# Raw Ingredients for Marine Aquaculture Fish

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## ABSTRACT

Raw ingredients, apparently routine topic and a risk for a recycling of data, are in reality becoming more and more important. It brings many additional constraints to the formulator. Just because consumers protection against dioxine, BSE, immunostimulants, or projection to 2010 concerning fishmeal utilization in aquaculture, will lead to eliminate some products or to replace others. Meat and bone meal, fishmeal and fish oil are particularly concerned. So aside from conventional protein sources, many ingredients especially from plant origin are tested and it opens up a new era for nutritional research. A short review of common ingredients for shrimp and fish brings new recommendations, evidences old papers, triggers the feed manufacturer, makes appear new set of additives (amino acids, attractants, concentrates, pigments,…) to face challenges of fast growing fish and shrimp in different culture systems. Biosecurity measures add up some considerations on the pertinence of recycling aquaculture by-products. Then from a routine topic, it comes up a fascinating one, which combines contribution from farmers, from manufacturers, scientists, and in particular nutritionists, all tended to make the best combination at a minimum price. Outlines of the paper were set to sum up 5 years research on sea bass culture (Aquacop, 1989-94) and draw a comparative approach (sea bass, snook, red drum) from an ADC point of view, optimum P/E at different stage of development : larvae, weaning, fingerlings at nursery, juveniles and grow-out making emphasis on animal *vs* plant protein : soybean meal with nutritional aspects and appetibility, fishmeal and fish oil criteria to end up with breeders requirement and feeding.

## INTRODUCTION

Raw materials are presented very regularly in order to update information on such basic knowledge, which is so important. Importance goes to the formulator of course as well as to the researcher. For that reason, a brief review on ingredients, its composition, but also origin, essentiality, relation with aspects such as succulence of the diet, environment concerns (in case of fishmeal for example), immune response, is certainly worth to be examined. Nutritionists, formulators, scientists are all mainly concerned to collect data for improvement of the current knowledge along such wide topic. Raw materials are basically

Cuzon, G., Gaxiola, G., Garcia, T. , Sanchez, A., Aquacop., 2002. Raw ingredients for marine aquaculture fish. In: Cruz-Suárez, L. E., Ricque-Marie, D., Tapia-Salazar, M., Gaxiola-Cortés, M. G., Simoes, N. (Eds.). Avances en Nutrición Acuícola VI. Memorias del VI Simposium Internacional de Nutrición Acuícola. 3 al 6 de Septiembre del 2002. Cancún, Quintana Roo, México.

the same for all species but can be selected differently according to development stage. Ingredients particles were emphasized again for shrimp feed (Obaldo *et al.*, 1998; 2000) as well as technology (extrusion) to improve nutritive value; technology is compared for shrimp pellets with and without heat (Rout, 1999).

In fish juveniles, Anon (1999) examined sea-bream performances; red drum and MCT which are fatty acids 8-12 carbon atoms gave indication on the fat deposition and its limitation through MCT (Davis *et al.*, 1999); feeding stimulants were examined for flounders (Fredette *et al.*, 2000), soybean products as a medicament for salmon (Kroglähl *et al.*, 2000) and copra was used for *Labeo rohita* fingerlings in spite of its high cellulose content (Mukhopadhgay, 1999); yellowtail could be fed on practical diet (Shimeno *et al.*, 1999) but steatosis should be avoided (Spiris *et al.*,1998) using choline-methionine as a lipotropic factor. ADC of ingredients for salmonids is still of importance (Sugiuro *et al.*, 1998) and minerals are surveyed. Antinutritional factor from soybean affected puffer fish (Takii *et al.*, 2001). Thoman *et al.*(1999), Watanabe *et al.* (2000) evaluated protein energy ratio in red drum and yellowtail; in yellowtail, Watanabe *et al.* (1998) examined the possibility of non-fish meal diets. Glencross *et al.* (1999) experimented a feed for tuna; Hardy (1999) investigated grains by-products for fish feeds; in *Sparus* soy and rapeseed could replace fish meal (Kissil *et al.*, 2000).

