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Reducing digestible protein in pond production of hybrid striped bass

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Study shows significant reduction is possible in commercial diets



This study showed that a significant reduction is possible in the digestible protein level in commercial diets for this hybrid striped bass using ideal protein diet formulation, a vigorous set of ingredient nutrient availabilities and a higher level (45 percent) of muscle profile as the formulation target.

There is growing interest to reexamine hybrid striped bass (HSB) feed formulas and nutrient requirements to maximize production efficiency, minimize the impacts of excreted dietary nutrients on pond water quality and support the industry against climate change.

Production ponds for sunshine hybrid striped bass (*Morone chrysops* × *M. saxatilis*) can reach extreme temperatures (29 to 33 degrees-C) during the summer, and due to peak feeding during this season, high total ammonia-nitrogen (TAN) and concurrently lower dissolved oxygen typically decrease feed consumption intermittently and increase the potential for fish stress, disease and mortality.

Reducing overall protein content of HSB feed can potentially decrease production costs and feed nutrient waste, and improve industry competitiveness if fish efficiencies and performance are not degraded. Previous research has shown that dietary protein can be reduced in fish diets by sensible supplementation of multiple amino acids.

In a previous, controlled tank study we determined that a 40 percent digestible protein (DP)/18 percent lipid diet formulated on an ideal protein basis would be the optimum among the six diets evaluated for summer HSB production up to 400-gram final weight. Results also suggested that, to reduce pond ammonia during high incidences of TAN that are typical during summer HSB pond production with the least compromise to production efficiency, an HSB producer farmer should feed the 40/18 diet at a reduced level instead of converting to a lower protein diet.

This article summarizes the results [original publication: *Aquaculture* 490 (2018) 217–227] of a study designed to extend the submarket-sized fish performance and water quality results of our previous tank study to market-sized HSB in pond production at commercial rearing densities, and concurrently explore

the hypothesis that intact protein can be further lowered in production diets by supplementing the first three limiting amino acids (Lys, Met, Thr).

Experimental setup

The feeding trial was carried out in twelve 0.1-hectare ponds at the Harry K. Dupree Stuttgart National Aquaculture Research Center (HKDSNARC). Each pond was stocked with 746 juvenile HSB (initial weight of 121.4 ± 3.21 grams) from a commercial supplier and cultured at the HKDSNARC over a 167-day period incorporating spring, summer, and fall temperatures of central Arkansas.

The fish were initially all fed a commercial hybrid striped bass diet for 26 days and then ponds were randomly assigned to one of three test diets in a completely randomized design (four ponds/diet) and hand fed once daily to apparent satiation not exceeding 100 kg of feed/ha for 141 days.

The three commercial test diets were formulated to contain one of three intact digestible protein (DP) levels (35, 38, 41 percent) and were supplemented with the first three limiting amino acids (Met, Lys, Thr) at an ideal protein level of 45 percent HSB muscle. Protein in the diets was supplied by a combination of commercial menhaden fish meal (MFM), soybean meal (SBM), pet food-grade poultry byproduct meal (PBM), poultry bloodmeal (BM), and poultry feather meal (FM), with a minor contribution from wheat flour.

To limit response variability from ingredient effects, ratios of digestible protein from the various ingredients were held as constant as possible among formulas, as follows: animal: plant protein (2:1), MFM: SBM (1:1), MFM: PBM (1:1), BM: FM (1:1), and (MFM+ SBM+PBM): (BM+FM) (6.98:1). In practice, however, actual ratios varied slightly; Similarly, test diets were formulated to contain 18 percent lipid supplied by a constant ratio (1:1.25) of fish lipid (8 percent) to poultry lipid (10 percent) from all ingredients.

However, total lipid measured in the test diets fell short of the target by 3 percentage points. Test diets were manufactured with commercial methods using a twin-screw cooking extruder to produce 3.5-mm floating pellets.

Water was added as needed to replace losses to evaporation and seepage. Ponds were fertilized after filling and then as needed through July with chemical and organic fertilizer to promote and maintain a phytoplankton bloom. Salt (2,241 kg/ha) was added to all culture units to ensure chloride concentration exceeded 100 mg/L.

