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Production performance and non-specific immunity of cage-raised red drum, *Sciaenops ocellatus*, fed soybean-based diets

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ABSTRACT

A 15-week feeding trial was conducted to evaluate the replacement of fishmeal (FM) with soybean products (SP) in the diet of advanced juvenile red drum, *Sciaenops ocellatus*. Two dehulled, hexane-extracted SP were evaluated: commodity soybean meal [SBM; 48% crude protein (CP)] and Navita 3010 [N-3010; produced from non-genetically modified, selectively bred varieties; 54% CP]. The additive effect of a prebiotic (GroBiotic®-A; GBA) on the overall performance of red drum also was evaluated. All diets were formulated to contain 35% digestible protein (DP), 15% lipid, and an estimated 3.5 kcal DE kg⁻¹. The reference diet was formulated to contain 44.6% Special Select® menhaden FM and 15.0% corn protein concentrate (CPC). Three test diets (SBM, SBM + GBA, and N-3010) were formulated to contain 6.0% FM and 15.0% CPC, together providing approximately 38% of the dietary DP. The remaining 62% DP was provided by 5.7–5.8% soy protein concentrate (SPC) and the specific SP treatment. Diets were fed to apparent satiation to advanced juvenile red drum (mean \pm SD = 68 \pm 5.1 g initial weight) in each of four replicate 1-m³ floating-cages. After 15 weeks of feeding, no significant differences were found among treatments for all growth performance and non-specific-immunity parameters evaluated. Based on these results, SBM or N-3010 in combination with SPC can replace approximately 86% of the DP provided by FM in the diet without negatively affecting production performance and health status of advanced juvenile red drum.

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1. Introduction

The aquaculture industry has long relied on fishmeal (FM) as the primary protein source in aquafeeds for carnivorous fish species (Tacon and Metian, 2008), but limited supply and increasing prices have prompted the utilization of alternative protein feedstuffs. Soybean meal (SBM) is currently the foremost feedstuff substituting for FM (Tacon et al., 2011; Troell et al., 2014). However, different degrees of tolerance to dietary SBM have been reported in a range of aquacultured fish species (Francis et al., 2001), largely attributed to the presence of anti-nutritional factors (ANFs).

Soy-protein concentrate (SPC) is an example of an highly refined soybean product (SP) with relatively higher protein content (NRC, 2011) and lower levels of ANFs compared to commodity SBM (Barrows et al., 2008); however, processing costs may limit its inclusion in aquafeeds (Gatlin et al., 2007). On the other hand, the selective breeding of nongenetically modified (non-GM) soybean varieties has led to the introduction of novel SP to the feed industry (Schillinger et al., 2012). Because they contain higher protein content and lower levels of ANFs, such novel ingredients may be more cost-effective for use in plant-based diets for aquacultured species.

* Corresponding author. *E-mail address:* d-gatlin@tamu.edu (D.M. Gatlin). Red drum, *Sciaenops ocellatus*, is a carnivorous and highly prized marine teleost to which plant-based diets have been evaluated (McGoogan and Gatlin, 1997; Moxley et al., 2014; Reigh et al., 1992; Rossi et al., 2013). These studies have shown that the red drum is able to tolerate relatively high dietary levels of plant feedstuffs without showing detrimental effects on production performance. However, these studies were restricted to early stage juveniles. Considering that the response to plant-based diets must be evaluated in larger fish.

With the increased utilization of alternative feedstuffs, the application of various feed additives in aquaculture also has increased (NRC, 2011). Particularly in the case of prebiotics, increasing evidence points out their beneficial effects in the diet of aquacultured species, as reviewed in Ringø et al. (2014). Responses of red drum to prebiotic supplementation have included increased apparent nutrient digestibility of SBM-based diets (Burr et al., 2008a), modulation of gut microbiota community (Burr et al., 2008b), and enhanced weight gain, feed efficiency, non-specific immunity, and gut morphology (Buentello et al., 2010). Enhanced gut morphology resulting from prebiotic supplementation in red drum also was evidenced in a latter study (Anguiano et al., 2013).

