The Open Access Israeli Journal of Aquaculture – Bamidgeh

As from **January 2010** The Israeli Journal of Aquaculture - Bamidgeh (IJA) will be published exclusively as **an on-line Open Access (OA)** quarterly accessible by all AquacultureHub (http://www.aquaculturehub.org) members and registered individuals and institutions. Please visit our website (http://siamb.org.il) for free registration form, further information and instructions.

This transformation from a subscription printed version to an on-line OA journal, aims at supporting the concept that scientific peer-reviewed publications should be made available to all, including those with limited resources. The OA IJA does not enforce author or subscription fees and will endeavor to obtain alternative sources of income to support this policy for as long as possible.

Published under auspices of
The Society of Israeli Aquaculture and Marine Biotechnology (SIAMB), University of Hawaii at Manoa Library
and
University of Hawaii Aquaculture Program in association with
AquacultureHub
http://www.aquaculturehub.org

ISSN 0792 - 156X

PUBLISHER:
Israeli Journal of Aquaculture - BAMIGDEH -
Kibbutz Ein Hamifratz, Mobile Post 25210, ISRAEL
Phone: + 972 52 3965809
http://siamb.org.il

Effects of Partial Replacement of Fishmeal with Isolated Soy Protein on Digestibility and Growth Performance in Sterlet *(Acipenser ruthenus)*

S. Ustaoglu[1]* and B. Rennert[2]

[1] *Faculty of Fisheries, University of Ondokuz Mayis, 57000 Sinop, Turkey*

[2] *Leibniz-Institute of Freshwater Ecology and Inland Fisheries, Muggelseedamm 310, 12587 Berlin, Germany*

(Received 18.12.05, Accepted 14.2.06)

Key words: *Acipenser ruthenus*, sterlet, nutrition, fishmeal replacement, isolated soy

Abstract

Fishmeal was partially replaced by isolated soy protein in diets for sterlet (*Acipenser ruthenus*) juveniles at an amount at which one-half or one-third of the protein was supplied by soy. Nutrient digestibility, growth, and whole body composition were determined after 79 days. Protein digestibility was significantly higher in the diet in which half the protein derived from isolated soy protein (84.9%) than in the diet in which one-third of the protein derived from soy (82.6%). Otherwise, digestibility, growth, feed efficiency, and whole body composition did not differ between the two diets.

Introduction

Fishmeal is a major ingredient in dry commercial fish diets because of its high protein quality and palatability. It provides high essential amino and fatty acid contents and is usually well digested by fish. However, fishmeal production already uses approximately 35% of the total global fish catch. About 4 kg of wet fish is needed to produce 1 kg dry fishmeal. Therefore, if a diet contains more than 17% fishmeal and the feed conversion ratio exceeds 1.5:1, the aquaculture process results in a net loss of fish protein (Allan et al., 2000).

One consequence of the world-wide decline of fish stocks is the uncertain availability of fishmeal, necessitating reduction of the fishmeal content in fish diets and a search for suitable replacements for protein sources. Successful replacements are mostly of animal origin and include milk and whey powder, krill meal, single-cell protein, and algae meal. Unfortunately, most of these materials are as scarce or expensive as fishmeal.

Soybean products are the most promising protein sources for replacing part or all of the fishmeal protein in fish diets because of their high protein content, well-balanced amino

* Corresponding author. E-mail: ustaoglu@omu.edu.tr

acid profile, consistent composition, reasonable price, and steady supply.

Some workers concluded that soybean protein can only partially replace fishmeal. Olli et al. (1995) showed that it is possible to substitute 20% of fishmeal with soybean meal in diets for Atlantic salmon (*Salmo salar*) and Storebakken et al. (1998) reported rapid growth in Atlantic salmon fed diets with 75% total protein from soy protein concentrate. However, Kaushik et al. (1995) found that soy protein concentrate could replace 100% of fishmeal in diets for rainbow trout (*Oncorhynchus mykiss)* without negative effects on the growth rate while poor growth and protein utilization resulted from the inclusion of soybean meal in diets for rainbow trout (Dabrowski et al., 1989) and channel catfish (Wilson and Poe, 1985). The differences among results could be due to the source of the soy protein or the fish species.

