Journal of Animals and Veterinary Advances 4 (4): 473-477, 2005

Soybean Meal Substitution with Roselle (*Hibiscus sabdariffa* L.) Seed Meal in Dry Practical Diets for Fingerlings of the African Catfish, *Clarias gariepinus* (Burchell 1822)

Oyedapo A. Fagbenro
Department of Fisheries and Wildlife, Federal University of Technology, PMB 704 Akure, Nigeria

Abstract: Roselle (*Hibiscus sabdariffa* var. *sabdariffa* L.) seed meal was evaluated, in digestibility and feeding experiments, as a protein source in dry pelleted diets (400g crude protein, 120g crude lipid and 19.0 MJ gross energy.kg^{-1} DM) for the African catfish, *Clarias gariepinus* (Burchell 1822). The diets contained Roselle seed meal as replacement for soybean meal in a control diet and were hand-distributed to catfish fingerlings (mean initial weight, 15.6±0.5g) to apparent satiation twice daily for 70 days. Catfish mortality was low (<10%) and was not diet related. Inclusion of Roselle seed meal, replacing up to 60% of soybean meal protein did not affect weight gain, growth response, feed conversion, protein utilization or carcass composition of catfish. Catfish growth was however retarded and feed was poorly utilized when Roselle seed meal replaced >60% of soybean meal protein, caused by reduced appetite, apparent deficiency in lysine and effects of residual antinutrients (tannin, protease inhibitors, phytic acid and gossypol) in Roselle seed meal. The toxicity of the antinutrients was however light as it did not manifest histological abnormalities in the liver. Results indicated that up to 60% of soybean meal protein can be replaced with Roselle seed meal without affecting growth performance and nutrient utilization in catfish.

Key words: Roselle seed meal, African catfish, diets, growth response, nutrient digestibility

Introduction

Oilseed cakes and legume seeds are considered suitable as alternative dietary plant protein sources for cultivated fish and are available in sub-Saharan Africa on a large scale. Soybean meal protein is the most commonly used oilseed protein within feeds for omnivorous fish species used in African aquaculture such as tilapias (*Oreochromis* species) and African catfishes (*Clarias* species). Soybean meal is palatable and is available at a cost much below fish meal, but recently, it has become relatively expensive and scarce due to competitive demands in poultry and livestock feeding (Rumsey,1993). The use of soybean meal in fish diets is also limited by the extent to which the endogenous anti-nutritional factors are destroyed or inactivated (Lovell,1988) hence the need to evaluate alternative plant protein sources with less antinutrients or lower anti-nutritional activity. To this end, few oilseed meals, other than soybean meal, that have been evaluated as protein sources in African catfish (*Clarias gariepinus*) nutrition are limited to palm kernel meal (Saad *et al.*,1994 andNg & Chen,2002), peanut meal (Fasakin & Balogun,1996), benneseed meal (Olukunle and Falaye,1998), rapeseed and cottonseed meals (Davies *et al.*,2002).

Roselle, *Hibiscus sabdariffa* Linnaeus (Family Malvaceae), has two main varieties, of which the more important economically is *H. sabdariffa* var. *altissima* Wester and the other is *H. sabdariffa* var. *sabdariffa* Linnaeus. Both varieties are annual herbs extensively cultivated in tropical Africa, Asia, Central America and the Caribbean for the jute-like fibre or the red calyces surrounding the fruit; the basis of a popular red non-alcoholic drink, hams, jelly and colouring material for foods and beverages. Seeds of both varieties are sources of protein and lipid (Kalyane,1986; El and Khalil,1994 and Rao,1996) and are used for small-scale edible oil production by parching, soaking in water containing ashes for 3 - 4 four days, and then pounding the seeds, or by crushing and boiling. The resulting meals from this process contain low levels of antinutrients - tannins, α-amylase inhibitors, protease (chymotrypsin, trypsin) inhibitors, phytic acid, gossypol (Abu-Tarboush & Ahmed,1996; Hansawadi & Kawabata,2000). Roselle seed meal is presently sold in Nigeria at one third of the cost of soybean meal, and hence justifies investigating its use in fish feeding.

