NUTRITION/DIETS[1]

Project ***Termination Report*** for the Period
September 1, 2007 to August 31, 2009

NCRAC FUNDING: $80,000 (September 1, 2007 to August 31, 2009)

PARTICIPANTS:

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REASON FOR TERMINATION

The objectives for this project were completed.

PROJECT OBJECTIVES

(1) Develop a least-cost diet for bluegill *Lepomis macrochirus* by:
- (a) Evaluating amino acid availability of dietary ingredients for bluegills,
- (b) Evaluating amino acid composition of bluegills,
- (c) Evaluating limiting amino acid requirements of bluegills, and
- (d) Making a least-cost diet formulation model available to the industry within a two-year period.

PRINCIPAL ACCOMPLISHMENTS

Objective 1a

Apparent digestibility of dry matter and energy, and availability of amino acids from blood meal (BM), fish meal (FM), porcine meat and bone meal (MBM), poultry byproduct meal (PBM), soybean meal (SBM), corn gluten meal (CGM), and corn, wheat, and yellow grease (YG) were determined for juvenile bluegill *Lepomis macrochirus* (mean weight, ~57 g [2.01 oz]). Apparent dry matter digestibility values ranged from 50% (corn) to 87% (BM). Apparent energy digestibility values ranged from 53% (corn) to 92% (BM) for bluegill. Apparent digestibility of most amino acids exceeded 90% for evaluated protein sources, except for MBM which showed slightly lower values (80–90%). Isoleucine digestibility from BM was relatively low (82%). High digestibility values for SBM, PBM, and BM indicate good potential for replacing FM in the diets of bluegill.

Objective 1b

Four randomly selected wild-caught juvenile bluegills (31.2 ± 16.4 g [1.10 ± 0.58 oz], mean weight ± SD) were sampled to determine amino acid compositions of whole-body tissue. Amino acid ratio for 10 essential amino acids were found to be 2.53 (arginine), 0.88 (histidine), 2.08 (isoleucine), 3.25 (leucine), 3.19 (lysine), 1.30 (methionine), 2.10 (phenylalanine),

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[1]NCRAC has funded two Nutrition/Diets projects. The Termination Report for the first project is contained in the 1997-98 Annual Progress Report. This Termination Report is for the second Nutrition/Diets project which was chaired by Robert S. Hayward. It was a 2-year project that began September 1, 2007.

1.85 (threonine), 0.52 (tryptophan), 2.51 (valine). Except for leucine, contents of all other amino acids were lower than lysine. This ratio was used for determining essential amino acid requirements (EAAs).

Objective 1c

Two, 60-day experiments were conducted to determine: (1) lysine requirement of juvenile bluegill based on the dose-response method, (2) requirements for other EAAs using whole-body amino acid profile, and (3) whether differences in growth rates of group-versus individually-housed bluegills led to different lysine requirement levels due to the presence and absence, respectively, of bluegill social hierarchies. Seven experimental diets (isonitrogenous, isocaloric) were prepared to contain graded levels of digestible lysine (1.0–3.1 %). Experiment-1 involved group-housed bluegills (~27 g [0.95 oz], $N = 10$ fish/chamber, 4 chambers/diet) whereas experiment-2 involved individually-housed bluegills (~30 g [1.05 oz], $N = 1$ fish/chamber, 14 chambers/diet). Fish were fed twice daily to apparent satiation. Bluegill growth responses in both experiments generally improved ($P < 0.05$, ANOVA) with increasing dietary lysine levels from 1.0 to 1.6 %, and then leveled off with further increase in lysine level ($P > 0.05$). Optimal dietary lysine level (digestible basis) was calculated to be 1.5% based on broken-line regression analyses of specific growth rate and feed conversion ratio (FCR) with no differences observed between the two rearing methods. Determined dietary requirement levels for other EAAs ranged from 0.24% (trypophan) to 1.53% (leucine).