The effectiveness of soy products as protein sources for fish depends on the extraction and processing techniques (Oliva-Teles et al., 1994). Soy protein concentrate and soy protein isolate are produced using various techniques including heat treatment to remove or deactivate antinutritional factors, oligosaccharides, and fiber (Bureau et al., 1998). Isolated soy protein is almost-pure soybean protein (at least 90%; Poston, 1991).

The search for alternative protein sources for other species has received considerable attention, but there has been comparatively little work on sturgeon species and only one on sterlet (*Acipenser ruthenus*; Ustaoglu and Rennert, 2002). In that study, total replacement of fishmeal with isolated soy protein reduced the growth rate, isolated soy protein was better digested (93.6%) than fishmeal (91.2%), and it was recommended that sterlet diets containing a combination of fishmeal and isolated soy protein be evaluated. The purpose of the present study was to examine the partial replacement of fishmeal in juvenile sterlet diets, substituting isolated soy protein so that one-third or one-half of the dietary protein was supplied by isolated soy protein.

Sterlet is a relatively small (usually up to 3 kg) freshwater sturgeon. Husbandry and experimentation are relatively easy due to its small size, making it ideal for investigation in recirculating systems (Ustaoglu and Rennert, 2002). Thus, sterlet is a model fish for study of sturgeons. The results on sterlet are likely transferable to other sturgeons.

Materials and Methods

Experimental fish and maintenance conditions. Juvenile sterlet (*Acipenser ruthenus*) were obtained from a commercial fish farm in Wollershof, Germany. The experiment was performed in two thermoregulated semi-recirculating systems, each equipped with three 60-l cylindrical fiberglass tanks with an apparatus for automatic collection of fish feces (Choubert et al., 1982) and aerated water at a flow rate of 4 l/min. Water temperature, dissolved oxygen, and pH, measured each morning, were 19.8-20.2°C, 7.0-8.1 mg/l, and 7.8-7.9.

Twelve fish weighing about 36 g each were randomly allocated to each tank and triplicate groups of these fish were used. The fish were adapted to the diets and the experimental conditions for two weeks prior to the start of eight weeks of fecal collection. At the start of the trial 18 fish from the initial pool were killed, homogenized, and frozen for body composition analysis. At the end of the experiment, the fish were weighed after a 24-h fast. Seven fish were randomly taken from each tank for whole body composition analysis. The samples were immediately frozen and stored at -20°C until analyzed.

Experimental diets. Two experimental diets were formulated to compare the effects of a different amount of soy protein on the performance variables. In diet I, half the diet protein was from fishmeal and half was from isolated soy protein. In diet II, two-thirds of the diet protein was from fishmeal and one-third from isolated soy protein. Diet ingredients were obtained from commercial sources. Ingredients were thoroughly mixed, water was added to obtain a moisture level of 30%, the mixtures were passed through a pellet machine and compressed into 1 mm diameter pellets, and the pellets were dried at room temperature. Chromic oxide was incorporated into the test diets at an inclusion level of 0.5% and used as an inert marker for computing apparent digestibility coefficients (ADC). The diets were stored at 6ºC until use. The formulations, proximate analyses, and amino acid compositions are shown in Table 1.

Feeding and fecal collection. Diets were fed by hand three times per day and six days per week at 1.5% of the body weight per day. Fish were weighed at two-week intervals and the amount of feed was adjusted accordingly. The fish were not fed on the day of weighing. Feces were collected by filtering the effluent water from the rearing tanks. Feces were collected every 15 minutes for eight weeks starting after the first feeding. Samples were immediately frozen and stored at -20°C until chemical analysis.

Chemical analyses. Diets, feces, and body composition were analyzed by identical methods (Naumann and Bassler, 1976). Dry matter content was determined by freeze drying for 48 hours at -54°C (Christ, Alpha 1-4), crude protein (N x 6.25) according to the Kjeldahl method (Kjeltec System-Tecator), crude lipid by petroleum ether extraction in a Soxhlet apparatus (Soxtec System HT-Tecator), ash by incineration for 4 h at 750°C, and gross energy content in an oxygen bomb calorimeter (Framo-MK 200). The nitrogen-free extract plus fiber component was determined by the equation: NFE + fiber = 100 - (% protein + % lipid + % ash). Total phosphorus in diets was determined by the method of Zwirnmann et al. (1999) and chromic oxide by spectrophotometer after acid digestion (Petry and Rapp, 1970). Apparent digestibility coefficients were calculated as ADC (%) = 100 - [100 (% Cr_2O_3 in diet/% Cr_2O_3 in feces) x (% nutrient in feces/% nutrient in diet)] (Degani et al., 1997). Amino acids were analyzed following acid hydrolysis using high pressure liquid chromotagraphy at the Institute for Cereal Processing in Potsdam, Germany. All chemical analyses were carried out in duplicate.