These are some of available information concerning raw materials for different aquaculture species and different development stages to limit live food sequence and to replace ingredients such as fish meal to a certain extent. Rainbow trout faced a growth reduction at 50% soy flour replacing fishmeal (Kaushik *et al.*, 1995) but poultry by-product meal is suitable as a partial or complete replacement of fishmeal if LYS and MET are added (Steffens, 1994).Different processing treatments on soybean meal revealed that full-fat extruded soybean meal did not improve its nutritional value and 20% dietary protein replacement of brown fish meal was achieved with rainbow trout (Oliva-Teles *et al.*, 1994).In catfish, heat processed winged bean as partial replacement(80%) menhaden fish meal proved satisfactory (Fagbenro, 1999) and distillers grains with LYS replaced soybean meal up to 40% (in channel catfish (Webster *et al.*, 1991). Sea bream presented similar growth and feed efficiency when fed soybean meal or chicken offal meal at 25% substitution level with fish meal (El-Sayed, 1994). What are the other considerations which would lead to an improved ability to use raw materials in order to cope with nutritional, economical, and environmental constraints?

## DEVELOPMENT

***Fish nutrition work in Tahiti. A 5 years experience:*** *Lates calcarifer* (sea bass) is an easy fish to cultivate compared to other finfish species (*C. undecimalis…)* because larvae can be produced, mass produced in hatchery, weaning is short and consistent, juveniles grow well in tanks or floating cages or ponds, up to commercial size (4 to 300g in 4 months at 28°C) and then it is possible to keep some specimen for breeders.. Breeders kept in round tanks can be handled, injected and spawned regularly. Then, closing the life cycle in captivity is quite achievable and farms installed in French Polynesia produce today, few 10 m tons of marketable fish. Only limitation would reside in *nodavirus* present at larval stage up to $J_{25}$ and sometimes a parasite (*Cryptocaryon sp*; but this is not impairing a production for local market (<100mt/y).

It is an example where nutritional requirements , some of them, were approached using practical diets right from the beginning in 1986 and regular raw materials (fishmeal, soybean meal, wheat) were the main protein and carbohydrate source for juveniles , fingerlings and breeders.

Fish are raised in 10m3 tanks, and maturation in captivity all year round is achieved under environmental control (T°C and photoperiod) in order to get 1-7 million eggs /month/pond. Larval rearing and fry production in hatcheries, survival rates 30% after day 25 and food sequence with rotifers and *Artemia* at 10-20 larvae/l in 10 m3 tank in semi-intensive conditions or extensive conditions at 2-5 larvae/l in 500m2 earthen ponds. Weaning and nursing on artificial diets bring 50% survival after 2 months and artificial feeds placed on automatic feeders help to get a final weight of 1.5 to 2 g fry. Grow-out in cages or earthen ponds would be conducted with expected 90% survival and final mean weight of 350-400g after 3 months.

A brief comparison with *Centropomus undecimalis* of America: Barramundi (sea bass) have been found easier to spawn voluntarily or induced tank spawning, faster developing (25 *vs* 35 days, and faster growing (545 g within 9-11 vs 11-12 months) than *Centropomus undecimalis* (snook) according to Tucker *et al.* (2001).

Red drum can be raised from eggs to fingerlings size in fiberglass tanks and seawater with rotifers and *Artemia*: in Martinique (Soletchnik *et al.*, 1989) rearing larvae at 20-200 larvae/liter with a survival up to 35-65% for the first and second month was achieved; mortality could come from nutritional deficiency of *Artemia* strains out of any enrichment. Red drum adapt well to captivity and is very prolific (Holt, 2001), many work has been conducted on its nutritional requirement and recently it was investigated a replacement of fish meal (Davis & Arnold, 2002) as discussed in this paper.

**Larvae**

In larvae, aside from the live food sequence, Teshima *et al.* (2000) reviewed nutritional value of micro particles. Microencapsulated diets were tested on striped bass (Chu, 1999); Garcia Ortega (2001) evaluated with micro bound diets protein levels for fish larvae; a diet experienced for halibut larvae (Hamre *et al.*, 2001) and *Artemia* was needed to achieve success at metamorphosis (Gera *et al.*, 1998). Spray dried diets for *M. rosenbergii* and striped bass larvae were tested (Ohs *et al.*, 1998) while micro bound MBD diet worked in sea bass larvae (Partridge, 1999).

