · Kurt A. Rosentrater · Kasiviswanathan Muthukumarappan · Michael L. Brown**

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**Abstract** Increases in global aquaculture production, compounded with limited availabilities of fish meal for fish feed, has created the need for alternative protein sources. Twin-screw extrusion studies were performed to investigate the production of nutritionally balanced feeds for juvenile yellow perch (*Perca flavescens*). Five isocaloric (~3.06 kcal/g) ingredient blends, adjusted to a target protein content of 36.7% db, were formulated with 0%, 10%, 20%, 30%, and 40% distillers dried grains with solubles (DDGS) at an initial moisture content of 5–7%db, with appropriate amounts of fish meal, fish oil, whole wheat flour, corn gluten meal, and vitamin and mineral premixes. During processing, varying amounts of steam (6.9–9.7 kg/h) were injected into the conditioner and water (6.7–13.1 kg/h) into the extruder to modulate the cohesiveness of the final extrudates. Extrusion cooking was performed at 226–298 rpm using a 1.9 mm die. Mass flow rate and processing temperatures generally decreased with progressively higher DDGS content. Moisture content, water activity, unit density, bulk density, expansion ratio, compressive strength and modulus, pellet durability index, water stability, angle of repose, and color were extensively analyzed to quantify the effects of varying DDGS content on the physical properties of the final extrudates. Significant differences ($P<0.05$) among the blends were observed for color and bulk density for both the raw and extruded material, respectively, and for the unit density of the extruded product. There were also significant changes in brightness (L), redness (a), and yellowness (b) among the final products when increasing the DDGS content of the blends. Expansion ratio and compressive strength of the extrudates were low. On the other hand, all extruded diets resulted in very good water stability properties and nearly all blends achieved high pellet durability indices. In summary, each of the ingredient blends resulted in viable extrudates.

**Keywords** Aquaculture · DDGS · Extrusion · Physical properties · Protein · Twin-screw extruder

## Introduction

As a consequence of changes in energy policies based on moving the US towards greater energy independence from fossil fuels, renewable biofuel production is steadily growing. Amongst others, the production of conventional ethanol derived from corn starch reached 10.6 billion gallons in 2009 and will steadily grow up to a level of

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

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

M. L. Brown
Department of Wildlife and Fisheries Sciences,
South Dakota State University,
Brookings, SD 57007, USA

15 billion gallons by 2015 (ACE 2010; RFA 2010a). Viewed globally, the US produced almost half of the world's fuel ethanol production (RFA 2010b). Production of these enormous amounts of fuel also generated high quantities of coproducts, of which the major proportion was distillers grains with solubles (DDGS). In 2009, up to 30.5 million tons of distiller grains was produced (RFA 2010a). During the fermentation process, starch is converted to alcohol and carbon dioxide; consequently, DDGS contains approximately three times the amount of most nutrients, such as protein, fiber, minerals and fat, compared to corn (Spiehs et al. 2002; Jacques et al. 2003) and is therefore a suitable ingredient for many animal diets. Traditionally, it has been used for ruminant nutrition, and currently it is also being utilized as protein supplements in other terrestrial animal feeds (Ham et al. 1994; USDA 2006; Lim et al. 2009; De Godoy et al. 2009; Saunders and Rosentrater 2009). Beyond that, other research has been examining DDGS as a food ingredient for humans (Rosentrater and Krishnan 2006).

The world's increasing demands for seafood products makes aquaculture one of the fastest growing animal food-producing sectors, with a worldwide production of about 51.7 million tons in 2006 (FAO 2009). With an annual growth rate of 6.9% from 1970 to 2006, it exceeded by far the world's annual population growth rate of 1.5% between 1975 and 2009, and will soon supply the market with higher quantities of farmed fish food than the harvest yield of capture fisheries (FAO 2009; UN 2007). Great amounts of feed are required to produce these large quantities of seafood. Fish diets predominantly contain fish meal, which is commonly used to provide proteins that are essential for metabolism. In 2006, about 20.2 million tons of raw fish were consumed as bulk for fish meal; about 3.06 million tons (56%) of the global fish meal production was used by the aquaculture industry (FAO 2009), the remaining 44% were utilized in livestock and other animal feed. These high demands on fish meal and seafood have increasingly lead to reductions of wild fish stocks in the ocean, and will eventually reach the limit of available natural sources for seafood and fish meal (Naylor and Burke 2005). As a result, fish feed represents the largest aquaculture operating cost, in which fish derived protein is the most expensive part, and aquaculturists have begun looking for alternative, less-expensive dietary proteins (El-Sayed 1998; Webster et al. 1999; Tidwell et al. 2000; Sardar et al. 2009).

Much research has been pursued to find sustainable, cost efficient, and compatible alternatives with high protein availability for fish. In recent years, studies on plant protein sources, such as peas (Carter and Hauler 2000; Schulz et al. 2007), lupins (Glencross et al. 2006; Farhangi and Carter 2007), rapeseed meal (Przybyl et al. 2006; Wu et al. 2006), corn gluten meal (Goda et al. 2007; Guimarães et al. 2008), and cottonseed meal (Mbahinzireki et al. 2001; Robinson and Li 2008), have been successful in using these ingredients as partial replacements for fish meal. Based on low palatability, potential presence of anti-nutritional factors, low protein content or imbalances in amino acids in certain plant feedstocks (Carter and Hauler 2000; Hansen et al. 2007), high inclusion levels of plant proteins in fish feed are often only achievable with supplementations of essential amino acids (such as methionine and lysine), and preprocessing of the plant materials (Francis et al. 2001; Oliva-Teles and Gonçalves 2001).

DDGS presents another potential alternative for protein. In contrast to other plant-derived nutrient sources, such as soybean meal (SBM; which is one of the most studied and widely used protein supplements in fish diets) DDGS contains none of the anti-nutritional factors found in most plant protein sources (US Grains Council 2008), and is less expensive than SBM on a per unit protein basis (Garcia and Taylor 2006; Lim et al. 2007). However, lysine and methionine must be supplemented when using DDGS in fish diets (Cheng and Hardy 2004; Stone et al. 2005). In preceding studies, DDGS supplemented with vitamins and minerals could be successfully integrated in diets for several fish species, such as channel catfish, including 30–40% DDGS or up to 70% DDGS with 0.4% crystalline L-lysine supplementation (Webster et al. 1991; Robinson and Li 2008; Lim et al. 2009), rainbow trout, with 22.5% DDGS inclusion level or up to 75% DDGS supplemented with lysine and methionine (Cheng and Hardy 2004), and Nile tilapia, including 20% DDGS, 30% with an animal-based protein or up to 40% DDGS inclusion level supplemented with lysine (Coyle et al. 2004; Lim et al. 2007).

Yellow perch (*Perca flavescens*) is a fish species that is widespread in North America (Pierron et al. 2009), and historically, most of the commercial food fish supply had been provided from capture fisheries in the Great Lakes region. Currently, aquaculture for yellow perch is increasing to meet the demands (Malison 2000). After being trained to accept prepared food, pellets can be used for their diet (Heidinger and Kayes 1986). To date, only a few nutritional studies have been conducted on yellow perch. Recommended dietary crude protein requirements are in the range of 21–27% (Ramseyer and Garling 1998). Kasper et al. (2007) performed formal evaluations of alternative proteins in diets for yellow perch. They concluded that yellow perch are able to utilize solvent-extracted dehulled SBM and expelled-extruded SBM as main feed ingredients to replace fish meal, and suggested levels of 300 g/kg diet.