Statistical analysis. Differences between results were analyzed by *t* test using SPSS for Windows (Release 6.1.3; standard version 1989-1995) and considered significant if $p<0.05$.

Results

Acceptance of the experimental diets was good and no mortality was observed during the experiment. Apparent digestibility and growth performance are presented in Table 2. Protein digestibility was significantly higher in diet I, indicating good digestion of isolated soy protein by sterlet. Growth performance did not significantly differ between treatments, indicating that including 50% protein from isolated soy protein in sterlet diets has no adverse effects on growth performance or feed efficiency. Body composition is shown in Table 3. The composition of fish from both treatments was identical.

Discussion

Of all the protein-rich plant feedstuffs, soybean has one of the best amino acid profiles for meeting the essential amino acid requirements of fish. However, the replacement of fishmeal protein by soy protein has met variable success. Many fish studies resulted in considerable success in partial or total replacement with soybean meal and other soybean products. In other cases, growth decreased in direct relation to the level of fishmeal replacement.

There are few investigations on the substitution of fishmeal with soy products in strugeon feeds. Inclusion of 40% soy protein concentrate in a diet for juvenile white sturgeon resulted in poorer growth compared to a purified test diet (Stuart and Hung, 1989). Ustaoglu and Rennert (2002) found that total replacement of fishmeal with isolated soy protein reduced the growth rate in sterlet, although the isolated soy protein diet was better digested (93.6%) than the fishmeal diet (91.2%). They suggested that the growth retardation may have resulted from methionine and phosphorus deficiencies in the isolated soy protein diet. Although soy protein has a well-balanced amino acid profile for fish, it is low in methionine (Storebakken et al., 2000) and contains phytic phosphorus which is of limited availability for fish (Lall, 1991; NRC, 1993). Therefore, it may be best to combine fishmeal and isolated soy protein in feeds.

Viola et al. (1982) reported that for practi-

Table 1. Ingredients, chemical composition, and amino acid contents of experimental diets.

	Diet I	*Diet II*
Ingredient (g/kg)		
Fishmeal	319.0	425.0
Isolated soy protein	261.0	174.0
Fish oil	113.0	101.0
Wheat starch	232.0	225.0
Vitamin-mineral premix[1]	70.0	70.0
Cr_2O_3	5.0	5.0
Proximate composition (%)		
Dry matter	92.80	92.84
Crude ash[2]	10.49	11.46
Crude lipid[2]	15.38	15.33
Crude protein[2]	47.31	48.49
Total phosphorus[2]	0.88	1.00
NFE + fiber[3]	26.82	24.72
Cr_2O_3	0.48	0.47
Gross energy (kJ/g)	21.98	21.86
Amino acid (% of diet)		
Aspartic acid	3.41	3.27
Glutamic acid	6.12	5.19
Serine	1.50	1.50
Histidine[4]	1.16	0.83
Glycine	2.22	2.04
Threonine[4]	1.42	1.31
Arginine[4]	3.03	2.48
Alanine	2.07	2.22
Tyrosine	1.13	0.94
Valine[4]	2.27	1.90
Phenylalanine[4]	1.88	1.67
Isoleucine[4]	2.41	1.73
Leucine[4]	3.06	2.76
Lysine[4]	2.70	3.35
Methionine[4]	0.94	1.02
Cystine	0.36	0.31

1 Vitamin-mineral premix (per kg diet): vitamin A, 210,000 IU; vitamin D3, 35,000 IU; vitamin E, 7000 mg; vitamin K3, 322 mg; vitamin B1, 588 mg; vitamin B2, 252 mg; vitamin B6, 294 mg; vitamin B12, 826 mcg; niacin, 1400 mg; biotin, 7583.1 mcg; folic acid, 182 mg; pantothenic acid, 1722 mg; inositol, 17,220 mg; vitamin C, 933.31 mg; Ca, 1414 mg.

2 Dry matter basis.

3 Nitrogen-free extract + fiber = 100 - (% protein + % lipid + % ash).

4 Essential amino acids.

Table 2. Apparent digestibility of the experimental diets and growth performance factors (means of three replicates±standard error).