Each pond was equipped with an electric paddlewheel aerator (11.1 kW/ha) that was operated nightly. Dissolved oxygen and temperature in each pond were monitored continuously, and water samples were collected weekly from each pond to determine water quality parameters and assure adequate conditions for the fish.

Animal care and experimental protocols used in this work were approved by the HKDSNARC Institutional Animal Care and Use Committee and conformed to USDA/ARS Policies and Procedures 130.4 and 635.1.

A number of fish were randomly collected at the beginning and at the end of the study for analysis of whole body composition and for determination of condition indices that included hepatosomatic index (HSI), intraperitoneal fat (IPF) ratio and muscle ratio (MR). Proximate composition of diets and fish was determined according to standard methods, and protein, energy and amino acid retention efficiencies (RE) were estimated.

For more detailed procedures on the study setup – including experimental design and diets; fish, feeding, and pond management; fish and tissue sampling; diet and tissue chemical analyses; and statistical analyses – refer to the original publication.



Results of this study regarding fish yields, average weights at harvest, FCR, protein retention and survival were markedly better when compared to several published hybrid striped bass pond studies.

Results and discussion

Total fish biomass per pond (520 to 540 kg), gross fish yield (5,139 to 5,342 kg/ha), survival (95.1 to 96.7 percent), FCR (1.34 to 1.37), weight gain (480 to 498 percent), average fish weight at harvest (732 to 734 grams), and average maximum fish weight at harvest (1,030 to 1,122 grams) were not correlated to diet DP level (Table 1). The average minimum fish weight at harvest was numerically larger (534 grams) in ponds fed the 38 percent DP diet than in ponds fed the 35 percent (477 grams) or 41 percent (448 grams) DP diets though not statistically different.

Rawles, digestible protein, Table 1

Response ^b	Intact DP (%) 35	Intact DP (%) 38	Intact DP (%) 41	ANOVA Pr > F ^c	LINEAR Contrast ^d
Total fish weight	520.0 ± 14.3	540.0 ± 16.5	533.3 ± 14.3	0.528	0.948
Gross Fish Yield	5139 ± 104 (4587 ± 140)	5342 ± 189 (4768 ± 169)	5271 ± 191 (4705 ± 170)	0.528	0.431
Survival	95.1 ± 1.2	96.7 ± 1.4	96.7 ± 1.2	0.302	0.17
Total feed	585 ± 11	616 ± 18	616 ± 9.0	0.536	0.079

FCR	1.34 ± 0.01	1.36 ± 0.03	1.37 ± 0.01	0.849	0.231
Gain	480.2 ± 7.4	490.0 ± 24.2	498.5 ± 14.3	0.681	0.37
Avg wt	736 ± 19	764 ± 22	732 ± 19	0.588	0.892
Max wt	1122 ± 46	1112 ± 54	1030 ± 46	0.338	0.152
Min wt	477 ± 24	534 ± 28	448 ± 24	0.071	0.964
Percent fish > 680g	59 ± 5.8	71 ± 6.7	59 ± 5.8	0.352	0.959
CV	0.205 ± 0.008	0.176 ± 0.009	0.200 ± 0.008	0.086	0.889
Skewness	0.52 ± 0.15	0.62 ± 0.17	0.25 ± 0.15	0.261	0.489
Kurtosis	-0.03 ± 0.26 ^a	0.31 ± 0.30 ^a	-0.89 ± 0.26 ^b	0.053	0.502
HSI	3.17 ± 0.15	2.81 ± 0.17	2.60 ± 0.15	0.08	< 0.001
IPF ratio	8.60 ± 0.18	8.64 ± 0.21	8.21 ± 0.18	0.213	0.121
MR	50.7 ± 0.57	50.5 ± 0.66	50.1 ± 0.57	0.782	0.221

Table 1. Growth, feed performance, size distribution parameters, and composition indices of hybrid striped bass (initial weight: 121.4 ± 0.8 grams/fish; mean ± SE) reared in ponds to market size on extruded diets containing one of three intact digestible protein (DP) levels (35, 38, 41 percent) and supplemented with Met, Lys, and Thr at an ideal protein level of 45 percent hybrid striped bass muscle^a.