Objective 1d

Two, 60-day experiments were conducted to determine optimal levels of protein and energy in the diets of juvenile bluegill. In experiment-1, eight experimental diets were formulated to contain digestible protein levels ranging from 35–49% by 2% increments. Dietary energy level was maintained at 3,800 kcal/kg (digestible basis) across diets. Four bluegill groups (~20 g [0.71 oz], $N = 10$ fish/group) were fed the experimental diets twice daily to apparent satiation. Feed consumption, specific growth rate (SGR), and protein efficiency ratio (PER) were significantly affected by dietary protein levels ($P < 0.05$, ANOVA). Fish fed the diets containing 49% protein exhibited higher feed consumption and SGR than those fed the diets containing 35% and 37% protein. However, fish fed the diets containing 35% and 39% dietary protein levels showed better PER than those fed the diets containing 47% and 49% dietary protein levels ($P < 0.05$, ANOVA). No differences were observed among dietary groups for feed conversion ratio, hepato-somatic index (HSI), viscera-somatic index, or whole-body composition ($P \geq 0.05$, ANOVA). Optimal dietary protein level (digestible basis) was calculated to be 41.3% based on broken-line regression analysis of SGR.

In experiment-2, seven experimental diets were prepared to contain dietary energy levels (digestible basis) ranging from 3,000 kcal/kg–4,200 kcal/kg with digestible protein levels fixed at 41% across diets. Four bluegill groups (~21 g [0.74 oz], $N = 10$ fish/group) were fed experimental diets twice daily to apparent satiation for 60 days. No significant differences were detected among fish groups for feed consumption, SGR, or FCR ($P > 0.05$, ANOVA). However, HSI values and whole body fat content were significantly affected by dietary energy levels with fish fed diets containing <3,600 kcal/kg showing significantly less fat deposition than those fed diets containing ≥4,000 kcal/kg. Diets containing 3,500 kcal/kg produced the maximum SGR. Results of both experiments indicated that bluegills require 41% digestible energy and 3,500 kcal/kg digestible energy in diets for optimal growth performance.

Least-Cost Diet Formulation for Juvenile Bluegill

A 60-day study was conducted to determine the least-cost diet formulation for bluegill based on the ingredients tested for digestibility and nutrient requirement levels from previous experiments. Current market price of the ingredients and the digestible nutrient levels determined for each of the tested ingredients were provided in the software program WUFFDA (Windows-based User Friendly Feed Formulation). A total of seven experimental diets were prepared for evaluation. Diets 1 through 6 were computer-formulated by gradually replacing fish meal with a protein blend; ingredients for the protein blend were determined by the software for the cheapest possible formulation while satisfying the specified minimum nutrient requirement level and the fish meal level. FM inclusion levels in experimental diets 1 through 6 were 55% (diet 1), 40% (diet 2), 30% (diet 3), 20% (diet 4), 10% (diet 5), and 0% (diet 6), respectively. Digestible energy level was maintained at 3,500 kcal/kg and the digestible protein level was maintained at 41% across the six diets. Adequate levels of EAAs were maintained in all the diets. Nutrient levels for vitamins and minerals were maintained at levels generally recommend for fish by the National Research Council in 1993. Ingredient costs of the six diets varied from $865/metric ton ($785/ton) (diet 1) to $527/metric ton ($478/ton) (diet 6). To further reduce the feed cost; diet 7 was prepared by reducing the protein level from 41% to 40% and the energy level from 3,500 kcal /kg to 3,333 kcal/kg, thereby reducing the ingredient cost from $527/metric ton ($478/ton) (0% FM) to $480/metric ton ($435/ton) (0% FM). Protein sources included in the seven experimental diets were FM, PBM, MBM, BM, SBM, and CGM.