Extrusion cooking is the primary method of aquafeed production. It can improve feed physical properties as well as fish growth performance and digestibility (which is influenced by starch content and anti-nutritional factors) of feed containing commonly used plant protein sources (Booth et al. 2002; Allan and Booth 2004; Barrows et al. 2007; Sørensen et al. 2009). Investigating the use of DDGS in fish diets notwithstanding, few studies have been conducted to analyze the processing aspects of DDGS feeds. Previous single- and twin-screw extrusion studies were conducted to produce floating feeds for tilapia and channel catfish, respectively, and to investigate the effects of various levels of DDGS, die dimensions, screw speeds, barrel temperatures, ingredient moisture contents, and protein contents, respectively, on various extruder processing parameters and extrudate physical properties (Chevanan et al. 2007a, b, c; 2008; 2009; 2010; Kannadhason et al. 2009a, 2009b, 2010; Rosentrater et al. 2009a, 2009b). So far, examination of the processing of DDGS-based feeds for yellow perch has only been conducted by Ayadi et al. (2009a, b). They determined that using single screw extrusion diets containing between 10% and 50% DDGS could result in viable extrudates, depending upon processing conditions used.

Single-screw food extrusion was established in the 1940s to produce macaroni and cereal pellets. Single-screw extruders are high-temperature, short-time bioreactors that can process a wide range of raw materials (Harper 1989). Twin-screw extruders started to be used in the 1970s and have the advantages handling wider varieties of ingredients due to their self-wiping properties and greater conveying angles (Harper 1989). Compared to single-screw extruders, twin-screw extruders can use two co-rotating or counter-rotating screws in the barrel. In contrast to counter-rotating extruders, where high localized pressures and large separation forces can appear, and limitations in operational screw speeds exist, co-rotating screws rotate in the same directions and are generally preferred (Brent 1989). More information about extrusion processing can be found in Mercier et al. (1989), Kokini et al. (1992), Chang and Wang (1998), Riaz (2000), and other literature sources. Combining heat and pressure generally leads to better digestibility of food, and can be used to sterilize the feedstock; this enhances the shelf life of the final product. Extrusion of fish feed often results in fewer problems with disease and requires fewer quantities to increase weight gain (Kiang 1998). Furthermore, extrusion cooking allows control of pellet density, greater water stability, enhanced durability, and better production efficiency and versatility (Kiang 1998; Brent 1989).

The objectives of this study were: (1) to develop feed for juvenile yellow perch using DDGS as an alternative protein source and (2) to examine the effects of varying DDGS content on the physical properties of the extrudates and on extruder processing behavior.

## Materials and Methods

### Feed Blend Preparation

Five isocaloric (3.06 kcal/g) ingredient blends (Fig. 1) with a similar protein content of 36.7% db, each with increasing contents of DDGS (0%, 10%, 20%, 30%, and 40%db) and decreasing amounts of herring fish meal, varying amounts of Celufil and menhaden oil, but a nearly constant ratio of whole wheat flour, corn gluten meal, vitamin and mineral mix (Table 1), were used to prepare nutritionally balanced diets for juvenile yellow perch. Table 2 shows the physical properties of the raw feed blends. Approximately 23 kg of each blend were extruded. DDGS was provided by Poet Nutrition (Sioux Falls, SD, USA) and were ground with a pilot-scale mill (Model DA 506, Fitzpatrick Co., Elmhurst, IL, USA) to a particle size of 500 µm, in order to achieve uniformity before extrusion. Herring fish meal was obtained from Lortscher Agri Service, Inc. (Bern, KS, USA). The Celufil (used as a fiber source) was purchased from USB Corporation (Cleveland, OH, USA); menhaden fish oil from Omega Protein, Inc. (Houston, TX, USA); whole wheat flour from Bob's Red Mill Natural Foods, Inc. (Milwaukie, OR, USA); corn gluten meal from Consumers Supply Distributing Company (Sioux City, IA, USA); vitamin C from DSM Nutritional Products France SAS (Village-Neuf, France); vitamin mix and mineral mix from Lortscher Agri Service, Inc. (Bern, KS, USA). The corn gluten meal, the menhaden fish oil, and the whole wheat flour were mixed (Model 600, Hobart Corporation, Troy, OH, USA) for about 3 min until the fish oil dispersed well into the mixture; then the mineral and vitamin mix were added to the mixture; this premix was then added to the rest of the ingredients and mixed in a twin shell dry blender (The Patterson-Kelley Co. Inc., East Stroudsburg, PA, USA) at 60 rpm to produce a homogeneous bulk.

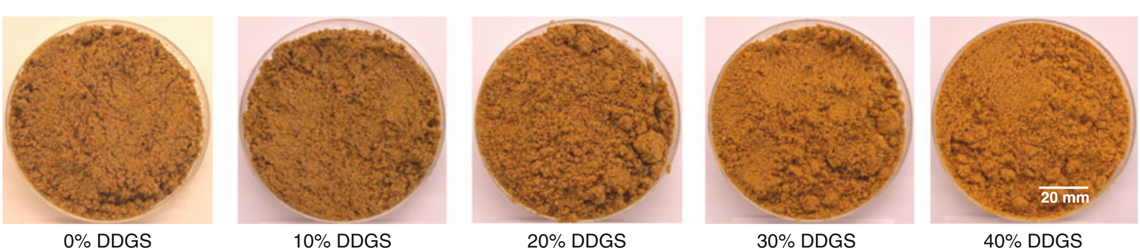



**Fig. 1** Raw blends

**Table 1** Ingredient components (g/100 g) in the feed blends and their compositions (dry basis) used in the study

| Ingredients (% db) | Diet 1 | Diet 2 | Diet 3 | Diet 4 | Diet 5 |
|---|---|---|---|---|---|
| | Dry weight of ingredients (g/100 g) | | | | |
| DDGS[a] | 0.00 | 9.98 | 19.98 | 30.00 | 40.05 |
| Fish meal (herring)[b] | 40.35 | 30.30 | 20.22 | 10.12 | 0.00 |
| Corn gluten meal[c] | 25.75 | 25.78 | 25.81 | 25.84 | 25.87 |
| Whole wheat flour[d] | 14.99 | 15.01 | 15.02 | 15.04 | 15.06 |
| Celufil[e] | 9.96 | 9.31 | 8.57 | 7.92 | 8.31 |
| Menhaden oil[f] | 5.54 | 6.21 | 6.97 | 7.64 | 7.27 |
| Vitamin/mineral premix[g] | 2.92 | 2.93 | 2.93 | 2.93 | 2.94 |
| Vitamin C mix[h] | 0.50 | 0.50 | 0.50 | 0.50 | 0.50 |
| Total | 100.00 | 100.00 | 100.00 | 100.00 | 100.00 |
| | Diet composition (% db) | | | | |
| Protein | 42.66 | 39.70 | 36.74 | 33.78 | 30.82 |
| Fat | 10.12 | 10.36 | 10.70 | 10.94 | 10.08 |
| Crude fiber | 2.40 | 3.16 | 3.92 | 4.68 | 5.44 |
| Ash | 5.25 | 4.41 | 3.57 | 2.73 | 1.89 |

All blends were formulated on a dry basis

[a] Dakota Gold HP DDG, Poet Nutrition (Sioux Falls, SD, USA)

[b] Lortscher Agri Service, Inc. (Bern, KS, USA)

[c] Corn gluten meal, Consumers Supply Distributing Company (Sioux City, IA, USA)

[d] Whole wheat flour, Bob's Red Mill Natural Foods, Inc. (Milwaukie, OR, USA)

[e] Celufil-Non Nutritive Bulk, USB Corporation (Cleveland, OH, USA)