^a Values are least squares (LS) means of N = 4 replicate ponds of fish for diets 35 percent and 41 percent intact DP and 3 replicate ponds for 38 percent intact DP; least squares means in the same row with different letters are different (P < 0.10).

^b Total pond weight (kg/pond) after 167 days; Gross Fish Yield, kg/ha (lbs/acre); Survival (percent); Total feed (Kg, dry weight basis) consumed; FCR: feed conversion ratio = g dry feed consumed / g weight gained; Gain (percent) = (final weight - initial weight) * 100 / initial weight; Avg wt: average fish weight (g) at harvest; Max wt: maximum fish weight (g) at harvest; Min wt: minimum fish weight (g) at harvest; percent > 680 g: percent of fish weighing > 680 g (1.5 lbs) at harvest; CV: coefficient of variation in fish size distributions; Skewness - denotes whether the fish size distribution is weighted toward smaller fish with fewer larger fish, i.e., right-tailed (+), heavier toward larger fish with fewer smaller fish, i.e., left-tailed (-), or symmetric (0) about the mean; HSI: hepatosomatic index (percent) = liver mass * 100 / fish mass; IPF: intraperitoneal fat (percent) = intraperitoneal fat mass * 100 / fish mass; MR: muscle ratio (percent) = skinless fillet with rib mass * 100 / fish mass.

^c ANOVA, Pr > F. LS means in the same row with different letters are different (P ≤ 0.05).

^d Linear contrast, Pr > F. Linear effect of DP deemed significant at P ≤ 0.05.

The percent of harvested fish over 680 grams did not differ statistically but was 59 percent in ponds fed the 35 percent or 41 percent DP diets, as opposed to 71 percent in ponds fed the 38 percent DP diet. Maximum daily ration did not differ significantly among treatments and averaged 99, 95, and 90 kg/ha for the 35 percent, 38 percent, and 41 percent DP diets, respectively. The daily feed ration was never restricted due to water quality, and total feed fed showed a slight linear increase with diet DP level.

Fish size at harvest was slightly more variable (higher CV) in ponds fed the 35 percent DP diet. The distribution of market size fish fed the 41 percent DP diet was slightly more flattened about the mean compared to distributions of market size fish fed the 35 to 38 percent DP diets; otherwise, size distributions did not appear significantly skewed toward larger or smaller size classes with respect to diet DP. Liver size (HSI) decreased linearly, from 3.17 percent to 2.60 percent, with increasing diet DP. Body fat content and muscle ratios were not significantly influenced by diet DP level (Table 1).

Gross fish yields (5139–5342 kg/ha or 4587–4768 lb/acre), average weights at harvest (732–764 g), FCR (1.34–1.37), protein retention (PRE; 22–25 percent), and survival (95 percent–97 percent) were markedly better in this study than in several published hybrid striped bass (HSB) pond studies, but similar to performance in our previous pond trial investigating the graded replacement of fish meal with pet-food grade poultry byproduct meal (PBM) on an ideal protein (IP) basis. In our previous study, yields ranged from 5,802 to 6,697 kg/ha, average weights at harvest were 814 to 932 grams, FCRs ranged from 1.99 to 2.15, PRE was 19 to 23 percent and survival ranged 95 to 99 percent.

The lack of significant growth and feed performance differences, or whole-body composition differences, compared to the results obtained in Rawles et al. (2012), suggest that digestible protein in HSB diets can be lowered significantly (100 grams/kg) through multiple amino acid supplementation. Furthermore, whole body protein retentions and amino acid retention efficiencies increased linearly by 3 percentage points (12 percent increase between 36 DP and 41 DP treatments) with decreasing intact DP.