The feed formulation program selected MBM, SM, and CGM as the primary protein sources in the protein blend as the FM level was gradually reduced. Three commercial diets (Silver Cup Fish Feed, Nelson & Sons, Inc., Utah) were included in the study (control diets): high-energy trout diet (diet 8: 46% protein, 16% fat, 4,325 kcal digestible energy/kg); low-energy trout diet (diet 9: 42% protein, 12% fat, 3,550 kcal digestible energy/kg); and catfish diet (diet 10: 36% protein, 7% fat, 3,450 kcal digestible energy/kg). Four bluegill groups (~22 g [0.78 oz], *N* = 10 fish/group) were fed the experimental diets twice daily to apparent satiation. Response variables, SGR, feed consumption, FCR, HSI, viscerosomatic index (VSI), and whole-body composition (moisture, crude lipid, crude protein and crude ash) were determined to evaluate the experimental and commercial diets. Cost of the experimental diets was calculated by multiplying the ingredient cost involved in each of the diets and the amount of diet consumed by the fish group.

Among fish fed computer-formulated diets 1 through 7, no significant differences were observed in SGR, feed consumption, FCR, HSI, and VSI. However, fish fed diets 1 and 2 produced higher body fat and showed lower moisture content than fish fed diets 3 through 7 ($P < 0.05$, ANOVA). No significant differences were observed among the fish in terms of whole-body protein and ash contents. Fish fed diets 1 and 2 consumed slightly higher levels of diet than fish fed the other experimental diets which may have caused higher body fat deposition for those fish.

Comparison of fish fed the experimental diets versus those fed the industry standard diets showed fish fed the catfish feed (diet 10) to have a poorer SGR than those fed diets 1 through 3, and diet 8. Feed consumption was higher for fish fed experimental diets 1 through 7 than for the fish fed diet 10. However, fish fed diets 3 through 7 showed higher FCR than the fish fed diet 8. Similarly, fish diets 3 and 5 showed higher FCR than fish fed diet 10. Fish fed diets 8 and 9 produced significantly higher HSIs than the fish fed diets 4, 5, and 6. Similarly, fish fed diet 10 produced a higher HSI than those fed diets 2 through 6. Fish fed diet 8 produced a higher VSI value than fish fed diets 4 and 5. Similarly, fish group fed diets 8 and 9 showed higher whole-body-lipid deposition than the fish fed diets 3 through 7.

Therefore, fish fed two of the commercial trout feeds (diets 8 and 9) gained body weight more through fat deposition than protein deposition. Higher energy levels in the trout feeds versus the optimal dietary energy level determined in the present study likely caused the increased body-fat deposition. Also, crude lipid level in the experimental diets were maintained at ≤8.3%, whereas estimated lipid levels in trout diets 8 and 9 were 13.7% and 9.7%, respectively.

Fish fed diets 6 and 7 showed significantly higher gain to cost ratios than fish fed diets 1 through 5, while no differences were observed in this ratio between fish fed diets 6 and those fed diet 7. Comparison of ingredient costs of diets 6 and 7 to that of diet 1 showed the cost of diet 6 to be 39% lower, whereas the ingredient cost of diet 7 was 45% lower than the cost of diet 1. However, fish fed diet 7 (6.7%) showed slightly higher whole-body lipid deposition than fish fed diet 6 (7.3%). While the feed cost of diet 6 was ~40% below that of diet 1, diet 6 also reduced whole-body lipid deposition by 25%, 33%, and 27% versus diets 1, 8, and 9, respectively. Cost comparisons against the commercial diets could not be made as the dietary formulations of trout and catfish diets are not reported. Based on these study results diet 6 is recommended as the least-cost diet for bluegill. Formulation of diet 6 is given in Table 1 of the Appendix to this termination report.