	Diet I	*Diet II*
Apparent digestibility (%)		
Dry matter	69.8±0.91	68.2±0.80
Crude protein	84.9±0.34[a]	82.6±0.35[b]
Crude lipid	96.2±0.34	95.8±0.55
NFE + crude fiber	45.5±1.53	44.5±1.03
Gross energy	79.6±0.72	78.7±0.65
Growth performance		
Initial body wt (g)	37.70±0.67	36.17±0.09
Final body wt (g)	62.34±1.23	62.92±0.63
Wt gain (g)	24.64±0.90	26.76±0.56
SGR (%)[1]	0.63±0.02	0.70±0.01
FCR[2]	1.5±0.14	1.3±0.03

Values in a row with different superscripts significantly differ (p<0.05).

[1] Specific growth rate = 100 x [(ln final body wt - ln initial body wt)/79 days]

[2] Feed conversion ratio = total diet fed/total wt gain

cal uses the partial replacement of half the fishmeal in carp feeds has several advantages. At this level, the deficiency of amino acids is barely noticeable. However, there is little information on the amino acid requirements of sturgeon. According to Kaushik et al. (1991), the quantitative requirements of Siberian sturgeon (*Acipenser baeri*) for most essential amino acids (except lysine and phenylalanine) are in the same range as in other teleosts. In our study, the amino acid composition, especially the amount of methionine, was relatively similar in both test diets. Further studies on the amino acid requirements are needed to formulate a protein-efficient diet for sterlet and other sturgeons.

In the present study, the experimental diets were well-accepted, indicating palatability, and there was no difference in feed intake between treatments. In other studies, high levels of plant protein feedstuffs reduced growth, mainly due to lower feed intake (Reigh and Ellis, 1992; Gomes et al., 1995). The intake of feeds containing high levels of vegetable feedstuffs was low in tilapia (Jackson et al., 1982) and extremely low in juvenile chinook salmon fed at 15% and 30% replacement levels of soybean meal and soy protein isolate (Hajen et al., 1993). On the other hand, rainbow trout and Atlantic salmon adapted to soybean meal diets and, after the adaptation period, achieved an equal feed intake as with a fishmeal diet (Refstie et al., 1997, 1998)

Feed conversion was better with the diet containing the higher proportion of fishmeal, but the difference was insignificant. Likewise, there was an insignificant difference in SGR between the experimental diets. These insignificant differences indicate that isolated soy protein may supply up to 50% of the protein in sterlet diets without adverse effects on growth. Similarly, Day and Plascencia Gonzales (2000) found that soy protein con-

Table 3. Whole body composition (% dry matter) of sterlet fed experimental diets (means of three replicates±standard error).

	Initial	*Diet I*	*Diet II*
Dry matter	24.74	26.80±0.17	26.03±0.33
Crude protein	56.84	54.56±0.59	55.98±1.62
Crude lipid	22.75	20.71±0.94	21.56±0.17
Ash	13.18	13.16±0.50	13.04±1.04
Gross energy (kJ/g)	23.70	23.12±0.27	23.41±0.21

centrate could replace 50% of fishmeal in diets for turbot without growth reduction.

The isolated soy protein was well digested by the sterlet (82 and 84%), showing that protein from isolated soy is more digestible than fishmeal protein. In Ustaoglu and Rennert (2002), isolated soy protein was 93.6% digestible by sterlet. These high apparent digestibility values are in agreement with those reported for rainbow trout and Atlantic salmon (Refstie et al., 2000) and turbot (Day and Plascencia Gonzales, 2000). Lovell (1990) reported that the protein of dehulled solvent extracted soybean meal was 85% digestible, equal to or higher than that of whole fishmeal protein. Van Der Meer et al. (1996) reported that soya protein was better utilized by *Colossoma macropomum* than fishmeal protein. This was explained by the high digestibility of soy protein. In fish, protein digestibility generally ranges 75-90% (NRC, 1993).

Lipid digestibility among fish normally ranges 85-95% (NRC, 1993). In the present study, the lipid was quite well digested by the sterlet (96%), possibly due to the high lipase activity in sterlets and most other fish (Sargent et al., 1989). Overall, fish oil is a high-energy ingredient, generally thought to be highly digestible by fish (Hardy, 1989).