[f] Omega Protein, Inc. (Houston, TX, USA)

[g] Lortscher Agri Service, Inc. (Bern, KS, USA)

[h] Rovimix Stay-C 35, DSM Nutritional Products France SAS (Village-Neuf, France)

## Extrusion

Extrusion was performed using a co-rotating, fully intermeshing, self-wiping, twin-screw extruder (Wenger TX-52, Sabetha, KS, USA), with a feed hopper and preconditioner. The extruder had 52 mm diameter twin screws, with a barrel length of 1340 mm, and a 25.5:1 barrel length-to-diameter ratio. The screw speed could operate from 100 to 1,800 rpm, and the barrel temperatures could be adjusted from 60°C to150°C. The screw had 25 individual sections (Fig. 2), and the configuration from the feeding section down the length of the barrel to the die section consisted of: four conveying screws, three shear locks, one conveying screw, one conveying screw backward, three conveying screws, one conveying screw backward, four conveying screws, one shear lock, one interrupted flight conveying screw, one conveying screw, one interrupted flight conveying screw, one shear lock, and one cone-shaped screw at the end. The raw material was scooped in the feed hopper and conveyed by screws into the preconditioner. The conditioner was an intermediate unit operation of the extruder, where the feedstock was heated and water at rates between 6.72 and 11.10 kg/h and steam at rates between 6.86 and 9.70 kg/h was added to adjust the blends to specific temperature and moisture contents. The conditioned ingredients were conveyed into the extruder at a feeder speed varying between 12 and 16 rpm (1.3 and 1.7 rad/s). The screw speed was maintained at levels varying between 226 and 298 rpm (23.7 and 31.2 rad/s). Between the feeding zone and the die section the barrel was divided into eight different temperature zones that were maintained at varying temperature combinations (15–90°C; see Table 3 for the process settings used during processing) depending on the actual temperature of the temperature zones and the final

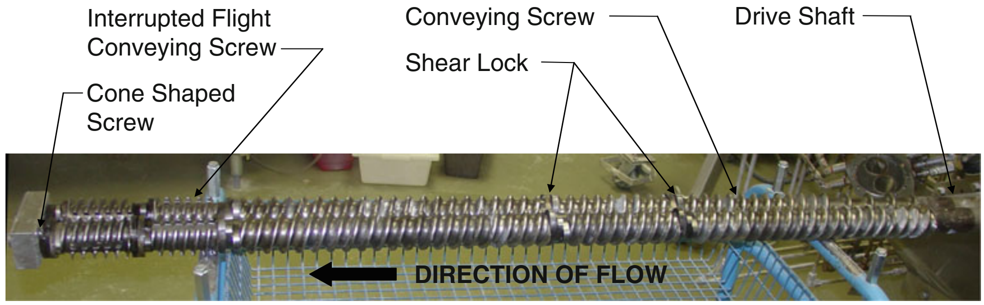
**Table 2** Physical properties of the raw feed blends

| Properties | Diet (% DDGS) | | | | |
|---|---|---|---|---|---|
| | 0 | 10 | 20 | 30 | 40 |
| MC raw (% db) | 4.98a (0.16) | 5.80ab (0.42) | 5.86ab (0.35) | 6.55b (0.80) | 6.63b (0.72) |
| $a_w$ (−) | 0.37a (0.01) | 0.39b (0.01) | 0.39b (0.00) | 0.4d (0.01) | 0.41d (0.00) |
| BD (kg/m$^3$) | 381.55a (3.85) | 356.08a (4.63) | 401.74b (4.10) | 409.65c (2.14) | 440.71d (1.54) |
| $k$ (W/(m °C)) | 0.06a (0.01) | 0.07bc (0.01) | 0.07ab (0.01) | 0.07ab (0.00) | 0.08c (0.00) |
| α (mm²/s) | 0.14b (0.01) | 0.13ab (0.01) | 0.13ab (0.01) | 0.13ab (0.00) | 0.13a (0.01) |
| $L$ (−) | 36.70a (0.18) | 39.09b (0.54) | 40.37bc (0.53) | 41.3c (1.64) | 45.76d (0.22) |
| $a$ (−) | 7.08a (0.04) | 7.79b (0.02) | 9.06c (0.07) | 10.59d (0.31) | 11.92e (0.16) |
| $b$ (−) | 16.41a (0.04) | 18.05b (0.16) | 19.78c (0.17) | 21.47d (0.52) | 23.98e (0.06) |

Means followed by similar letters for a given dependent variable are not significantly different at $P<0.05$, LSD. Values in parentheses are standard deviation

*MC* moisture content, $a_w$ water activity, *BD* bulk density, $k$ thermal conductivity, α thermal diffusivity, *L, a, b* Hunter color parameters

**Fig. 2** Screw profile used in the extruder




product property. In addition, depending on extrudates physical characteristics, the amount of water entered the extruder was applied at levels between 9.00 and 13.14 kg/h. The two dies each had circular openings of 1.9 mm. A rotating cutter with three blades was positioned at the end of the dies and was adjusted to specific speeds to cut the exiting extrudates to desired lengths. Furthermore, during extrusion processing, the moisture (% db) content at the die was monitored, and the temperatures (°C) of the raw material, the blend in the preconditioner, and the extrudates exiting the extruder, were measured with an infrared thermometer (Model 42540, Extech Instruments Corporation, Waltham, MA, USA). Table 4 shows the behavior during processing for each blend.

## Processing Behavior

Mass flow rate (MFR) was determined by collecting two ($n$=2) samples of extrudates exiting the die. Extrudates were collected at intervals of 30 s during extrusion processing and weighed on an electronic balance (Defender 3000 Series, Ohaus, Pine Brook, NJ, USA).

Moisture content (MC) was determined according to AACC method (2000) using a laboratory oven (Thelco Precision, Jovan, Wincester, VA, USA) at 135°C for 2 h. Moisture content of three ($n$=3) raw blend samples, exiting the preconditioner, and extrudates exiting the die were measured.

After the prepared blends were extruded, they were cooled for 72 h at room temperature (21±1°C), and they were then dried in a laboratory oven (Model TAH-500, The Grieve Corporation, Round Lake, IL, USA) for 24 h at 45°C, and then subjected to extensive physical property testing.

After cooling, triplicates ($n$=3) were then analyzed for moisture content (% db), water activity (−), bulk density (kg/m$^3$), pellet durability index (%), water stability (min), angle of repose (mm), and color (−); length and diameter (mm), unit density (kg/m$^3$), expansion ratio (−), compressive strength (MPa), and compressive modulus (MPa) were determined with $n$=10 replications.

## Moisture Content and Water Activity

The moisture content of the raw materials and extrudate samples for each blend were determined using a laboratory oven (Thelco Precision, Jovan, Wincester, VA, USA) at 135°C for 2 h according to AACC (2000).