IMPACTS

Fish producers have often used trout and catfish diets when rearing bluegill. However, trout diets, as demonstrated in the present study, cause high body-fat deposition in bluegill whereas catfish diets substantially reduce bluegill growth rates. Profit margins, particularly for bluegill producers aiming to rear larger, food-size bluegill, have undoubtedly been negatively impacted in many cases by feeding high-cost trout diets to bluegills to achieve rapid growth. The present study has developed a fish-meal-free diet based on locally available, low-cost ingredients. This diet's ingredient costs are not only ~32% cheaper versus a control diet (55% fish meal diet), but also satisfy optimal dietary nutrient requirements as determined in the present study for juvenile bluegill. The newly developed least-cost diet also produced a ~30% reduction in the whole-body fat deposition compared to the trout diets. Reducing body fat deposition while maintaining desirable fish growth rates (similar to those produced by industry standard diets) should lead to higher fillet yields.

RECOMMENDED FOLLOW-UP ACTIVITIES

The identification of a new least-cost diet for bluegills through laboratory studies still need to be tested under field conditions with production scales used by regional producers. It is anticipated that these fish reared in ponds will not only feed on the provided diets but also on natural feed stuffs including aquatic invertebrates. The combination of a commercial diet with these natural feedstuffs may result in different production parameters. Previous NCRAC-funded projects have revealed the importance of these natural feedstuffs to bluegill reared in ponds.

SUPPORT

NCRAC provided $80,000 to the University of Missouri-Columbia which was the entire amount of funding allocated for this project.

PUBLICATIONS, MANUSCRIPTS, WORKSHOPS, AND CONFERENCES

See the Appendix for a cumulative output for all NCRAC-funded Nutrition/Diets activities.

APPENDIX

Table 1. Formulation of a least-cost diet developed for juvenile bluegill.

Ingredients[Source]	Price*		Amount (percent by weight)
	$/metric ton	$/ton	
Porcine meat and bone meal[1]	270.07	245.00	38.01
Soybean meal[2]	336.21	305.00	36.99
Corn gluten meal[3]	518.09	470.00	15.29
Corn[4]	142.45	129.23	3.60
Fish Oil[5]	1,477.10	1,340.00	4.00
Lecithin[6]	4,188.79	3,800.00	0.30
Dicalcium phosphate	11,023.12	10,000.00	0.20
Vitamin premix[7]	11,023.12	10,000.00	1.00
Vitamin C	1,543.24	1,400.00	0.07
Choline chloride	1,543.24	1,400.00	0.14
Mineral mix[7]	1,543.24	1,400.00	0.10
Binder[8]	2,314.86	2,100.00	0.30
			100.00

*Prices in $/ton of porcine meat and bone meal, soybean meal, corn gluten meal, and corn are average values for the price of those ingredients for the first week of every month from the period January 2008 to December 2009 as reported in the weekly newspaper "Feedstuffs." The prices in $/ton of other ingredients are those charged by the respective sources when this diet was prepared.

[1]American Midwest Distributors, LLC, Kansas City, Missouri.
[2]ADM Soybean Meal Plant, Mexico, Missouri.
[3]Grain Processing Corporation, Muscatine, Iowa.
[4]Bourn Feed, Columbia, Missouri.
[5]Refined Menhaden Oil (Virginia Prime Gold), Omega Protein, Inc., Houston, Texas.
[6]Archer Daniels Midland Company, Decatur, Illinois.
[7]Nelson's Silvercup Fish Feed, Nelson & Sons, Inc., Murray, Utah.
[8]Ultra-Bond™, Uniscope, Inc., Johnstown, Colorado.

APPENDIX

NUTRITION/DIETS

Publications in Print

Brown, P.B. 2006. Nutrition. Pages 45-50 *in* S.D. Hart, D.L. Garling, and J.A. Malison, editors. Yellow perch (*Perca flavescens*) culture guide. North Central Regional Aquaculture Center, NCRAC Culture Series #103. Iowa State University, Ames.

Brown, P.B. 2008. Utilization of soy products originating from soybeans in diets fed to freshwater fishes. Pages 225-260 *in* C. Webster, C. Lim, and C.-S. Lee, editors. Alternative protein sources in aquaculture diets. Haworth Press, Taylor and Francis Group, New York.

