Apparent digestibility coefficients of Empyreal 75 determined in vivo using post-juvenile rainbow trout Oncorhynchus mykiss

2010

RONALD W. HARDY, PH.D.
University of Idaho – Aquaculture Research Institute
Hagerman Fish Culture Experiment Station
3059F National Fish Hatchery Road
Hagerman, ID 83332
Tel: 208-837-9096 | Fax: 208-837-6047
E-mail: rhardy@uidaho.edu

INTRODUCTION

Corn gluten meal is a mainstay among plant protein concentrates for use in feeds for salmon and trout. However, its use is limited by a defined and actual protein content of 60%. In modern, high-energy *Salmonid* feed formulations, all ingredients must have a purpose in the formulation and not bring unwanted or poorly digested components into the feed. Conventional corn gluten meal contains 60% crude protein but also 40% other material (starch, soluble, fiber, ash). Cargill has developed a corn protein concentrate (Empyreal 75) that addresses this issue.

AIM

The aim of this study was to determine the digestibility of nutrients from the Empyreal 75 meal by rainbow trout. Establishing apparent digestibility coefficients (ADC) values for protein, lipid, energy, amino acid, and phosphorus will provide a better understanding of the suitability of the product as an alternative to fishmeal. The information will also enable us to correctly formulate experimental diets for further growth studies if such a course of action is warranted.

MATERIALS AND METHODS

The determination of *in vivo* digestibility of an ingredient is the first logical step to take when screening the potential of an ingredient as an alternative to fishmeal for cultured fish. Due to possible differences in nutrient digestibility due to fish size, we assessed the digestibility of nutrients from ADF products (ingredient nutrient composition, Table 1) using post-juvenile (150g/fish) rainbow trout. We analyzed the product at one inclusion level (30%). This is the recommended inclusion content to analyze a product for *in vivo* nutrient digestibility in fish.

Feeds: To investigate the digestibility of Empyreal 75 by rainbow trout, we manufactured two separate diets (Table 2):

1. A 5-kg batch of reference diet.
2. A 5-kg batch of test diet containing 30% Empyreal 75 meal and 70% of the reference diet.

An indigestible inert marker (yttrium oxide) was included in each diet at 0.01%. Pellet size was 3 mm. In order to determine the digestibility of the product by rainbow trout, the analyses' results of the test diet containing Empyreal 75 were compared to the analyses of the control diet and the relevant calculations were made according to the methods (see below).

Diets were made by compression pelleting at the Hagerman Fish Culture Experiment Station. Samples of each diet were taken for proximate, energy, amino acid, mineral, and yttrium analyses. The study was designed as a completely randomized arrangement for statistical evaluation of data, and diets were assigned randomly to tanks within the fish-rearing laboratory at the Hagerman Fish Culture Experiment Station.

Fish maintenance and feeding regime: At stocking, rainbow trout were weighed and counted into groups of 60 and placed into 450-L digestibility tanks. There were two treatment groups, with two replicate tanks of fish for each treatment. Each tank was supplied with untreated, constant-temperature (15°C) spring water at a flow rate of 8-L min^{-1}. Photoperiod was maintained at a constant 14 h light: 10 h dark with fluorescent lights.

Fish were fed their respective diets twice daily, at 0830–0900 and 1530–1600h to apparent satiation for two weeks. Feces were then collected by manually stripping each fish after partial anesthesia (MS-222, 100 mg L^{-1}) during the second week. Fecal material from fish within each tank was pooled, and each tank was analyzed separately. Fish were handled and treated in accordance with the guidelines approved by the Animal Care and Use Committee of the University of Idaho.

Chemical analyses: The ingredient, two diets, and four fecal samples were dried in a convection oven at 105°C for 12 h to determine moisture level according to AOAC (1990). Dried samples were finely ground by mortar and pestle and analyzed for crude protein (total nitrogen x 6.25) using a LECO FP-428 nitrogen analyzer (LECO Instruments, St. Joseph, Michigan). Crude lipid was analyzed using an ANKOM XT15 extraction apparatus (ANKOM Technology, Macedon, New York) with petroleum ether as the extracting solvent, and ash by incineration at 550°C in a muffle furnace. The energy content of samples was determined using a Parr Adiabatic Calorimeter (Parr Instruments, Moline, Illinois). AAA Laboratories at Mercer Island, Washington analyzed amino acids (excluding tryptophan). Analyses of phosphorus and yttrium were conducted by the University of Idaho, Holm Center, Moscow, Idaho.