In sea bass, larval rearing and fry production in hatcheries provided survival rates around 30% after $day_{25}$ while providing food sequence with rotifers and *Artemia* at 10-20 larvae/l in 10 $m^3$ tank (semi-intensive conditions) or extensive conditions at 2-5 larvae/l ($500m^2$

earthen ponds). Rotifers density of 10-20 pieces/ml was required; *Tetraselmis* or *Chlorella* should be added daily to maintain 8-10x$10^3$ or 2-4 x$10^4$ cells/ml respectively: the algae serve a dual purpose as a direct food to rotifer and a water conditioner in the rearing tank. *Artemia* were used to feed larvae together with rotifers from age 8 days. (Tookwinas, 1989).Clear water technique derived from European sea bass and sea bream larvae rearing was adapted to tropical sea bass. Enriched preys were more nutritious than rotifers grown on algae and yeast. Rotifers concentration started at 1 per ml to increase at 10-15/ml (Duray *et al.*, 1986) then 20/ml (Rimmer *et al.*, 1989). Survival rates ranged 50-60% on day 25. Sea bass larvae can accept dry particles after a few days; survival and growth after 20 days are satisfactory compared to control fish on live *Artemia.* A suitable feeding program included: from 20mg larvae, live *Artemia* up to day12 (5g/5400 larvae), micro particles ACAL (Aquacop, 1983) up to day10 (5-12 g feed weight / 5400 larvae) and sevbar, granulated feed from day7 onwards, 5-50g feed weight / 5400larvae (Aquacop, com pers.). Only limitation with sea bass at larval stage up to $J_{25}$ would reside in presence and virulence of *nodavirus*.

Snook references on larval culture indicate a need for redefinition of culture technique to increase survival from 1.2% to 7% with probiotic and at least 10-15% survival rate if an effective control of cannibalism is achieved (Tucker *et al.*, 2001). Lipid content in live preys or dry particles is like other marine species, essential, not only in terms of triglycerides amount: the n-3 HUFA's are required during the first feeding too (Seiffert *et al.*, 2001). In larvae, rotifer enriched with superselco showed highest levels of EPA (13mg/g dwt) and DHA (6mg/g dwt) with a survival rate still low at 2-3% (Seiffert *et al.*, 2001).

Red drum larvae fed on artificial diets (Holt *et al.*, 1991) evidenced an absence of a well developed digestion system and lipid dietary supply is of prime interest to mimic the natural food and its lipid content (30-50% dwt).

As far as lipid nutrition (Izquierdo *et al.*, 2000) is concerned, larvae received unsaturated fatty acids(HUFA's) from marine oils; but the hydrolysis of triglycerides with non-specific lipase lead to micelle formation and such micelles contain phospholipids. In enterocytes the reformation of glycerides and phospholipids contribute to synthesis of VLDL and transport to different tissues. There is a need for phospholipids (PC and PE noticeably) for the constitution of bio membranes while larvae is presenting a high tissue growth. There is a complex timing from a feeding point of view consisting in providing HUFA's and phospholipids according to ontogeny of digestive enzymes in order to supply triglycerides and lecithin in proportion for optimum absorption, reformation of key molecules to ensure adequate membrane structure. There are metabolic enzymes such as desaturase, phosphorylase, regulating the flow of nutrients and bringing to tissues what is essential for growth and adaptation to changes in environmental conditions.

## Weaning

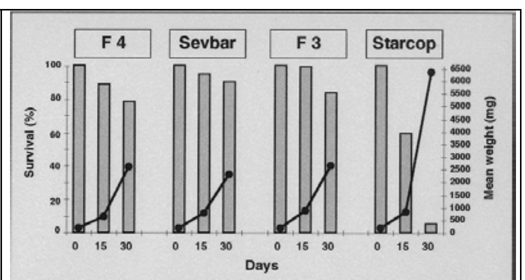
In order to facilitate digestive enzymes adaptation to a new diet (Peres *et al.*, 1996), larvae are fed with an overlapping feeding schedule during a few days with a mixture of frozen *Artemia* and micro particles (MBD,..) or extruded feeds (56/16, sevbar) . The incorporation of 19 and 38% hydrolysate (CPSP [70] ) in larvae diets (day 10-41) induced a high level of two membranous enzymes of intestine, alkaline phosphatase and amino peptidase N as early as day 20. The cytosolic enzyme leucine-alanine peptidase (leu-ala) was also assayed (Cahu *et al.*, 1999).