The raw material and extrudate samples from each treatment were analyzed for water activity ($a_w$) with a

**Table 3** Process settings used during extrusion of each diet

| Parameters | Diet (% DDGS) | | | | |
|---|---|---|---|---|---|
| | 0 | 10 | 20 | 30 | 40 |
| Conditioner steam (kg/min) | 6.86 (0.18) | 6.88 (0.20) | 6.96 (0.25) | 9.70 (0.02) | 7.13 (0.14) |
| Extruder Water (kg/h) | 9.15 (0.21) | 11.10 (0.00) | 13.14 (0.00) | 6.72 (0.00) | 9.60 (0.00) |
| Set Temperatures (°C) | | | | | |
| Head 2 zone 1 | 25.00 | 25.00 | 25.00 | 25.00 | 25.00 |
| Head 3 | 35.00 | 35.00 | 35.00 | 10.00 | 10.00 |
| Head 4 zone 2 | 15.00 | 15.00 | 15.00 | 15.00 | 15.00 |
| Head 5 zone 3 | 65.00 | 65.00 | 65.00 | 15.00 | 15.00 |
| Head 6 zone 4 | 65.00 | 65.00 | 65.00 | 15.00 | 15.00 |
| Head 7 zone 5 | 90.00 | 90.00 | 90.00 | 15.00 | 37.50 |
| Head 8 | 80.00 | 80.00 | 80.00 | 81.50 | 65.00 |
| Head 9 zone 6 | 80.00 | 80.00 | 80.00 | 81.50 | 65.00 |

Values in parentheses are standard deviation

**Table 4** Treatment effects on the measured processing behavior

| Parameters | Diet (% DDGS) | | | | |
|---|---|---|---|---|---|
| | 0 | 10 | 20 | 30 | 40 |
| MC conditioner (% db) | 31.66d (1.21) | 35.22e (1.14) | 20.46c (0.72) | 15.61a (0.78) | 17.71b (0.98) |
| MFR (kg/min) | 1.08c (0.06) | 1.08c (0.00) | 0.98b (0.03) | 0.90ab (0.03) | 0.82a (0.03) |
| MC die (% db) | 51.84c (0.62) | 62.03d (2.14) | 47.96b (1.23) | 34.93a (0.44) | 32.96a (0.23) |

Means followed by similar letters for a given dependent variable are not significantly different at $P<0.05$, LSD. Values in parentheses are standard deviation

*MC* moisture content, *MFR* mass flow rate

water activity measuring system ($a_w$ Sprint TH-500, Novasina, Pfäffikon, Switzerland). A sample bowl was filled with each sample and then placed in the measuring chamber of the instrument for analyzing the water activity.

## Expansion Ratio and Unit Density

Extrudates at approximate lengths of 25.4 mm were weighed on an analytical balance (Adventurer™, Item No: AR 1140, Ohaus Corp. Pine Brook, NJ, USA) and measured with a digital caliper (Digimatic caliper, Model No: CD-6"C, Mitutoyo Corp., Tokyo, Japan) to determine their diameter. According to Rosentrater et al. (2005) the unit density (UD) was calculated as the ratio of the mass $M$ (kg) to the volume $V$ ($m^3$) of each measured and weighed sample, assuming a cylindrical shape for each extrudate:

$$UD = \frac{M}{V} \quad (1)$$

The ratio of the diameter of the dry extrudates, measured with a digital caliper (Digimatic caliper, Model No: CD-6"C, Mitutoyo Corp., Tokyo, Japan), to the diameter of the die nozzle (1.9 mm) was used to determine the expansion ratio (ER). The results were displayed as the mean of ten measurements.

## Bulk Density

Bulk density (BD) was determined as the ratio of the mass of extrudates and raw material, respectively that they filled up to a given bulk volume and measured using a standard bushel tester (Seedburo Equipment Company, Chicago, IL, USA) following the method recommended by USDA (1999).

## Compressive Strength and Modulus

For each treatment, the extruded samples of equal length were tested for their compressive strength and modulus (i.e., stiffness) using a dual column universal materials testing machine (Model No. 5564, Instron, Canton, MA, USA).

## Pellet Durability Index

The pellet durability (PDI) index was determined according to Method S269.4 (ASAE 2004). Approximately 100 g extrudate samples of each blend were manually sieved (USA standard testing, ASTM E-11 specification, Daigger, Vernon Hills, IL, USA) for about 10 s and then tumbled in a pellet durability tester (model PDT-110, Seedburo Equipment Company, Chicago, IL, USA) for 10 min. Afterwards, the samples were again sieved for approximately 10 s and weighed on an electronic balance (Explorer Pro, Model: EP4102, Ohaus, Pine Brook, NJ, USA). For blend 1–4 sieve No. 7 (2.80 mm) was used, but for blend 5 and 6 sieve No. 8 (2.36 mm) was used. Relating the extrudates sample weights before and after tumbling, the PDI was calculated as

$$PDI = \left(\frac{Ma}{Mb}\right) \times 100 \quad (2)$$

where, $M_a$ was the mass (g) after tumbling and $M_b$ was the sample mass (g) before tumbling.

## Water Stability

For extrudates of each blend, a 1-g sample was placed in 200 mL of distilled water and stirred with a magnet stirrer (PMC No. 524 C, Barnstead International, Dubuque, IA, USA) at low speed until the extrudates broke to determine the stirred water stability. In the case of still water stability (WS), the same process was used without stirring. Water stability of aquafeeds gives conclusions about the overall performance of the extrudates. It represents one of the most important properties of the feed, since water stability determines how much time it takes before an extrudate breaks and, therefore, is no longer available for the fish. In addition, disintegration of extrudates involves leaching of nutrients into water. There were two methods used to draw conclusions about WS: (1) to determine how long extrudates were stable when spun around and (2) how long extrudates were stable when they were not moved, as compared to the act of sinking to the ground tank bottom when not being consumed by fish.

### Angle of Repose

Angle of repose was determined by letting an extrudate sample fall onto a 99-mm circular plate, using a standard bushel tester (Seedburo Equipment Company, Chicago, IL, USA) with the measuring cup turned upside down. Angle of repose (AOR) was determined by calculating the $\tan^{-1}$ of (the height of the pile/radius of its base (49.5 mm)).

### Color

A spectrophotometer (LabScan XE, HunterLab, Reston, VA, USA) was used to determine the extrudate color, where L* quantified the brightness/darkness, a* the redness/greenness and b* the yellowness/blueness of the samples. Likewise, the color was tested on the raw material blends.

### Thermal Properties

Each raw blend was analyzed for thermal conductivity (k) and thermal diffusivity ($\alpha$). From each blend, raw material was placed in a 250 mL beaker. The sensor needle of the thermal properties analyzer (KD2 Thermal Properties Analyzer, Decagon Devices, Inc., Pullman, WA, USA) was inserted into the medium, and the thermal conductivity and thermal diffusivity were measured three times on three different positions in the material at room temperature (21±1°C).

### Statistical Analysis

All data were analyzed with SAS (1999) software (SAS Institute, Cary, NC, USA), using a type I error rate ($\alpha$) of 0.05 by analysis of variance, to determine if there were significant differences between treatments and, if differences existed, post hoc least significant difference (LSD) tests were used to determine where they occurred. Pearson linear correlation analyses were also conducted among all independent and dependent variables to test for linear relationships.

## Results and Discussion

DDGS level influenced both the processing behavior as well as the resulting extrudate characteristics, both externally (Fig. 3, which shows that the extrudates changed in texture and appear to be more homogeneous with increasing DDGS levels), and internally (Fig. 4, which illustrates cross sections of the final extrudate samples at a magnification of ×60).