Calculations of apparent digestibility coefficients of test ingredients: Apparent digestibility coefficients (ADCs) of ingredients for dry matter, protein, amino acids, lipid, energy, and phosphorus were calculated using the following formulae described by Sugiura et al. (1998):

Step 1. Diet ADC:
ADC test diet (%) = 100 x [1 – (% marker in diets/% marker in feces) x (% nutrient in feces/% nutrient in diets)]

Step 2: Ingredient ADC:
ADC ingredient (%) = (Nutrient test diet x ADC test diet – 0.7 x nutrient reference diet x ADC reference diet) / (0.3 x nutrient test ingredient)

RESULTS AND DISCUSSION

NUTRIENT PROFILE OF TEST INGREDIENTS

The nutritional profile of the test ingredient (Table 1) indicates that it has a favorable protein level to replace a substantial proportion of dietary fishmeal for rainbow trout as well as most other cultured species of fish. The protein content of the Empyreal 75 meal is comparable to or higher than levels found in fishmeals. With regard to essential amino acids, and compared to fishmeal, which is the standard dietary protein source used for carnivorous fish, the levels of essential amino acids found in the Empyreal 75 meal were lower than fishmeal for all essential amino acids except PHE. The amino acid profile of Empyreal 75 was similar to that of other corn protein products. The lipid level was low, as expected.

GENERAL OBSERVATIONS WITH DIGESTIBILITY EXPERIMENT

This experiment examined the nutrient digestibility and availability to fish and did not attempt to measure growth. Growth of fish fed diets containing ADF products would have to be measured in a specifically designed study. However, there were no signs of disease or mortality in this study and fish accepted the experimental diets well, indicating that the test ingredient (Empyreal 75) was highly palatable at the 30% inclusion level.

DIGESTIBILITY OF THE TEST INGREDIENTS

Apparent digestibility coefficients (ADCs) of nutrients from the Empyreal 75 meal for rainbow trout was excellent and in line with values for corn gluten meal measured in earlier studies (Table 2). With the exception of phosphorus and valine, the essential nutrient ADCs of the Empyreal 75 meal were all above 81%. The Empyreal 75 meal was similar in digestible protein level (55.6%) compared to digestible protein values obtained using the settlement technique for herring meal (64%) and menhaden meal (53%) reported for rainbow trout by Cheng and Hardy (2002).

Additionally, all digestible essential amino acid levels in the Empyreal 75 meal were higher when compared to herring meal and menhaden meal and poultry meals reported for rainbow trout by Cheng and Hardy (2002). The Empyreal 75 meal provided respectable digestible energy for rainbow trout. The digestible energy value of the Empyreal 75 meal (3,891 kcal kg^{-1}) is in the middle of the range of values reported for poultry and fishmeals, e.g., menhaden meal (3,106 kcal kg^{-1}), feed-grade poultry meal (4,061 kcal kg^{-1}), prime poultry meal (3,822 kcal kg^{-1}) and herring meal (4,420 kcal kg^{-1}) (Cheng and Hardy, 2002).

The ADC value for phosphorus from Empyreal 75 was better than expected. Bearing in mind that phosphorus excretion by fish is one of the major concerns associated with effluents from fish farms, this is an important point. The ADC_{phos} value was in line with values reported for other products, e.g., feed-grade poultry meal (ADC 49.4%), prime poultry meal (ADC 45.8%) and refined poultry meal (ADC 56.0%), and herring meal (ADC 58.2%) (Cheng and Hardy, 2002).

CONCLUSIONS AND RECOMMENDATIONS

Based on the digestible nutritional profile of Empyreal 75 compared to herring meal, Empyreal 75 meal has the potential to replace a significant proportion of fishmeal protein in diets for rainbow trout and other species of cultured *Salmonid* fish. It is critical, however, to remember that digestibility does not necessarily equal potential utilization for growth or value in a feed formulation. Amino acid profiles, especially levels of limiting amino acids, determine the extent to which ingredients are used. Digestibility determinations are the first step in determining the potential of an ingredient for inclusion into aquafeeds. As the outcome of the digestibility trial was positive, Cargill may wish to consider conducting a growth experiment to further investigate the potential of the Empyreal 75 meal as an alternative ingredient to fishmeal in diets for rainbow trout. A growth study would investigate any positive or negative impacts that these ingredients may have on growth performance, effluent levels, and quality of the finished product (fillet). Ultimately the use of Empyreal 75 as ingredients for rainbow trout feeds will be dependent upon pricing relative to competing ingredients.

Proximate, energy, amino acid, and phosphorus content of Cargill Empyreal 75[a] compared to herring fishmeal.