In a comparison between several diets (Table 1), weaning of larvae and further growth feeds without sorting. Survival (%) and mean weight (mg). Feed and carcass composition are given for protein and energy (MJ/kg DE).

The results were similar in terms of survival (80-100%) and growth rates were not different whichever diet considered while carcass composition did not show significant difference even with the treatment (star cop) while the survival dropped at day30 (Fig in table1) .this experiment was conducted without sorting of animals; grading of larvae is recommended before weaning This operation is conducted generally to refrain cannibalistic tendency of fingerlings (Cuzon *et al.*, 1990).

Table.1 nursing under experimental conditions of fingerlings sea bass fed extruded

| | moist | prot | lipid | energy | ash |
|---|---|---|---|---|---|
| F4diet | 15 | 49 | 11 | 17 | 15 |
| sevbar | 12 | 56 | 11 | 18 | 12 |
| F3 diet | 12 | 47 | 13 | 17 | 12 |
| starcop | 10 | 52 | 15 | 19 | 10 |
| carcass | 1 | 65 | 15 | 20 | 17 |
| carcass | 3 | 62 | 17 | 20 | 15 |
| carcass | 1 | 59 | 19 | 20 | 16 |
| carcass | 1 | 65 | 15 | 20 | 17 |




Snook will begin to eat dry feed within 35 days after hatching and then raised on pellets in small or large tanks (Tucker, 1987).

## Fingerlings

At nursery stage (Table 2), optimum protein level is determined using a commercial diet (sevbar).

Table 2.nursery stages of sea bass fingerlings fed on dry feed (sevbar or 56/16)

| age | cycle | growth | survival | FCR | final load |
|---|---|---|---|---|---|
| $J_{75}$ to $J_{140}$ | 2 m. | 2 to 30 g | >60 % | <1.5 :1 | 18 kg/$m^3$ |
| $J_{80}$ to $J_{142}$ | 2 m. | 2 to 29 g | 90 % | 1.3 :1 | 11 kg/$m^3$ |

High quality protein diets can promote growth rates of about 6g/15days provided that protein content range between 41-50% and similar conclusions were drawn on snook (Tucker, 1987) with a starter feed which should contain at least 50% protein

## Juveniles

Red drum amino acid nutrition (Moon & Gatlin, 1990) provided information on limiting factor. Requirements of red drum were described (Robinson, 1991). In intensive recirculating systems, red drum feeding trials were reported (Gatlin *et al.*, 1996), with a recent description on its culture (Holt, 2001); in clear water, feeding of cultured sea bass (Williams *et al.*, 1994) were conducted by replacing fishmeal with greaves meal (Aquacop, 1993) and amino acid requirement in sea bass described to be correlated with muscle amino acid composition. But first and before all, research on sea bass focused on ADC and optimum protein energy determination.

**ADC:** Digestibility values (Table 5) on ingredients for marine fish are a prerequisite and with *L.calcarifer*, a cooperative study Asian / UE project, 1994) gave consistent data for the species and considering fishmeal as a major component. In a range of diets from 38-60% protein content, ADC protein were found at 95%, energy at 79-87% lipid at 95-98% and DP/DE in mg protein /MJ ranged from 18-27.

Table 5. Juveniles ADC nutrient and energy for some ingredients

| | sea bass | red drum | snook |
|---|---|---|---|
| $ADC_{protein}$ fishmeal | 90 | 74-100 | - |
| $ADC_{protein}$ soybean meal | 85 | - | - |
| $ADC_{starch}$ | 55 | - | - |
| $ADC_{lipid}$ | 72-83 | - | - |
| $ADC_{energy}$ | - | 56-60 | - |

Red drum (*Sxianops ocellatus*) were found to digest protein from different sources (blood meal, corn grain, cottonseed meal, sorghum, meat, menhaden fish meal, rice bran, soybean meal and wheat mids in a range of 74-100% with a relation between high values and high (>60%)protein content (McGoogan & Reigh,1996). ADC energy was low for animal products (54-60%) and lower for plant products (12-52%). Digestive enzymes (Table 6) were found to vary with the feeding frequency in snook (Gaxiola *et al.*, 2000) and the peak of proteinases increased with residence time of feed in the gut leading to a maximum digestion of dietary protein (Garcia *et al.*, 2000). All species presented a good aptitude to digest fishmeal based diets containing 420-500 g $kg^{-1}$ dietary protein.