Fagbenro & Davies (2000) and Fagbenro *et al.*(2004) evaluated roselle seed meal as a partial substitute for soybean meal in diets for Nile tilapia (*Oreochromis niloticus*) fingerlings and the results showed that roselle seed meal could not replace 75% of soybean meal protein in Nile tilapia diets without affecting growth and protein utilization, because of low nutrient digestibility. This study evaluates the nutritive potential of mechanically-extracted meal derived from roselle seeds (*sabdariffa* variety) as alternative plant protein feedstuffs to mechanically-extracted soybean meal in dry practical pelleted diets for fingerlings of the African catfish, *Clarias gariepinus* (Burchell 1822).

Materials and Methods

Roselle seed meal was obtained from Arewa Oil Mills, Kano (northern Nigeria) while menhaden fish meal, soybean meal and other ingredients were obtained locally from a feed/ feedstuff market. The protein feedstuffs were separately milled, screened to fine particle size (<250μm), and triplicate samples were analysed for proximate composition according to AOAC (1990) methods. The essential amino acids were determined using an LKB 4151 Alpha$^+$ amino acid analyzer after treating the samples with 6 mol.L^{-1} HCl under reflux for 24h at 110 °C. Tryptophan was determined colorimetrically after hydrolyzing samples in 4.2 mol.L NaOH (Fishcl,1960). Protein feedstuffs were analyzed for Calcium and phosphorus contents by digesting the samples with a mixture of nitric and perchloric acids. Calcium content was measured by using flame photometry while phosphorus content was determined using a spectrophotometer. Gross energy content was determined in protein feedstuffs and diets with an adiabatic bomb calorimeter.

Apparent digestibility coefficient (ADC) of crude protein and gross energy in meals of roselle seed meal were determined as follows: a purified reference diet (containing g.kg^{-1}: casein 320, gelatin 80, α-starch 400, fish oil 100, vitamin-mineral mix 90, and chromic oxide 10) and two test feedstuff diets containing 700 g.kg^{-1} of the reference diet mixture and 300 g.kg^{-1} of roselle seed meal was prepared. *C. gariepinus* fingerlings were distributed in groups of 15 fish into 20-litre cylindrical plastic tanks supplied with aerated water. Each diet was assigned to duplicate tanks and fish were fed to apparent satiation twice daily (8.30–9.00h and 16.0–16.30h) for 14 days. On the last day, faeces were collected from each anaesthetized fish (2.5ml quinaldine.L^{-1} of water) eight hours after feeding using the dissection method. Dry matter and crude protein were analysed in triplicate samples of diets and faeces according to AOAC (1990) methods and gross energy content was determined by bomb calorimetry. Chromium content of diets and faeces was determined spectrophotometrically (Bolin *et al.*,1952). ADC of crude protein and gross energy in the diets were calculated as: $ADC = 10^2 - [10^2 \times (I_d/I_f \times N_f/N_d)]$, where: N_d = nutrient in diet, N_f = nutrient in faeces; I_d = Cr_2O_3 in diet; I_f = Cr_2O_3 in faeces. The $ADC_{crude\ protein/gross\ energy}$ in test feedstuff was calculated as: $ADC_{crude\ protein/gross\ energy} = 100/30\ (ADC_{test\ diet} - 70/100\ ADC_{reference\ diet})$.

Based on the nutrient composition of the protein feedstuffs (Table 1), a control diet and five test diets (400 g crude protein, 120 g crude lipid and 19.0 MJ gross energy.kg^{-1}dry matter) were formulated (Table 2) to meet the nutritional requirements of catfish (Wilson & Moreau,1996). The control diet (SB100) contained soybean meal protein, providing 60% of total protein while the test diets contained roselle seed meal replacing 20%, 40%, 60%, 80% or 100% of soybean component of the diet (Table 2). Lipid content was adjusted with corn and cod liver oils, while gelatinized corn starch was supplemented to adjust energy content. Chromium III oxide was added as the inert digestibility marker. The feedstuffs were blended, moistened, steam-pelleted, oven-dried for 24 h, and stored in sealed (air-dry) plastic containers at ambient temperature (32.5 °C).