### Moisture Content

Extrudate physical properties were impacted by the level of DDGS (Table 5). Moisture content has a major impact on the extrusion process. It does not only affect most of the extrusion processing parameters such as mass flow rate and color (Chevanan et al. 2007a), but it is also a determining factor for the cohesiveness of the final product. Variations in steam and water may result in soft, brittle or, as desired, cohesive extrudates. Another important effect of MC on extrudates' texture is that water plasticizes proteins (Zhang et al. 2001) and makes the final product less brittle and fragile. Analyzing the moisture content of the raw materials (on a dry basis), a slight increase in MC occurred when adding more DDGS to the diets. The levels varied between 4.98% MC for the control diet and 6.63% MC for the blend including 40% DDGS. Similar observations for increases in MC of the raw blends with higher DDGS levels were made in different extrusion studies (Chevanan et al. 2007c, a, b; Ayadi et al. 2009a, b). Regarding the samples taken from the conditioner and samples taken directly when exiting the die, no clear pattern of changes in MC could be observed. MC for the samples from the conditioner showed significant differences between all blends. The blend including 30% DDGS resulted in the lowest MC of 15.61%, and the blend with 10% showed the highest MC at 35.22%. These findings are not consistent with the amount of steam added to the conditioner cylinder. The largest amount of steam,


**Fig. 3** Resulting extrudates


**Fig. 4** Cross-sections of the resulting extrudates (magnification of ×60)

9.70 kg/min, was added to the blend that yielded the lowest amount in MC. The remaining blends were supplied with 6.86 and 7.13 kg/min of steam.

Similar effects in MC of the product exiting the die could be observed when injecting varying amounts of water into the extruder barrel. The blend at the lowest die MC (32.96%) had a water injection with 9.60 kg/h and the blend at the highest die MC (62.03%) was supplied with 11.10 kg/h of water. The blend with 20% DDGS was supplied with the largest amount of water (13.14 kg/h) and yielded a die moisture content of 47.96%; no clear relation to the water supply could be determined.

MC of the final extrudates showed differences that were similar to the raw materials. An almost constant increase in MC together with an increase in DDGS could be determined, except that the blend containing 20% DDGS had the lowest MC (1.10%). The blend containing 40% yielded the highest MC (3.70%). Similar results were obtained by Ayadi et al. (2009a, b).

As expected, extrudates MC was lower compared to the raw material. On the one hand, this was due to an extensive flashing-off of internal moisture at the die exit caused by the sudden pressure drop from high values inside the extruder to atmospheric levels outside. On the other hand, the long time period of drying the final products at room temperature and additionally in an oven, resulted in the low extrudate MC. This is in agreement with results discussed by Ayadi et al. (2009a, b).

### Water Activity

Water activity is defined as the ratio between the pressure of a solution to that of pure water under the same condition (Koop et al. 2000). It measures the free water existing in a

**Table 5** Treatment effects on the extrudate physical properties

| Properties | Diet (% DDGS) | | | | |
|---|---|---|---|---|---|
| | 0 | 10 | 20 | 30 | 40 |
| MC extrudate (% db) | 1.32a (0.43) | 1.64a (0.81) | 1.10a (0.05) | 2.50b (0.43) | 3.70c (0.22) |
| $a_w$ (−) | 0.11a (0.01) | 0.13b (0.00) | 0.14bc (0.00) | 0.14c (0.00) | 0.12a (0.00) |
| ER (−) | 1.43b (0.02) | 1.35a (0.05) | 1.35a (0.03) | 1.44b (0.03) | 1.36a (0.04) |
| UD (kg/m$^3$) | 855.31a (47.42) | 917.05b (65.74) | 911.19b (52.98) | 867.89ab (64.10) | 990.09c (59.77) |
| BD (kg/m$^3$) | 427.59a (5.54) | 447.19c (2.72) | 463.89d (1.68) | 434.00b (0.52) | 470.29e (0.24) |
| Compressive strength (MPa) | 1.13a (0.29) | 1.51ab (0.33) | 1.91b (0.43) | 1.58b (0.19) | 3.00c (0.86) |
| Compressive modulus (MPa) | 12.39a (4.60) | 17.27a (4.15) | 17.68a (10.64) | 14.11a (10.64) | 61.05b (25.49) |
| PDI (%) | 93.35b (0.36) | 96.26c (0.37) | 96.77c (0.54) | 89.89a (0.89) | 96.27c (0.12) |
| WS stir (min) | >30 (0.00) | >30 (0.00) | >30 (0.00) | >30 (0.00) | >30 (0.00) |
| WS still (min) | >30 (0.00) | >30 (0.00) | >30 (0.00) | >30 (0.00) | >30 (0.00) |
| AOR (°) | 36.28c (0.43) | 35.77c (0.44) | 35.51c (1.17) | 33.15b (0.47) | 31.78a (0.48) |
| *L* (−) | 39.51c (0.39) | 40.16c (0.11) | 39.73c (0.55) | 34.26b (0.64) | 30.16a (0.45) |
| *a* (−) | 4.38a (0.03) | 4.87b (0.09) | 5.61c (0.05) | 8.10d (0.13) | 10.30e (0.09) |
| *b* (−) | 15.51a (0.16) | 16.17b (0.08) | 16.62c (0.17) | 17.07d (0.27) | 15.76a (0.21) |

Means followed by similar letters for a given dependent variable are not significantly different at $P<0.05$, LSD. Values in parentheses are standard deviation

*MC* moisture content, $a_w$ water activity, *ER* is expansion ratio, *UD* unit density, *BD* bulk density, *PDI* pellet durability index, *WS* water stability, *AOR* angle of repose, *L, a, b* Hunter color parameters material that is not bound to molecules and therefore available for the growth of microorganisms, such as bacteria, molds, and yeast. Hence, products with a low $a_w$ have a lower chance for spoilage, whereas materials with a high $a_w$ are at a greater risk for quick spoilage; water activity determines the shelf life of a product and is generally considered to guarantee a stable product with long storage stabilities at levels below 0.6 (Lowe and Kershaw 1995).

Water activities of the raw materials varied between 0.37 and 0.41 and partially increased with higher DDGS levels in the blend. The water activity of the extrudates showed no significant differences between all blends and varied between 0.11 for the blend with 0% DDGS to 0.14 for the blends with 20% and 30% DDGS content. The values for $a_w$ were very low and can be traced back to the fact that extrudates were air- and oven-dried post extrusion. Thus, they all had high expected shelf-life properties. Likewise, it can be said that the raw materials had a low-water activity that allows long storage times without the risk of fast spoilage.

### Expansion Ratio

The degree of expansion determines the structure and therefore the texture of the extrudates. During expansion, water is nucleated and forms bubbles in the extruded material upon die exit (Arhaliass et al. 2009). The internal structure of the expanding melt is affected by the radial expansion that occurs at the die exit and results in different textures of the extrudates (Arhaliass et al. 2003). ER is generally inversely related to the unit density (Bhatnagar and Hanna 1996). The levels of expansion ratio were calculated between 1.35 and 1.44 and hence were very low. Diets including 10%, 20%, and 40% showed no significant differences to each other in ER; the control diet showed no significant difference to the blend with 30% DDGS. Contrary to the initially mentioned assumption, there was no noticeable relation determined between UD and expansion ratio. Similarly, Ayadi et al. (2009a, b) observed almost no significant differences in expansion ratio when performing single-screw extrusion studies on yellow perch.

Expansion depends on the flashing of water vapor and flow properties of molten starch (Colonna et al. 1989). High expansion ratios were not expected due to the fact that DDGS naturally has low starch contents that vary between 4.7% and 5.9% (Rosentrater and Muthukumarappan 2006) and that the present diets were designed for fish feed, accordingly, containing high amounts of protein. Furthermore, expansion ratios were only calculated based upon radial expansion, neglecting longitudinal and volumetric expansions.