Table 6. Juveniles feed performances and utilization *data for larvae (Walford *et al.*, 1991)

| | sea bass | red drum | snook |
|---|---|---|---|
| feeding frequency | continuous | 2 | 3 |
| gastric evacuation | 1-2 hour* | | 4 hours |
| FCR | 1 :1 | | 0.9 :1 |
| appetability | | | |
| FM | ++ | ++ | ++ |
| SBM | - | - | - |

**Optimum protein and Energy**: fishmeal remains the basic ingredient when examining the requirement (Lee *et al.*, 1994) and results (Fig. 1) gave an expression of sea bass requirements not only in percentage but in quantity per 100g of juvenile per day. Improvement in previous recommendations led to a sparing effect on protein; however, these recommendations whose acquisition was made under laboratory conditions were needed to be confirmed on large fish raised in field conditions i.e. for example in floating cages.

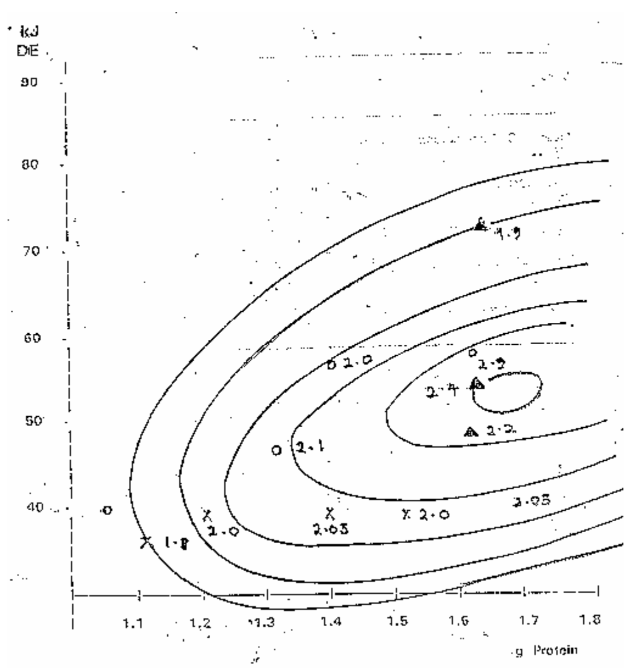



Fig.1. Contour lines for SGR plotted according to g protein/ MJ DE for juveniles *Lates calcarifer.*

Growth performances were best with the diet containing 43% protein and 17MJ/kg when fish were fed at a feeding rate of 4.1% per day.

SGR at 2.4% gave a requirement for maximum growth which was 1.5 g balanced protein(DP) and 64 MJ DE (Fig.1) and similar approach on FCR provided another value: 1.3g (Table 3) while body lipid content and DP g was not addressed yet (Lee *et al.*, 1994).

Table 3. Juveniles sea bass performances according to DP and DE

| | DP g | DE kJ |
|---|---|---|
| SGR | 1.5-1.7 | 55 |
| feed efficiency | 1.3 | 60 |
| body lipid content | - | - |

But, if feed efficiency is taken into account, DP is lower (1.3 g) and DE =60kJ then it could be looked into body lipid content and optimum DP and DE.

Juveniles red drum diets should contain 35-45 % high –quality protein and 15-17 kJ/g energy with 5-6% fat and below 12 % to prevent depressed weight gain (Chamberlain *et al.*, 1990).