Clarias gariepinus fingerlings were acclimated to experimental conditions for 14 days prior to the feeding trial. Groups of 20 catfish fingerlings (mean initial weight, 15.6±0.5g) were stocked into indoor system comprising 60-litre cylindrical plastic tanks each supplied with aerated tap water (water flow, 1.L.min^{-1}). Each diet was hand-fed to the catfish in triplicate tanks, to apparent satiation twice daily (09.00h, 16.00h) for 70 days. Fish mortality was monitored daily, fish weight in each tank was determined at two-week intervals. Growth and feed utilization indices were estimated. Water temperature and dissolved oxygen (DO) were measured daily using a combined digital YSI DO meter (YSI Model 57 YSFI; Yellow Springs, Ohio); pH was monitored weekly using a pH meter (Metler Toledo-320, Jenway, UK). Ten catfish and five catfish per treatment were respectively sacrificed at the start and end of the feeding trial and analysed for carcass composition (AOAC,1990). Livers of ten catfish per treatment were removed, weighed and used to estimate the hepatosomatic index; and later fixed in 1:10 buffered formalin solution, dehydrated in graded ethanol series, cleared with xylene and blocked in paraffin. The blocks were sectioned at 5μ, placed on glass slides, stained with haematoxylin and eosin, and examined under a light microscope.

All data obtained were subjected to one-way analysis of variance (ANOVA) test (P<0.05). Duncan's multiple-range test (Zar,1996) was applied to characterize and quantify the differences between treatments using Statgraphics 5 plus package for Windows.

Results and Discussion

$ADC_{crude\ protein}$ and $ADC_{gross\ energy}$ for soybean meal and roselle seed meal were similar (Table 1), however calculated values for digestible protein and digestible energy values of roselle seed meal were lower that the corresponding values for soybean meal. Roselle seed meal had higher fibre and ash contents than soybean meal (Table 1) and correspondingly reflected in the proximate composition of the diets (Table 2). Calculated digestible protein was similar for all diets while the estimated digestible energy of diets showed slight reductions as dietary inclusion of roselle seed meal increased. Except for apparently lower isoleucine, lysine and threonine contents, the essential amino acid (EAA) profile of roselle seed meal was similar to that of soybean meal protein (Table 3). The EAA profile of the different diets, calculated on the basis of their levels present respective protein feedstuffs (Table 3) shows that lysine content of diets RS40, RS60, RS80 and RS100 did not meet the requirements for catfish.

The ranges of the water quality parameters were: temperature 26 -28 °C, dissolved oxygen concentration 6.5-8.1 mg.L^{-1}, total ammonia 0.1-0.25 mg.L^{-1} and pH 6.8-8.2. No critical values were detected for NO_2 and NO_3. Catfish mortality was low (<10%) in all diet treatments and did not differ significantly (P>0.05). Acceptance of the diets was good and catfish became accustomed to the diets within five days. Weight gain, growth response, feed efficiency and protein utilization by catfish (Table 4) showed the best overall response in catfish fed with the control diet (SB100). Except for catfish fed with diet RS80 or RS100, no significant differences occurred in weight gain, growth response, feed conversion and protein utilization by catfish fed with control diet and other test diets (Table 4).

Hepatosomatic indices showed no trend relating to diet treatment (Table 5) and histological examination of livers of catfish fed any of the diets revealed no alterations or lesions suggestive of nutritional disorders. Carcass composition of catfish at the end of the feeding trial (Table 5) showed that catfish fed with the roselle seed meal-based diets generally had higher moisture and ash contents as well as lower protein and lipid contents compared with catfish fed the control diet. However, no clear trend was observed in relation to the dietary inclusion level of roselle seed meal.