Brown, P.B., S.J. Kaushik, and H. Peres. 2008. Protein feedstuffs originating from soybeans. Pages 205-223 *in* C. Webster, C. Lim, and C.-S. Lee, editors. Alternative protein sources in aquaculture diets. Haworth Press, Taylor and Francis Group, New York.

Brown, P.B., B.J. Brown, S. Hart, J. Curry, and A. Hittle-Hutson. 2008. Comparison of soybean-based practical diets containing 32, 36, or 40% crude protein fed to hybrid striped bass in earthen culture ponds. North American Journal of Aquaculture 70:128-131.

Gatlin, D.M., III, F.T. Barrows, P. Brown, K. Dabrowski, T.G. Gaylord, R.W. Hardy, E. Herman, G. Hu, A. Krogdahl, R. Nelson, K. Overturf, M. Rust, W. Sealey, D. Skonberg, E.J. Souza, D. Stone, R. Wilson, and E. Wurtele. 2007. Expanding the utilization of sustainable plant products in aquafeeds: a review. Aquaculture Research 38:551-579.

Kasper, C.S., B.A. Watkins, and P.B. Brown. 2007. Evaluation of two soybean meals fed to yellow perch (*Perca flavescens*). Aquaculture Nutrition 13:431-438.

Lewis, H.A. 2006. Minimum dietary fish oil requirement to maintain highly unsaturated fatty acid concentrations in the fillets of sunshine bass fed diets containing little or no fish meal. Master's thesis. Southern Illinois University-Carbondale.

Lewis, H.A. and C.C. Kohler. 2008. Minimizing fish oil and fish meal in sunshine bass diets without negatively impacting growth and fillet fatty acid profile. Journal of the World Aquaculture Society 39:573-585.

Lewis, H.A. and C.C. Kohler. 2008. Corn gluten meal partially replaces dietary fish meal without compromising growth or the fatty acid composition of sunshine bass. North American Journal of Aquaculture 70:50-60.

Twibell, R.G., M.E. Griffin, B. Martin, J. Price, and P.B. Brown. 2003. Predicting dietary essential amino acid requirements for hybrid striped bass. Aquaculture Nutrition 9:373-382.

Manuscript

Hart, S.D., B.J. Brown, N.L. Gould, M.L. Robar, E.M. Witt, and P.B. Brown. In press. Predicting optimal dietary essential amino acid profile for growth of juvenile yellow perch with whole body amino acid concentrations. Aquaculture Nutrition.

Papers Presented

Lewis, H.A. and C.C. Kohler. 2006. Plant-based protein sources partially replace menhaden fish meal in practical diets fed to juvenile sunshine bass. Aquaculture America 2006, Las Vegas Nevada, February 13-16, 2006.

Lewis, H.A. and C.C. Kohler. 2006. Plant-based protein sources partially replace menhaden fish meal in practical diets fed to juvenile sunshine bass. Illinois Chapter of the American Fisheries Society, Rend Lake, Illinois, March 9, 2006.

Lewis, H.A. and C.C. Kohler. 2006. Plant-based protein sources partially replace menhaden meal in practical diets for juvenile sunshine bass. American Fisheries Society Annual Meeting, Lake Placid, New York, September 10-14, 2006.

Brown, P.B. 2007. Recent advances in yellow perch nutrition. Aquaculture 2007, San Antonio, Texas, February 26-March 2, 2007.

Lewis, H.A. and C.C. Kohler. 2007. Minimizing fish oil and fish meal in sunshine bass diets without negatively impacting growth and fillet fatty acid profile. Annual Meeting of the American Fisheries Society, San Francisco, California, September 2-7, 2007.

Lewis, H.A., and C.C. Kohler. 2007. Dietary menhaden oil requirement of sunshine bass *Morone chrysops* × *M. saxatilis* fed diets containing 20% menhaden meal to maintain aquaculture production and fillet quality. Aquaculture 2007, San Antonio, Texas, February 26-March 2, 2007.