### Unit Density

Extrudates decreased in length as DDGS level increased (Fig. 3). Unit density measures the mass of individual extrudates to their volume. In contrast to bulk density, unit density quantifies the density for a single extrudate, whereas BD relates to the volume occupied by a given mass. Unit density is related to the floatability of the feed, and is therefore a decisive factor in aquaculture feeds. Values for UD varied from 855.31 kg/m$^3$ for the control diet to 990.09 kg/m$^3$ for the blend including 40% DDGS content, which was an overall increase of 15.8%. Blends with 10%, 20%, and 30% DDGS content showed no significant differences among each other. Furthermore, there was no significant difference between the control diet and the blend containing 30% DDGS; only the diet with the 40% DDGS level, which had the highest UD, was significantly different from all other blends. In other studies, Chevanan et al. (2007b) observed an increase of 159% in unit density when increasing the DDGS content from 20% to 60% in twin-screw extrusion for fish feed. Studies in single-screw extrusion for yellow perch feed determined an increase of 17% when raising DDGS levels from 10% to 50% (Ayadi et al. 2009a, b). Hence, it can be assumed that changes in UD can be related to the amounts of DDGS and fish meal added to the ingredients, since the remaining ingredient quantities were not altered. Although all extrudates were found to be below the density of water (1,000 kg/m$^3$), none of the samples floated and instead sank slowly to the bottom of the beaker. The same reaction of non-floating DDGS extrudates with lower UD than water was observed by Ayadi et al. (2009a, b). This behavior could be ascribed to the porous structure of the pellet, which might have rapidly absorbed the water and let them slowly sink.

### Bulk Density

Bulk density is a key parameter in the design and utilization of storage reservoirs (Rosentrater 2006). It determines the size of the storage volumes that are needed for extrudates and feedstocks. At the final step of the extrusion process, small voids inside the extrudate's texture are formed by expansion and hence, affect the bulk density. Likewise, the space between extrudates and particles of the feedstock affects the size of storage space.

With increasing DDGS inclusion levels from 10% to 40%, the values for BD of the raw material increased from 356.08 to 440.71 kg/m$^3$ and resulted in a steady increase of 23.8%. Regarding the extrudates, similarly, a significant increase of 10.0% in bulk density from 427.59 to 470.20 kg/m$^3$ could be observed when raising the DDGS content from 0% to 40% (except in the diet including 30% DDGS, which did not conform, and showed the second lowest BD of 434.00 kg/m$^3$). Chevanan et al. (2007b) determined an increase of 61.4% in bulk density for extrudates when raising the DDGS content from 20% to 60% in twin-screw extrusion of aquaculture feed. Since the fishmeal was constantly lowered in the initial ingredient blends with increasing levels of DDGS, and the amount of fish oil added was enhanced for the diets, it can be assumed that the bulk density increased due to the higher DDGS and/or fish oil levels.

### Compressive Strength and Modulus

Compression tests were performed to determine the capacity of the extrudates to resist forces without breaking or deforming. Tests were completed when extrudate samples reached the yield point (i.e., when changes in the structure of the material were permanent, which occurred when the first force peak was reached). The values for compressive strength ranged from 1.13 to 1.91 MPa for diets containing 0–30% DDGS. The diet containing 40% reached the highest value of 3.00 MPa. This significant difference from all other blends could be ascribed to the significantly higher MC of the diet, since the moisture content in a material generally affects the hardness (in this case the fracturability) of the extruded product (Ding et al. 2005; Li et al. 2005). Water plasticizes the starch and protein and reduces its viscosity (Zhang et al. 2001; Li et al. 2005). Therefore, the high value of the compressive strength could be ascribed to the lower porosity of the extrudates' texture. This could also be supported when comparing the compressive strength with the UD: the higher the UD, the higher the values for the compressive strength. Regarding all the values, however, the levels for the compressive strength were very low and showed that the extrudates did not resist high forces and broke quickly. Similar observations were determined by Ayadi et al. (2009a, b) who also noted low levels for compressive strength between 3.06 and 4.28 MPa for DDGS-based yellow perch feeds, which varied with the moisture content.

Compressive modulus gives indicated the stiffness of the material. Except for the diet containing 40% DDGS, which achieved 61.05 MPa, there were no significant differences among the other diets for compressive modulus; the other values varied between 12.39 and 17.68 MPa. The diet with 40% DDGS achieved the highest level and had the highest standard deviation, which can be one factor for the large variation towards the other blends; another reason could again be attributed to the significantly higher MC, which resulted in a more highly plasticized product.

### Pellet Durability Index

High quality fish feed extrudates should be durable and made to last during transportation, stacking, and feeding. This property of extrudates is a decisive factor for the tumbling economics of aquafeeds. Durability is quantified by the amount of fines returned from a batch under standardized tumbling conditions (Hansen and Storebakken 2007). The values for durability for the blends containing DDGS ranged between 89.89% and 96.77%, and all were significantly higher than the control diet (93.35%) except for the blend containing 30% DDGS (which had a PDI of 89.89%). These levels indicated high durability, and thus very good resistance against destructive forces. In preceding single-screw extrusion studies on yellow perch feeds, likewise, high levels of PDI (up to 96%) were determined when including DDGS in the diets (Ayadi et al. 2009a, b). Overall, no clear pattern between amounts of DDGS included in diets affecting PDI of extrudates were observed.

### Water Stability

Water stability is another measurement for quality of fish feed, and is defined as the amount of time a pellet requires before breaking up after it has been placed in water. WS quantifies the dissolving period and loss of nutrients once they are exposed to water. Generally, long times for WS demonstrate high physical stabilities of extrudates. Besides, the time fish need to consume their rations is decisive for the duration of feed's stability in water.

All times for WS were above 30 min, irrespective of whether samples were rotated or unmoved. These results indicate excellent water stability properties for all extruded diets, and were similar to the values that Ayadi et al. (2009a, b) determined; they observed times for WS between 24 and 30 min for extrudates containing DDGS.

### Angle of Repose

AOR provides information about the storage behavior of extrudates and feed ingredients. It quantifies how easily the material flows and how much storage space is required. Low AOR values are advantageous because they indicate better flowability, improve dispersion, and therefore need less room for storage. Additionally, increased flowability results in better filling properties. Angle of repose varied between 36.28° and 31.78° and decreased with higher DDGS levels. Hence, diets with higher amounts of DDGS showed a better flowability, which is desired. The differences in values for AOR may be attributed to the surface and shape irregularities of the extrudates and occurring friction and other forces.

## Color

Extrudates became browner and darker in color with higher DDGS amounts (Fig. 3). Presently, color of fish feed plays no tangible role in the aquaculture feed sales market. However, it may provide an indication about the loss of lysine in the extruded product, which is an essential amino acid for fishes. The Maillard reaction, favored by high temperatures in combination with low water content, causes reduction of sugars and free amino groups in proteins, and decreases the protein digestibility and availability of amino acids, particularly lysine (Björck et al. 1985). Cromwell et al. (1993) determined that DDGS with lighter color tended to have the highest concentrations of lysine compared to dark colored DDGS.

DDGS is generally golden brown and lighter in color than fish meal, the level of which was reduced from 40% to 0% with increasing amounts of DDGS in the raw blends. These raw blends became lighter and more yellow, red brownish in color with increasing DDGS levels. This was confirmed in changes in the raw materials' color: a significant increase in brightness (Hunter L), redness (Hunter a), and yellowness (Hunter b) of 24.7%, 68.4%, and 46.1%, respectively, was observed with increasing DDGS levels from 0% to 40%. Regarding the visual differences in the final product, the color became darker and more red brownish as DDGS increased. Analyzing color instrumentally, there were no significant differences in brightness among blends containing 0%, 10%, and 20% DDGS; diets with 30% and 40% DDGS had significant lower values in brightness than all blends and were also significantly different to each other. Yellowness of the extrudate samples showed the lowest values for the blends with 0% and 40% DDGS; blends containing 10%, 20%, and 30% DDGS were significantly different from all other blends.