One of the most important characteristic of feedstuffs is the bioavailability of nutrients, particularly digestible protein and digestible energy, hence reliable data on the digestibility of different ingredients for each species might well be considered as a necessary prerequisite. In spite of the high crude fibre plus ash contents of roselle seed meal, there were no differences in $ADC_{crude\ protein}$ and $ADC_{gross\ energy}$ values for roselle seed meal compared with that for soybean meal earlier reported (Fagbenro,1998). Although digestibility of nutrients are species specific, digestible protein and energy content of roselle seed meal were similar to values reported for Nile tilapia, *Oreochromis niloticus* (Fagbenro & Davies,2000).

Roselle seed meal replaced peanut meal with no adverse effects on growth of broiler chicks and laying hens (Mohammed & Idris,1991 and Backeit *et al.*,1994). It has also been included in broiler diet up to 300 g.kg^{-1} diet without lowering growth response or feed utilization (Cortes & Avila,1996; Jinez *et al.*,1998), and in Nile tilapia diets (Fagbenro and Davies,2000 and Fagbenro *et al.*,2004) roselle seed meal could not replace 75% of soybean meal without affecting growth and protein utilization, because of low nutrient digestibility. Weight gain and feed utilization by catfish decreased significantly when fed diets RS 80 and RS100, attributable to lower digestible energy caused by high dietary fibre level (Table 2). Fibre level of 100g.kg^{-1} is ideal for omnivorous fish species, above which reduced feed efficiency and digestibility have been reported (Leary *et al.*,1975 and Anderson *et al.*,1984). Higher dietary fibre could result in dilution of nutrients, thereby evoking poor growth response as has been reported for rainbow trout and tilapia (Hilton *et al.*,1983 and Shiau,1989). Poorer protein utilization in diets RS80 and RS100 was also due to lower digestible energy content of roselle seed meal (Table 1), which could indicate that more protein would have been used to produce energy. If this is true, the metabolic use of roselle seed meal at high dietary inclusion levels could be improved by increasing the digestible energy level in the diet through gelatinized starch supplements. Similarly, lower energy digestibility in palm kernel meal-based diets fed to catfishes was attributed to its high crude fibre content (Saad *et al.*,1994 and Ng & Chen,2002). Apparent digestibility coefficients and growth parameters can be used as indicators to evaluate the protein quality, but the availability and optimal balance of essential amino acids must also be considered. From the amino acid composition of protein feedstuffs (Table 3), it appears that there might be other factors in addition to energy digestibility, which reduce the nutritional value of roselle seed meal for catfish. Possible deficiencies in isoleucine and lysine contents of roselle seed meal relative to soybean meal were apparent (Table 3), which correspondingly indicated apparent lower levels of both amino acids with increasing dietary levels, particularly in diet RS80 and RS100 (Table 3). This did not compromise catfish growth and protein utilization when fed diets RS20, RS40 and RS60. Roselle seed meal contains residual amounts of antinutrients – tannins and phytic acid (Abu-Tarboush *et al.*,1997) that are characteristic of members belonging to the plant family Malvaceae. High levels of these antinutrients reduce lysine bioavailability and cause depressed appetite and loss of body weight. Toxicity was however light and did not manifest histological abnormalities in the liver.

Table 1: Proximate composition and nutrient digestibility of protein feedstuffs

g. kg^{-1} DM)	Menhanden fish meal	Soybean meal	Roselle seed meal
Dry matter	921	890	926
Crude protein	675	446	394
Crude lipid	104	46	61
Crude fibre	10	49	177
Tortal ash	204	58	114
Calcium	56.5	2.9	6.6
Phosphorus	31.6	6.8	7.0
Gross energy (MJ. kg^{-1})	18.69	17.78	17.91
ADC crude protein (%)	92.8[1]	86.9[1]	86.6
ADC gross energy (%)	85.1[1]	77.4[1]	74.9
Digestible protein (g.kg^{-1})	626.4	387.6	342.1
Giestible energy (MJ. kg^{-1})	15.91	13.76	13.41