Results for final whole-body protein (16 percent), lipid (17 percent), energy (8.3 to 8.5 J/kg), moisture (62 to 63 percent) and amino acid content did not differ significantly among dietary treatments on a fresh weight basis retention efficiencies of protein (PRE), Asx, Glx, Leu, Lys, Ser, and Val decreased linearly with increasing diet DP. Retention efficiencies of Arg, His, and Ile also declined numerically with increasing DP but the trend was not strongly linear. However, the range in retention efficiencies among dietary treatments was less than two percentage points in all nutrients. Retention efficiencies of energy (ERE), Ala, Gly, Met, Phe, Thr, and Tyr did not differ significantly with respect to diet DP level.

During “summer” (June 11 to Sept. 14) the number of times weekly TAN exceeded 1.5 mg/L exhibited a positive linear trend ($P=0.086$) with increased digestible protein. But the number of times weekly TAN exceeded 2.0 mg/L did not differ among treatments ($P=0.203$). Weekly TAN exceeded both threshold concentrations most frequently during June and the frequency of occurrence decreased during each subsequent month. The change in the quantity of feed fed on the day of the TAN spike, the previous day or the following 1 to 3 days was not correlated significantly with the TAN spike concentration.

Chlorophyll *a* (Chl *a*) concentrations exceeded 100 mg/m³ during 15.4 percent, 20.0 percent, and 26.7 percent of the TAN>1.5 mg/L events for the 35 DP, 38 DP, and 41 DP diets, respectively. Otherwise, chlorophyll *a* concentration was low and averaged 35.3, 47.5 and 42.4 mg per cubic meter, respectively. Total ammonia-nitrogen and Chl *a* concentrations were correlated negatively for all diets. Total ammonia-nitrogen and soluble reactive phosphorus (SRP) concentrations were positively correlated. Soluble reactive phosphorus and Chl *a* were correlated positively for the 35 DP and 41 DP diets, but not the 38 DP diet. Feed input and SRP concentrations were correlated positively for all diets. Feed input and Chl *a* concentrations were correlated positively for all diets. No other correlations were detected.

Diet lipid was lower (15 percent) than targeted (18 percent) in the test diets and raises the question of whether the lower lipid level affected protein retention efficiencies to some extent.

Therefore, one might hypothesize that higher dietary fat might have potentially spared more protein for deposition in the current study, since this study included the hottest months of the growing season. With the caveat that our previous results are for tank-reared fish up to 400 grams while current results are for pond-reared, market-sized fish (>500 grams), there is some indication that dietary fat at the targeted level might have potentially improved protein retention and reduced lipid deposition, since body fat (IPF) ranged from 6.4 to 7.5 percent in our previous study and 8.2 to 8.6 percent in the current study.

Food conversions in our pond study were markedly better (≈ 1.35) than pond studies currently in the literature and better than current commercial experience. This is a result of several efficiencies in our research setting that are not necessarily feasible in a commercial setting.

Although fish were fed similar total quantities of feed and FCR's suggest minimal feed wastage, the total nitrogen delivered differed by dietary treatment. Since protein retention decreased linearly with increasing diet DP, we might have expected more correlations between pond TAN and dietary treatment.

Perspectives

While post-prandial TAN excretion by hybrid striped bass is known to increase with increasing dietary DP content, pond ecosystem services (phytoplankton and microbial uptake, and soil adsorption), standardized nightly aeration, and setting a maximum daily feed ration in the current experiment did not allow stark treatment differences in water quality to manifest themselves. Results likely would be different if daily feed ration was to exceed quantities fed in the present experiment and nightly aeration was provided on an as needed basis; additional research could verify this.

In addition to supplementing the first three limiting amino acids in the commercial formulas tested, we paid attention to the balance of multiple ingredient/nutrient inputs in the test diets to reduce both confounding ingredient derived influences and potential deficiencies in other nutrients. Specifically, attention was given to available phosphorus, macro minerals and vitamins that are now considered important to overall diet balance.

From the results of our study, it appears that significant reductions can be made in digestible protein level in hybrid striped bass commercial diets using ideal protein diet formulation, a robust set of ingredient nutrient availabilities, and a higher level (45 percent) of muscle profile as the formulation target.

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