The objective of this study was to evaluate the performance and health status of advanced juvenile red drum in response to the effects of: i) a FM- or SP-based diets; ii) a SBM- or a novel SP-based diet; and iii) a yeast-based prebiotic supplement in a SBM-based diet. The study







was conducted in cages suspended in aquaculture ponds to allow the use of larger fish than used in previous studies and incorporate the variable conditions of the pond environment.

2. Material and methods

2.1. Diets

A 15-week feeding trial was conducted to evaluate the replacement of FM with SP in the diet of red drum. The SP evaluated included SBM, and Navita Premium Feed Ingredients 3010 (designated as N-3010), a SP produced from non-GM, selectively bred soybean varieties. All diets were formulated to contain 35% digestible protein (DP), 12% lipid, and an estimated digestible energy (DE) level of 350 kcal/100 g (Table 1). The reference diet (designated REF) was formulated such that 71% of the DP was provided from FM, and the remaining DP (29%) was provided from corn protein concentrate (CPC). Two test diets (designated

Table 1
Composition of experimental diets fed to advanced juvenile red drum for 15 weeks.

Ingredients	REF	SBM	SBM + GBA	N-3010	
	% of dry matter				
Menhaden fishmeal ^a	44.6	6.0	6.0	6.0	
Corn protein concentrate ^b	15.0	15.0	15.0	15.0	
Soy protein concentrate ^c	-	5.7	5.8	5.8	
Soybean meal ^d	-	40.0	40.0	-	
Navita 3010 ^e	-	-	-	35.6	
Wheat flour ^f	20.5	7.5	5.5	11.8	
Menhaden oil ^g	9.8	13.3	13.3	13.6	
Vitamin premix ^{h,i}	3.0	3.0	3.0	3.0	
Mineral premix ^{h,i}	4.0	4.0	4.0	4.0	
Dicalcium phosphate ^j	-	1.0	1.0	1.0	
GroBiotic®-A ^k	-	-	2.0	-	
Glycine ^l	2.0	2.0	2.0	2.0	
Taurine ¹	1.0	1.5	1.5	1.5	
Lysine HCl ¹	-	0.5	0.5	0.2	
DL-Methionine ¹	-	0.5	0.5	0.5	
Analyzed proximate composition					
Moisture	2.4	2.7	3.1	3.3	
Protein	46.6	47.6	46.9	46.9	
Lipid	14.7	12.5	12.8	11.6	
Ash	11.8	6.5	6.5	6.2	
Total phosphorus	1.9	1.0	1.0	1.0	
Crude fiber	2.9	2.1	2.0	1.6	
Analyzed amino acid composition					
Arg	2.3	3.0	2.4	3.0	
His	0.9	1.0	1.0	1.1	
Ile	1.9	2.3	1.9	2.2	
Leu	4.3	5.2	4.3	5.0	
Lys	2.4	2.7	2.2	2.4	
Met	1.1	1.2	0.9	1.3	
Phe	2.2	2.9	2.4	2.9	
Tau	1.1	1.7	1.5	1.7	
Thr	1.7	1.9	1.5	1.8	
Val	2.2	2.6	2.1	2.4	

^a Special Select [™] Omega Protein Inc., Abbeville, LA, USA. Crude protein = 68.4%; Lipid = 10.2% on a dry-matter basis.

 $^{\rm b}\,$ Empyreal 75, Cargill Corn Milling (Blair, NE, USA). Crude protein = 81.3%; Lipid = 4.2% on a dry-matter basis.

 $^{\rm c}\,$ The Solae Company, St. Louis, MO, USA. Crude protein = 71.9%; Lipid = 0.44% on a dry-matter basis.

^d Producers Coop. Association, Bryan, TX, USA. Crude protein = 53.9%; Lipid = 10% on a dry-matter basis.

 $^{\rm e}$ Navita Premium Feed Ingredients, West Des Moines, IA, USA. Non-GM. Crude protein = 60.6%, crude lipid = 0.44%, on a dry-matter basis.

^f Manildra Milling. Crude Protein = 18.4%; Lipid = 1.6% on a dry-matter basis.

^g Omega Protein, Reedville, VA, USA.

^h Moon and Gatlin (1991).

ⁱ MP Biomedicals, Solon, OH, USA.