Values of ADC previously determined by Fgbenro (1998)   DM, Dry Matter

Table 2: Ingredient and proximate composition of catfish diets

Ingredients (g. Kg^{-1} diet)	SB100	RS20	RS40	RS60	RS80	RS100
Menhanden fish meal	300	300	300	300	300	300
Soybean meal	450	360	270	180	90	0
Roselle seed meal	0	102	204	306	408	510
Cod liver oil	30	30	30	30	30	30
Corn oil	40	38	36	34	32	30
Cassava starch	160	150	140	130	120	110
Carboxymethyl cellulose	20	20	20	20	20	20
Proximate composition (g. kg^{-1} DM)						
Crude protein	403.2	403.3	403.3	403.3	403.4	403.4
Crude lipid	121.9	122.0	122.1	122.2	122.2	122.3
Crude fibre	45.1	58.7	72.3	86.0	99.6	113.3
Total ash	87.3	93.7	100.1	106.5	112.9	119.3
Gross energy	19.13	19.11	19.08	19.06	19.03	19.00
Digestible protein1	362.4	362.4	362.4	362.4	362.4	362.4
Digestible energy1	16.26	16.15	16.04	15.93	15.82	15.71

1Calculated from digestible protein and energy values of feedstuffs in Table 1 DM, dry matter

Table 3: Essential amino acid composition (g.kg^{-1} protein) of the protein feedstuffs and experimental diets

	Protein feedstuffs			Diets						
	MFM1	SBM2	RSM3	SB100	RS20	RS40	RS60	RS80	RS100	Catfish requirements[4]
Arginine	61	71	96	50.3	53.7	57.1	60.5	63.9	67.3	43
Histidine	24	26	27	18.9	19.3	19.7	20.1	20.6	21.0	15
Isoleucine	47	61	47	41.6	40.9	40.1	39.5	38.8	38.1	26
Leucine	73	86	80	60.6	61.0	61.4	61.9	62.3	62.7	35
Lysine	77	65	49	52.4	51.5	50.6	49.8	48.9	48.1	51
Methionine	29	15	16	15.5	15.7	16.0	16.3	16.6	16.9	23*
Cystine[5]	9	13	21	8.6	9.5	10.5	11.5	12.4	13.4	
Phenylalanine	40	53	51	35.9	36.3	36.7	37.1	37.6	38.0	50**
Tyrosine[5]	32	39	36	27.2	27.3	27.4	27.6	27.8	28.0	
Threonine	41	40	34	30.3	30.2	30.0	29.9	29.8	29.6	20
Tryptophan	11	14	13	9.6	9.7	9.7	9.8	9.9	9.9	5
Valine	53	53	50	39.8	40.1	40.4	40.7	41.1	41.4	30

[1]Menhanden fish meal   [2]Soybean meal   [3]roselle seed meal   [4]Wilson and Moreau (1996)
[5]Non-essential amino acids   *Methionine + cystine   **Phenylalanine + tyrosine

Table 4: Growth, feed utilization and digestibility of catfish after 70 days of feeding experimental diets

	Diets						
	SB100	RS20	RS40	RS60	RS80	RS100	S.E.[6]
Initial weight (g)	15.6	15.6	15.6	15.6	15.6	15.6	2.01
Final weight (g)	58.2a	57.4a	56.9a	55.7a	48.1c	48.1c	3.98
WG1	273.1a	267.9a	264.7a	257.1a	235.3b	208.3c	
SGR2	1.88a	1.86a	1.85a	1.82a	1.73b	1.61c	0.05
FGR3	1.83a	1.85a	1.88a	1.92a	1.99b	2.07b	0.17
PER4	1.84a	1.82a	1.79a	1.76ab	1.70b	1.62c	0.26
PPV5	30.1a	29.5a	29.1a	27.4ab	24.8b	20.1bc	3.65
Survival (%)	93.3	96.7	96.7	96.7	93.3	96.7	

[1]weight gain (%) = [{final wt. - initial wt.)/initial wt.)] x 100
[2]specific growth rate (% day^{-1}) = [(ln final wt. - ln initial wt.)/ no of days] x 100
[3]feed gain ratio = feed intake (g)/body weight gain (g) [4]protein efficiency ratio=body wt. Gain (g)/protein intake (g)
[5]protein productive value (%) = protein gain (g)/protein intake (g)] x 100
[6]Standard error of treatment mean values in a row followed by dissimilar letters are significantly different (P<0.05)