The present study showed that sterlet were able to efficiently digest isolated soy protein. The digestibility of the diets was not affected by the level of isolated soy protein and isolated soy protein may account for up to half the total protein in sterlet diets without adverse effects on growth and feed efficiency when the remainder of the protein content is supplied by fishmeal. The combination of fishmeal and isolated soy protein was effective as a source of protein for sterlet, while total substitution of fishmeal by isolated soy protein significantly reduces the growth rate (Ustaoglu and Rennert, 2002). Further studies of the amino acid requirements are needed to formulate a protein-efficient diet for this species.

The price of isolated soy protein is higher than the price of fishmeal. However it seems to be an interesting alternative to a significant amount of fishmeal as a protein source in fish feeds. This can be done because isolated soy protein contains a high proportion of available protein and a well-balanced amino acid profile.

Acknowledgements

We would like to thank the Leibniz-Institute of Freshwater Ecology and Inland Fisheries, Berlin, for financial support for this experiment and for the use of the facilities at the institute.

References

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

Bureau P.D., Harris A.M. and C.Y. Cho,

1998. The effects of purified alcohol extracts from soy products on feed intake and growth of chinook salmon (*Oncorhynchus tschawytscha*) and rainbow trout (*Oncorhynchus mykiss*). *Aquaculture*, 161:27-43.

Choubert G., De La Noue J. and P. Luquet, 1982. Digestibility in fish: improved device for the automatic collection of feces. *Aquaculture,* 29:185-189.

Dabrowski K., Poczyczynski G., Kock G. and B. Berger, 1989. Effect of partially or totally replacing fishmeal protein by soybean meal protein on growth, food utilization and proteolytic enzyme activities in rainbow trout (*Salmo gairdneri*). New *in vivo* test for exocrine pancreatic secretion. *Aquaculture*, 77:29-49.

Day O.J. and H.G. Plascencia Gonzales, 2000. Soybean protein concentrate as a protein source for turbot *Scophthalmus maximus* L. *Aquac. Nutr.*, 6:221-228.

Degani G., Viola S. and Y. Yehuda, 1997. Apparent digestibility coefficient of protein sources for carp, *Cyprinus carpio* L.. *Aquac. Res.*, 28:23-28.

Gomes E.F., Rema P. and S.J. Kaushik, 1995. Replacement of fishmeal by plant proteins in the diet of rainbow trout (*Oncorhynchus mykiss*): digestibility and growth performance. *Aquaculture*, 130:177-186.

Hajen W.E., Higgs D.A., Beames R.M. and B.S. Dosanjh, 1993. Digestibility of various feedstuffs by post-juvenile chinook salmon (*Oncorhynchus tshawytscha*) in sea water. 2. Measurement of digestibility. *Aquaculture*, 112:333-348.

Hardy R.V., 1989. Diet preparation. pp. 475-548. In: J.E. Halver (ed.) *Fish Nutrition*, 2nd ed. Academic Press Inc., San Diego, CA.

Jackson A.J., Capper B.S. and A.J. Matty, 1982. Evaluation of some plant proteins in complete diets for the tilapia *Sarotheredon mossambicus*. *Aquaculture*, 27:97-109.

Kaushik S.J., Breque J. and D. Blanc, 1991. Requirements for protein and essential amino acids and their utilization by Siberian sturgeon (*Acipenser baeri*). pp. 25-39. In: *Acipenser*. Premier Colloque International sur l'Esturgeon, Oct 3-6, 1989, Bordeaux., Cemagref.

Kaushik S.J., Cravedi J.P., Lalles J.P., Sumpter J., Fauconneau B. and M. Laroche, 1995. Partial or total replacement of fishmeal by soybean protein on growth, protein utilization, potential estrogenic or antigenic effects, cholesterolemia and flesh quality in rainbow trout, *Oncorhynchus mykiss*. *Aquaculture*, 133:257-274.

Lall S.P., 1991. Digestibility, metabolism and excretion of dietary phosphorus in fish. In: C.B. Cowey, C.Y. Cho (eds.). *Nutritional Strategies and Aquaculture Waste*. Fish Nutrition Research Laboratory, Department of Nutritional Sciences, Univ. Guelp, Guelp, Ontario. 275 pp.

Lovell R.T., 1990. Use of soybean products in diets for aquaculture species. Revised. ASA Tech. Bull., AQ21-90 6/7.