Comparing the color of the raw material with the final extrudates, only changes in redness conformed to the differences of the raw material color. Likewise, redness increased steadily and significantly with rising DDGS levels; all values for redness were lower than the raw material, which thus resulted in greener extrudates. Changes in brightness and yellowness of the extruded products were not in accordance with behavior in color changes of the raw product. Diets with 0% and 10% DDGS of extrudates had higher values for L than the raw ingredients, and resulted therefore in brighter extrudates. Diets containing 20–40% DDGS had lower values for brightness for the extruded product and were therefore darker than the raw materials. All extruded blends had lower values for yellowness and hence, resulted in bluer extrudates. Comparing the changes in brightness among the raw material and the extruded product for the blends without DDGS and with 10% DDGS an increase of 7.66%, and 2.76%, respectively, and for the diets containing 20%, 30%, and 40% DDGS a decrease of 1.57%, 16.92%, and 34.08%, respectively, was observed.

Summing up, it can be assumed that for diets with 20%, 30%, and 40% DDGS, processing conditions related to the Maillard reaction could have caused a darker product. In future studies, the loss of lysine should be evaluated by additional laboratory nutrient analysis.

## Thermal Conductivity and Thermal Diffusivity

Thermal conductivity governs the heat transfer through a material by conduction. The values for all blends varied between 0.06 W/m·°C (for the control diet) and 0.08 W/m·°C (for the blend containing 40% DDGS). A significant increase occurred between the control diet, the diets containing 10–30% DDGS, and the diet with 40% DDGS could be observed with increasing levels of DDGS. The values for all raw materials were low and characterized the blends as materials with poor heat transfer properties.

The thermal diffusivity is defined as the ratio of the thermal conductivity to the volumetric heat capacity. It indicates the heat storage ability of a given sample (Kawasaki and Kawai 2006) and is basically a measure of the heating time in that material, i.e., the lower the thermal diffusivity the longer the heating time (Arámbula-Villa et al. 2007). The measured values for thermal diffusivities for all diets varied between 0.13 and 0.14 $mm^2/s$. This has negative effects on heating and cooling processes, since small variations in temperature require more time to be readjusted. Then again, it can be assumed that the thermal properties of the feedstock did not interfere with the intense mechanical shear that can break covalent bonds in biopolymers during extrusion processing (Asp and Björck 1989); and consequently the thermal properties of the dough may not have affected chemical reactions, residence time, and the viscosity during extrusion cooking.

## Mass Flow Rate and Processing Behavior

The amount of extrudates produced and the performance of the extruder during processing was quantified by the mass flow rate. MFR is influenced by the screw speed, the diameter of the die (Kannadhason et al. 2010), the shear rate, levels of DDGS, moisture content, and viscosity of the dough (Chevanan et al. 2008). The highest MFR was achieved at a level of 1.08 kg/min for the blends with 0% and 10% DDGS. MFR for the following blends slowly decreased with increasing levels of DDGS; the lowest MFR was recorded at 0.82 kg/min for the diet including 40% DDGS. A significant decrease in extruder throughput with increasing DDGS level was observed. These results are contradictory to the conclusions of Chevanan et al. (2008) and Ayadi et al. (2009a, b), who determined an increase in MFR when DDGS levels increased.

The temperature distribution throughout the extrusion process (Fig. 5) for each ingredient blend at the different head zones indicated that there were some differences between the blends. The temperature entering the extruder increased noticeable due to steam injection for adjusting the blends to the desired temperatures; temperatures in the head 2 zone 1 decreased because water was injected to the extruder. For the blends containing 0%, 10%, and 20% DDGS, the temperature zones were maintained at the same temperature combinations, whereas the temperature combination for blends with 30% and 40% DDGS varied for some head zones. Hence, the variations in temperature recordings occurred were based on settings, but can also be attributed to the nutrient composition and the particle size of the varying blends. As observed in Figs. 3 and 4, the different blends altered in texture. Regarding the blend containing 40% DDGS, the temperature setting was the lowest. Hence, higher temperature values than the other blends could be ascribed to the significantly higher recording for thermal conductivity compared to the other blends. Furthermore, temperature fluctuations, particularly in the final zones, can be ascribed to higher friction caused by less available free water due to further processing and cooking.

### Extrudate Nutrient Analysis

The values for the protein content in the extrudates showed high variances. Since the diets were initially designed to have similar protein levels, it can be concluded that discrepancies in the analysis of protein contents of the extrudates were caused by faults in formulations and experimental error. The fat content hardly showed differences among the different blends, whereas fiber increased by 127% and ash contents decreased by 64% with increasing DDGS levels. The increasing fiber values can be related to the higher fiber in DDGS compared to the other ingredients. Similar conclusions for changes in fiber content in DDGS-based fish feed were made by Kannadhason et al. (2010).

### Property Correlations

Pearson linear correlation analysis results can be found in Table 6. Many correlations with *r* values >0.80 and ≤0.8 were detected among the various properties, of which most were anticipated based on previous studies conducted, for example, by Chevanan et al. (2007a) and Kannadhason et al. (2009a). In this study, the majority of DDGS properties showed high correlations, and which have already been discussed. Hunter color values of the raw blends were strongly positively related to all of the raw properties (with *r* values >0.84), except for thermal diffusivity; mass flow rate was strongly, negatively related to Hunter color values of the raw feed blends, with *r* values ≤0.93. These correlations can be ascribed to the DDGS content of the raw blends, which showed similar correlations.

## Conclusions

Extrusion studies were performed using a twin-screw extruder with the intention of producing nutritionally