- ^j Fisher Scientific, Pittsburg, PA, USA.
- ^k International Ingredient Corporation, St. Louis, MO, USA.

¹ USB, Cleveland, OH, USA.

SBM and N-3010) were formulated to replace approximately 72% of the DP from FM in the REF diet with SBM or N-3010. An additional inclusion of SPC at 5.7% into each of the test diets resulted in a total of 86% replacement of DP from FM compared to the REF diet. Additionally, in order to evaluate the potential additive effect of a yeast-based prebiotic (GroBiotic®-A; GBA) on the overall performance of red drum, a third test diet (designated SBM + GBA) was formulated identically to the SBM diet with the addition of GBA at 2.0%, at the expense of the same amount of wheat flour. All diets were supplemented with mineral and vitamin premixes to meet or exceed the established requirements for red drum (NRC, 2011), along with the supplementation of taurine (1.0 or 1.5%). Supplementation of DL-methionine and L-lysine was provided to the test diets in excess of the established requirements for red drum, while glycine was supplemented at 2.0% for palatability enhancement (McGoogan and Gatlin, 1997).

All diets were manufactured at the U.S. Fish and Wildlife Service, Bozeman Fish Technology Center-Fish Feed Laboratory (Bozeman, MT). The manufacturing procedures, including ingredient grinding, mixing, and extruding, were performed as described by Read et al. (2014). All proximate composition analyses were performed in triplicate using AOAC (1990) procedures. The analyzed composition of the diets fed to advanced red drum juvenile closely matched the targeted formulation values for dietary CP of approximately 45% (35% DP), while lipid was slightly lower (11.6-12.8%) in the soybean-based diets (Table 1). The dietary phosphorus ranged from 1.0% in the SBM, SBM + GBA, and N-3010 diets to 1.9% in the REF diet and was in excess of the 0.8% established as the requirement for red drum (Davis and Robinson, 1987). The dietary concentration of lysine, methionine, and threonine, also was in excess of the established requirements for red drum (Boren and Gatlin, 1995; Brown et al., 1988; Moon and Gatlin, 1991).

2.2. Fish and feeding trial

Red drum were obtained from the Sea Center Texas Marine Aquarium, Fish Hatchery and Nature Center operated by Texas Parks and Wildlife Department in Lake Jackson, TX, and transported by truck to the Texas A&M Aquacultural Research and Teaching Facility. In an indoor setting (water temperature = ~ 26 °C; dissolved oxygen (DO) = near air saturation; pH = between 7 and 8; salinity = ~ 6 g/L, and 12 h light:12 h dark photoperiod), the fish were cultured in 6, 1200-L circular, fiberglass tanks operating as a recirculating system until over 60 g of mean weight was attained. During this period, red drum were fed at a rate approaching apparent satiation with a 40% crude protein and 12% crude fat commercial diet (Rangen, Inc., Angleton, TX).

At the commencement of the feeding trial, a sample of 15 fish was collected from the population and frozen (-20 °C) for subsequent analyses of initial whole-body composition. Thirty red drum juveniles were then stocked into each of 16, 1-m³ floating-cages divided equally between two, 0.05-ha, rubber-lined ponds. Constant mechanical aeration was provided to each pond and well water was added as necessary to cope with evaporation. In each cage, red drum were fed their assigned diet for a few days until active feeding was observed, then fish in each cage were group weighed for the commencement of the feeding trial. Fish [mean initial weight \pm standard deviation (SD) = 68 \pm 5.1 g] in each cage were fed their assigned diet once daily to apparent satiation during the entire feeding trial, which began on June 14, and ended on September 27, 2013 (15 weeks). Two cages in each pond were assigned either the REF or a test diet, resulting in a generalized randomized block (GRB) design with four replicate cages per dietary treatment.

Average morning temperature was 28.0 \pm 1.1 (mean \pm SD) and morning DO was 7.1 \pm 0.4, with total ammonia-nitrogen averaging 0.4 \pm 0.1 mg L⁻¹. Water salinity was increased by the addition of sea salt (Fritz Industries, Dallas, TX, USA) and food-grade NaCl (Producers Coop. Association, Bryan, TX, USA) at a 1:3 ratio to the well water and was maintained at 5.8 \pm 0.26 g L⁻¹.

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