Naumann K. and R. Bassler, 1976. *Handbuch der Landwirtschaftlichen Versuchs- und Unter-suchungsmethodik (Methodenbuch).* Die chemische Untersuchung von Futtermitteln, Verlag J. Neumann-Neudamm, Berlin.

NRC,1993. *Nutrient Requirements of Fish*. National Research Council, National Academy Press, Washington DC. 114 pp.

Oliva-Teles A., Gouveia A.J., Gomes E. and P. Rema, 1994. The effect of different processing treatments on soybean meal utilization by rainbow trout, *Oncorhynchus mykiss*. *Aquaculture*, 124:343-349.

Olli J.J., Krogdahl A. and A. Vabeno, 1995. Dehulled solvent-extracted soybean meal as a protein source in diets for Atlantic salmon, *Salmo salar* L. *Aquac. Res.*, 26:167-174.

Petry H. and W. Rapp, 1970. Zur Problematik der chromoxidbestimmung in verdauungsversuchen. *Z. fur Tierphysiol., Tierern. und Futtermittelkde*, 27:181-189.

Poston H.A., 1991. Response of rainbow trout to soy lecithin, choline and autoclaved isolated soy protein. *Prog. Fish-Cult*., 53:85-90.

Refstie S., Helland S.J. and Storebakken T., 1997. Adaptation to soybean meal in diets for rainbow trout, *Oncorhynchus mykiss*. *Aquaculture*, 153:263-272.

Refstie S., Storebakken T. and A.J. Roem, 1998. Feed consumption and conversion in

Atlantic salmon (*Salmo salar*) fed diets with fishmeal, extracted soybean meal or soybean meal with reduced content of oligosaccharides, trypsin inhibitors, lectins and soya antigens. *Aquaculture*, 162:301-312.

Refstie S., Korsoen O.J., Storebakken T., Baeverfjord G., Lein I. and A.J. Roem, 2000. Differing nutritional responses to dietary soybean meal in rainbow trout (*Oncorhynchus mykiss*) and Atlantic salmon (*Salmo salar*). *Aquaculture*, 190:49-63.

Reigh R.C. and S.C. Ellis, 1992. Effects of soybean and fish-protein ratios on growth and body composition of red drum (*Sciaenops ocellaatus*) fed isonitrogenous diets. *Aquaculture*, 104:279-292.

Sargent J. Henderson R.J. and D.R. Tocher, 1989. The lipids. pp. 153-218. In: J.E. Halver (ed.). *Fish Nutrition*, 2nd ed. Academic Press Inc., San Diego, CA.

Storebakken T., Shearer K.D. and A.J. Roem, 1998. Availability of protein, phosphorus and other elements in fishmeal, soy-protein concentrate and phytase-treated soy-protein-concentrate-based diets to Atlantic salmon, *Salmo salar. Aquaculture*, 161:365-379.

Storebakken T., Refstie S. and B. Ruyter, 2000. Soy products as fat and protein sources in fish feeds for intensive aquaculture. pp. 127-170. In: J.K. Drackley (ed.) *Soy in Animal Nutrition.* Fed. Anim. Sci. Soc., Savoy, IL.

Stuart S.J. and S.O. Hung, 1989. Growth of juvenile white sturgeon (*Acipenser transmontanus*) fed different proteins. *Aquaculture*, 76:303-316.

Ustaoglu S. and B. Rennert, 2002. The apparent nutrient digestibility of diets containing fishmeal or isolated soy protein in sterlet (*Acipenser ruthenus*). II. Rearing techniques of sturgeon. *Int. Rev. Hydrobiol*., 87:577-584.

Van Der Meer M.B., Huisman E.A. and M.C.J. Verdegem, 1996. Feed consumption, growth and protein utilization of *Colossoma macropomum* (Cuvier) at different dietary fishmeal/soya meal ratios. *Aquac. Res.*, 27:531-538.

Viola S., Mokady S., Rappaport U. and Y. Arielli, 1982. Partial and complete replacement of fishmeal by soybean meal in feeds for intensive culture of carp. *Aquaculture*, 26:223-236.

Wilson R.P. and W.E. Poe, 1985. Effects of feeding soybean meal with varying trypsin inhibitor activities on growth of fingerling channel catfish. *Aquaculture*, 46:19-25.

Zwirnmann E., Kruger A. and J. Gelbrecht, 1999. Analytik im Zentralen Chemielabor des IGB. *Berichte des IGB*, Heft 9:170.