Table 5: Carcass composition (% wet weight basis) and hepatosomatic index of catfish

	Diet					
	SB100	RS20	RS40	RS60	RS80	RS100
Moisture	79.16	76.50	76.63	76.66	76.63	76.64
Crude protein	15.88	17.45	17.39	17.28	17.28	17.32
Crude lipid	5.24	6.43	6.38	6.37	6.37	6.35
Total ash	1.91	1.29	1.36	1.34	1.34	1.36
Hepatosomatic index[1]	1.50	1.91	1.89	1.92	1.92	1.90

[1]hepatosomatic index (%) = (liver wt./body wt.) X 100

The presence of antinutrients in oilseeds and legumes, coupled with deficiencies in some essential amino acids (or their availability at sub-optimal levels, have put limitations on their use in replacing soybean meal completely (Fagbenro and Davies, 2000). Considering the absence of any histological change in catfish liver and its economic advantage in the cost of catfish nutrition, roselle seed meal may be recommended as an alternative to soybean meal as a protein feedstuff, provided it does not replace > 60% of soybean meal. High crude fibre content will almost certainly limit the use beyond this level in catfish diets.

Acknowledgement

I am grateful to the International Foundation for Science (IFS), Stockholm for providing funds for this study through an IFS/ DANIDA (Danish International Development Agency) award.

References

Abu-Tarboush, H. M. and S. A. B. Ahmed, 1996. Studies on karkade (*Hibiscus sabdariffa*): Protease inhibitors, phytate, in vitro protein digestibility and gossypol content. Food Chemistry, 56: 15-19.

Abu-Tarboush, H. M., S. A. B. Ahmed and H. A. Al Kahtani, 1997. Some nutritional and functional properties of Karkade (*Hibiscus sabdariffa*) seed products. Cereal Chemistry, 74: 352-355.

Anderson, J., A. Jackson, A. Matty and B. Capper, 1984. Effects of dietary carbohydrate and fibre on the tilapia *Oreochromis niloticus* (Linn.). Aquaculture, 37: 303-314.

Association of Official Analytical Chemists (AOAC), 1990. Official method of analysis. K. Helrich (ed), 15th edition, AOAC, Arlington, VA, pp: 684.

Backeit, M. H., E. A. El Zubeir and S. K. Jubarah, 1994. The nutritional value of Roselle seed meal in laying hen diets. J. Science of Food and Agriculture 65: 199-200.

Bolin, D. W., R. P. King and E. W. Klosterman, 1952. A simplified method for the determination of chromic oxide ($Cr_2 0_3$) when used as an index substance. Science, 16: 634-635.

Cortes, C. A. and G. E. Avila, 1996. Use of roselle seed (*Hibiscus sabdariffa*) in broiler diet. Veterinaria Mexico, 27: 205-209.

Davies, S. J., A. Gouveia and A. A. Tekinay, 2002. Rapeseed and cottonseed meals as component ingredients in diets for fingerling African catfish, Clarias gariepinus. Pp.121, in: Abstract Book, Feeding for quality: 10th International Symposium on Nutrition and Feeding in Fish, Rhodes, Greece.

El, A. T. A. and A. H. Khalil, 1994. Characteristics of roselle seeds as a new source of protein and lipid. J. Agriculture and Food Chemistry 42: 1896-1900.

Fagbenro, O. A., 1998. Apparent digestibility of oilseed cakes/meals by African catfish *Clarias gariepinus*. Aquaculture International, 6: 317-322.

Fagbenro, O. A. and S. J. Davies, 2000. Use of oilseed meals as fishmeal replacer in tilapia diets. In: K. Fitzsimmons, K., Filho, J. C. (editors.) Proceedings of Fifth International Symposium on Tilapia in Aquaculture (ISTA V), Rio de Janeiro, Brazil, pp: 145-153.