Nutrient or proximate category	Empyreal 75[b]	Chilean fishmeal
Crude protein (%)	82.9	71.0
Crude lipid (%)	2.4	9.2
Energy (kcal/kg)	5,902	5,072
Phosphorus (g/kg)	3.2	2.4
Essential Amino Acids (%)		
Arginine	2.77	4.50
Histidine	1.53	2.24
Isoleucine	2.99	3.24
Leucine	13.50	5.25
Lysine	0.96	5.40
Methionine	1.64	1.84
Phenylalanine	5.22	2.84
Threonine	2.88	3.10
Valine	3.96	3.63
Nonessential Amino Acids (%)		
Alanine	7.08	
Aspartic acid	4.71	
Glutamic acid	17.37	
Glycine	2.10	
Proline	8.37	
Serine	4.51	
Tyrosine	4.52	

[a] Cystine and tryptophan were not measured. | [b] Expressed on a dry-weight basis.

Table 1.

Apparent digestibility coefficients (%) of Cargill Empyreal 75 measured *in vivo* using post-juvenile rainbow trout[a]	
Nutrient or proximate category	**ADC Value**
Crude protein	74.2
Curde lipid	100[b]
Energy	65.93
Phosphorus	53.6
Essential Amino Acids	
Arginine	93.0
Histidine	84.9
Isoleucine	83.5
Leucine	81.4
Lysine	89.5
Methionine	82.7
Phenylalanine	84.9
Threonine	82.7
Valine	74.9
Nonessential Amino Acids	
Alanine	78.9
Aspartic acid	79.2
Glutamic acid	81.3
Glycine	77.8
Proline	758
Serine	83.6
Tyrosine	86.9

[a] Cystine and tryptophan were not measured.
[b] Empyreal 75 contains about 2% lipid, making it virurally impossible to accurately measure ADC $_{\text{lipid}}$.

Table 2.

Allan, G.L., Parkinson, S., Booth, M.A., Stone, D.A.J., Rowland, S.J., Frances, J., and Warner-Smith, R. 2000. Replacement of fishmeal in diets for Australian silver perch, *Bidyanus bidyanus*: I. Digestibility of alternative ingredients. Aquaculture 186: 293–310.

Alexis, M.N, Papaparaskeva-Papoutsoglou, E., and Theochari, V. 1985. Formulation of practical diets for rainbow trout (*Salmo gairdneri*) made by partial or complete substitution of fishmeal by poultry by-products and certain plant by-products. Aquaculture 50: 61–73.

AOAC. 1990. Official Methods of Analysis of the Association of Official Analytical Chemists, 15th Edition. Association of Official Analytical Chemists Inc. Arlington, Virginia. pp. 1,298.

Cheng, Z.J. and Hardy R.W. 2002. Apparent digestibility coefficients of nutrients and nutritional value of poultry by-product meals for rainbow trout *Onchorynchus mykiss* measured *in vivo* using settlement. Journal of the World Aquaculture Society 33: 458–465.

Dong, F.M., Hardy R.W., Haard N.F., Barrows F.T. Rasco, B.A. Fairgrieve, W.T., and Forster, I. P. 1993. Chemical composition and protein digestibility of poultry by-product meals for *Salmonid* diets. Aquaculture 116: 149–158.

Ferraris, R.P., Catacutan, M.R., Mabelin, R.L., and Jazul, A.P. 1986. Digestibility in milkfish, *Chanos chanos* (Forsskal): Effects of protein source, fish size, and salinity. Aquaculture 59: 93–105.

Kitamikado, M., Morishita, T. and Tachino, S. 1964. Digestibility of dietary protein in rainbow trout-II. Effects of starch and oil content in diets, and size of fish. Bulletin of the Japanese Society of Scientific Fisheries, 30: 50–54.

NRC (National Research Council) 1993. Nutrient Requirement of Fish. Committee on Animal Nutrition, Board of Agriculture, National Research Council. National Academic Press, Washington, D.C., p. 114.

Steffens, W. 1994. Replacing fishmeal with poultry by-product meal in diets for rainbow trout, *Oncorhynchus mykiss*. Aquaculture 124: 27–34. Cargill Corn Milling – Confidential 9.

Sugiura, S.H., Dong, F.M., Rathbone, C.K., and Hardy, R.W. 1998. Apparent protein digestibility and mineral availabilities in various feed ingredients for *Salmonid* feeds. Aquaculture 159: 177–202.

Sumagaysay-Chavoso, N.S. 2003. Nitrogen and phosphorus digestibility and excretion of different-sized groups of milkfish (*Chanos chanos Forsskal*) fed formulated and natural food-based diets. Aquaculture Research 34: 407–418.

Usmani, N., and Jafri, A.K. 2002. Effect of fish size and temperature on the utilization of different protein sources in two catfish species. Aquaculture Research 33: 959–967.

Windell, J.T., Foltz, J.W., and Sarokon, J.A. 1978. Effects of fish size, temperature, and amount fed on nutrient digestibility of a pelleted diet by rainbow trout, *Salmo gairdneri*. Transactions of the American Fisheries Society,107: 613–616.