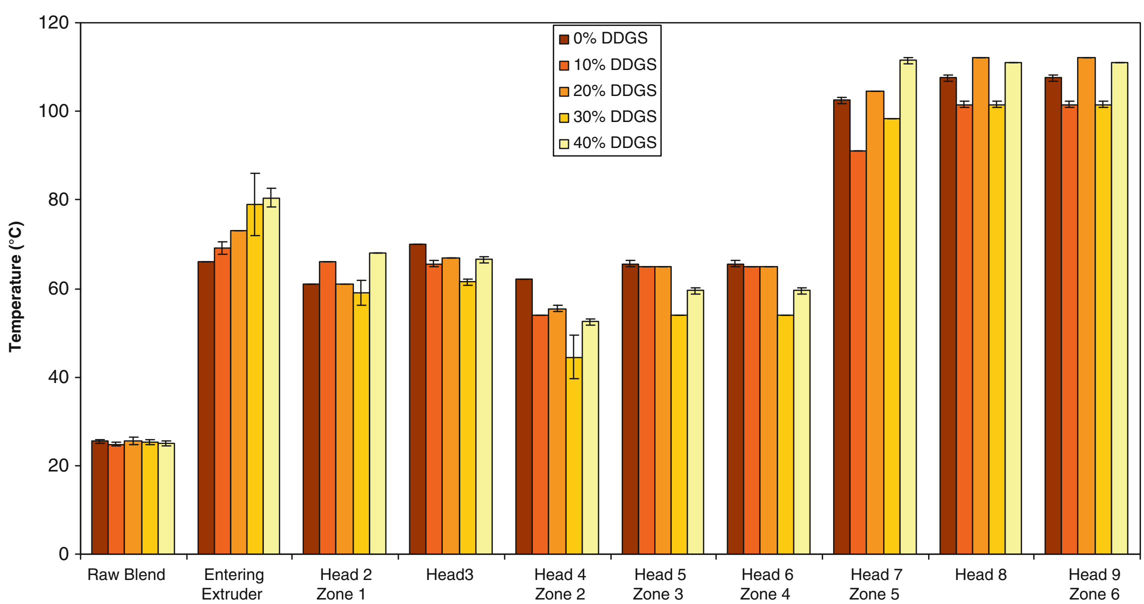



**Fig. 5** Temperature distribution throughout the extrusion process for each ingredient blend. *Error bars* ±1 SD

**Table 6** Linear correlations among all independent and dependent variables

| | | Extrudate properties | | | | | | | | | | | | | Processing properties | Raw properties | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | DDGS | MC | $a_w$ | ER | UD | BD | Compr. strength | Compr. modulus | PDI | AOR | *L* | *a* | *b* | MFR | MC | $a_w$ | BD | *k* | α | *L* | *a* | *b* |
| Extrudate Properties | DDGS | 1 | | | | | | | | | | | | | | | | | | | | | |
| | MC | 0.8351 | 1 | | | | | | | | | | | | | | | | | | | | |
| | $a_w$ | 0.3638 | −0.1240 | 1 | | | | | | | | | | | | | | | | | | | |
| | ER | −0.1755 | −0.0092 | −0.1872 | 1 | | | | | | | | | | | | | | | | | | |
| | UD | 0.6581 | 0.6617 | −0.0482 | −0.7397 | 1 | | | | | | | | | | | | | | | | | |
| | BD | 0.6190 | 0.3899 | 0.2184 | −0.8352 | 0.8983 | 1 | | | | | | | | | | | | | | | | |
| | Compressive strength | 0.8456 | 0.7863 | 0.0151 | −0.5147 | 0.9299 | 0.8639 | 1 | | | | | | | | | | | | | | | |
| | Compressive modulus | 0.7245 | 0.8451 | −0.2756 | −0.4154 | 0.9116 | 0.7287 | 0.9495 | 1 | | | | | | | | | | | | | | |
| | PDI | −0.0288 | −0.0962 | −0.1186 | −0.9344 | 0.6708 | 0.7540 | 0.4392 | 0.4105 | 1 | | | | | | | | | | | | | |
| | AOR | −0.9481 | −0.9548 | −0.1169 | −0.0170 | −0.6047 | −0.4409 | −0.7997 | −0.7722 | 0.1722 | 1 | | | | | | | | | | | | |
| | *L* | −0.8822 | −0.9719 | 0.0685 | −0.0903 | −0.5849 | −0.3806 | −0.7858 | −0.8095 | 0.1831 | 0.9814 | 1 | | | | | | | | | | | |
| | *a* | 0.9564 | 0.9478 | 0.1002 | −0.0333 | 0.6474 | 0.5021 | 0.8428 | 0.8096 | −0.1070 | −0.9961 | −0.9809 | 1 | | | | | | | | | | |
| | *b* | 0.3499 | −0.0809 | 0.9506 | 0.1230 | −0.2510 | −0.0275 | −0.1140 | −0.3733 | −0.4200 | −0.1671 | −0.0021 | 0.1330 | 1 | | | | | | | | | |
| Processing Properties | MFR | −0.9737 | −0.8556 | −0.2159 | 0.0312 | −0.5930 | −0.5419 | −0.8316 | −0.7439 | 0.1109 | 0.9626 | 0.9334 | −0.9754 | −0.2439 | 1 | | | | | | | | |
| Raw Properties | MC | 0.9565 | 0.7886 | 0.5109 | −0.1601 | 0.5726 | 0.5103 | 0.7158 | 0.5826 | −0.1174 | −0.8994 | −0.7989 | 0.8865 | 0.5059 | −0.8812 | 1 | | | | | | | |
| | $a_w$ | 0.9594 | 0.8250 | 0.4137 | −0.3217 | 0.7294 | 0.6471 | 0.8266 | 0.7130 | 0.0707 | −0.9000 | −0.8106 | 0.8990 | 0.3581 | −0.8778 | 0.9783 | 1 | | | | | | |
| | BD | 0.8593 | 0.7529 | 0.0335 | 0.0297 | 0.5281 | 0.5220 | 0.8020 | 0.7372 | −0.0664 | −0.8591 | −0.8743 | 0.8910 | 0.0691 | −0.9460 | 0.6841 | 0.6929 | 1 | | | | | |
| | *k* | 0.8944 | 0.7908 | 0.2712 | −0.5493 | 0.8999 | 0.8185 | 0.9280 | 0.8372 | 0.3543 | −0.8210 | −0.7498 | 0.8401 | 0.1397 | −0.8087 | 0.8713 | 0.9535 | 0.6613 | 1 | | | | |
| | α | −0.7071 | −0.3846 | −0.7717 | 0.5459 | −0.5595 | −0.6365 | −0.5461 | −0.3294 | −0.2222 | 0.5140 | 0.3482 | −0.5098 | −0.6328 | 0.5312 | −0.8216 | −0.8292 | −0.2898 | −0.7906 | 1 | | | |
| | *L* | 0.9621 | 0.8800 | 0.1810 | −0.3410 | 0.8289 | 0.7394 | 0.9514 | 0.8777 | 0.1852 | −0.9295 | −0.8885 | 0.9500 | 0.1151 | −0.9354 | 0.8892 | 0.9472 | 0.8402 | 0.9588 | −0.6599 | 1 | | |
| | *a* | 0.9930 | 0.8716 | 0.2758 | −0.0925 | 0.6318 | 0.5668 | 0.8407 | 0.7457 | −0.0846 | −0.9725 | −0.9265 | 0.9802 | 0.2879 | −0.9916 | 0.9317 | 0.9315 | 0.8983 | 0.8612 | −0.6212 | 0.9572 | 1 | |
| | *b* | 0.9961 | 0.8659 | 0.2856 | −0.1958 | 0.7029 | 0.6426 | 0.8823 | 0.7815 | 0.0136 | −0.9589 | −0.9064 | 0.9710 | 0.2655 | −0.9786 | 0.9363 | 0.9538 | 0.8774 | 0.9084 | −0.6694 | 0.9797 | 0.9944 | 1 |

*MC* moisture content, $a_w$ water activity, *ER* expansion ratio, *UD* unit density, *BD* bulk density, *PDI* pellet durability index, *AOR* angle of repose, *L, a, b* Hunter color parameters, *MFR* mass flow rate, *k* thermal conductivity, α is thermal diffusivity balanced diets for juvenile yellow perch using increasing levels of DDGS. The initial feed MC (5–7%) was modulated with varying amounts of steam (6.9–9.7 kg/h) and water (6.7–13.1 kg/h) during processing to obtain extrudates with adequate cohesive properties. With increasing DDGS levels, significant differences were detected for color on the raw and extruded material. The changes in extrudate color can be ascribed to the processing conditions and to the composition of the raw materials. Bulk density for the raw materials and the extrudates, respectively, showed significant increases with higher amounts of DDGS. Water activities were very low and depict a product with long shelf life properties. Furthermore, values for the compressive strength were very low and resulted in easily breakable extrudates, unlike the pellet durability index, which indicated high values for nearly all blends and provided a durable product with high resistance against destructive forces. Low compressive strength and high pellet durability are thus easily consumable by fish and have very good transport properties. In addition, extrudates showed excellent water stabilities that did not dissolve for at least half an hour when exposed to water. In summary, it can be concluded that extrusion processing with DDGS yielded viable extrudates. Future work should examine how changes in color, particularly brightness, are related to the loss of lysine affected by the extrusion process and availability of lysine and other essential amino acids in DDGS feed can be assimilated by fish.

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