Fagbenro, O. A., T. T. Akande, O. O. Fapohunda and Y. Akegbejo-Samsons, 2004. Comparative assessment of roselle (*Hibiscus sabdariffa* var. *sabdariffa*) seed meal and kenaf (*Hibiscus sabdariffa* var. *altissima*) seed meal as replacement for soybean meal in practical diets for fingerlings of Nile tilapia, *Oreochromis niloticus*. Proceedings of the Sixth International Symposium on Tilapia in Aquaculture (ISTA 6), pp: 277-288. (K. Fitzsimmons & R.B. Bolivar, editors). Manila, Philippines.

Fasakin, E. A. and A. M. Balogun, 1996. Replacement of groundnut cake with processed soybean meals in diets for the African mud catfish, *Clarias gariepinus*. Applied Tropical Agriculture, 1: 5-11.

Fischl, J., 1960. Quantitative colorimetric determination of tryptophan. J. Biological Chemistry, 235: 999-1001.

Hansawasdi, C. and J. Kawabata, 2000. alpha-Amylase inhibitors from Roselle (*Hibiscus sabdariffa* Linn.) tea. Bioscience, Biotechnology and Biochemistry 64: 1041-1043.

Hilton, J. W., J. L. Atkinson and S. J. Slinger, 1983. Effect of increased dietary fibre on the growth of rainbow trout (*Salmo gairdneri*). Canadian J. Fisheries and Aquatic Sci., 40: 81-85.

Hossain, M. A. and K. Jauncey, 1989. Nutritional evaluation of some Bangladeshi oilseed meals as partial substitutes for fish meal in the diets of common carp, Cyprinus Carpio L. Aquaculture and Fisheries Management, 20: 255-268.

Jínez, T. M., A. C. Cortés, E. G. Ávila, T. M. Casaubon and R. E. Salcedo, 1998. Efecto de niveles elevados de semilla de jamaica (*Hibiscus sabdariffa*) en dietas para pollos sobre el comportamiento productivo y funcionamiento hepático. Veterinaria Mexico 29: 35-40.

Kalyane, V. L., 1986. Nutritional quality of Hibiscus seed protein. International J. Tropical Agriculture, 4: 280-282.

Leary, D. F. and R. T. Lovell, 1975. Value of fibre in production-type diets for channel catfish. Transactions of the American Fisheries Society, 104: 328-332.

Lovell, R. T., 1988. Use of soybean products in diets for aquaculture species. J. Aquatic Production, 2: 27-52.

Mohammed, T. A. and A. A. Idris, 1991. Nutritive value of Roselle seed (*Hibiscus sabdariffa*) meal for broiler chicks. World Reviews in Animal Production : 59-62.

Ng, W. K. and M.L. Chen, 2002. Replacement of soybean meal with palm kernel meal in practical diets for hybrid Asian-African catfish, *Clarias macrocephalus* x *C. gariepinus*. J. Applied Aquaculture 12: 67-76.

Rao, P. U., 1996. Nutrient composition and biological evaluation of mesta (*Hibiscus sabdariffa*) seeds. Plant Foods in Human Nutrition, 49: 27-34.

Rumsey, G. L., 1993. Fish meal and alternative sources of protein. Fisheries, 18: 14-19.

Saad, C. R., S. H. Cheah and M. A. A. Hashimi, 1994. The use of palm kernel cake (PKC) in catfish practical diets. In: Chou, L.M. et al. (eds) Proceedings of The Third Asian Fisheries Forum, Asian Fisheries Society, Manila, Philippines, pp: 653-655.

Shiau, S. Y., 1989. Role of fiber in fish feed. In: Shiau, S-Y. (ed) Progress in Fish Nutrition: Proceedings of the Fish Nutrition Symposium, Marine Food Science Series No. 9, National Taiwan Ocean University, Keelung, Taiwan, pp: 93-119.

Wilson, R. P. and Y. Moreau, 1996. Nutrient requirements of catfishes (Siluroidei). Aquatic Living Resources, 9: 103-111.

Zar, J. H., 1996. Biostatistical analysis. 3rd edition., Prentice-Hall, Upper Saddle River, NJ, pp: 662.