## CRITERIA FOR FISHMEAL TO BE USED IN FISH RATION

As it is the main protein source, there is a constant need for definition of its quality. Quality can vary considerably from one origin to another. In order to get a quality fishmeal (FM), it should have been dried at a temperature which does not exceed 90°C, considering that 70°C is ideal. Its moisture content should be below 10% for storage reasons. Protein content should be around 70% and ash content is expected staying below 15% because for example phosphorus availability is 50% with FM or meat and bone meal, and it is a better source with mineral mix including dicalphos (80% phosphorus availability). Then there is another prerequisite for regularity in composition. Such regularity was not always achieved and that is why some manufacturers in Europe practiced the blending of 4 or 5 different fishmeal and then coping with one or two compounds variation without altering the overall composition of the final mix product. Common sense advice is sometimes given through the assertion "talk with your fishmeal supplier".

**- wastes** use is an alternative to produce fish at local level with a possibility to recycle fish offal and then decrease the possible pollution risk in an island area for example, where environment need to be preserved.

- other animal protein sources (meat and bone meal, poultry meal; feather meal, whey, ….

## CRITERIA FOR FISH OIL TO BE USED IN FISH RATIONS

There are several criteria to be looked into such as free fatty acids (<3%), unsaponifiable fraction (<2.5%) , POV < 5 meqv /kg, iodine value >135, n-3 fatty acids above 15% with EPA and 8% for DHA, and ethoxyquin at 500ppm. Moreover, polyunsaturated fatty acids level in the diet will condition the level of vitamin E to be added to the diet with a range between 20 to 50 mg/kg according to 1% to 10% PUFA's respectively.

Fish oil is used to meet essential fatty acid requirements of juvenile red drum or sea bass. As far as 0.5-1% n-3 HUFA (0.3-0.6%EPA+DHA) are provided to juveniles red drum , best growth, survival and feed efficiency were observed (Lochman & Gatlin, 1993). In experimental diets, dietary fish oil was utilized up to 13% for red drum better than soybean oil (Tucker *et al.*,1997) and fish were fatter with 13% dietary lipid than with 2% moreover, quantitative EFA (linolenic acid) requirement will increase with dietary lipid (Lochman & Gatlin, 1993); semi purified diets would contain 7% total lipid.

**Animal protein versus plan protein**: an outdated distingue: end of 50's nutritional value of animal protein were considered higher than plant protein; today, they have a higher ADC, a higher BV and APF (animal protein factor) but generalization is a fake, exceptions exist and APF is known to be a combination of $B_{12}$ + carnitine + attractants. Most difference does not refer to protein itself, but to factors such as anti-nutritional factors and cellulose in plant sources, vitamins of animal raw materials, attractants and succulence. It is a matter of thinking in terms of protein of a given characteristic, plant or animal raw materials source.

## PLANT *VS* ANIMAL PROTEIN: EXAMPLE OF SOYBEAN MEAL (SBM)

Soybean meal used in sea bass studies (10%moisture, 47.5% crude protein, 3.4 % crude fiber, 1.9% crude fat, 6% ash) contained 1.9 mg/g antigens such as glycinin and β-conglycinin with 14 and 8 wells of inhibition by hemagglutination testing); (Central soya, pers.com.): while trypsin inhibition was low, the antigens were quite high in this sample. This conventional protein source is used to try and replace fishmeal, and if inclusion level tends to increase the loss of succulence of the diet put formulators in front of an alternative which is to combine soy concentrate (procon) and fish soluble, $CPSP^{80}$ for example. Moreover, when lower than 15% of the ration in animal protein (fishmeal), a trace supplement (Mn, I, Cu, Zn Fe, Co) is recommended with an attention to phosphorus, availability of which is only 40% compared to 80% with dicalphos (Lovell, 1979). Ash content was examined recently in *L. calcarifer* diet (Chaimongkol and Boonyaratpalin, 2001) and low ash-diets (10%) provided higher Ca, Mg, P and Zn retention than high-ash diets (22%). Concerning Ca, there is a possible interference with direct absorption derived from water: for example, with goldfish, [between 0.2 and 0.8 % Ca in diet], Ca derived from water (i) decreases as concentration in food increases (ii) increases as concentration in water increases from 4 to 20mg Ca/l water. Marine fish are sometimes raised in waters from 35ppt down to 12ppt salinity or even less in case of sea bass and then consideration in consideration in calcium load should be taken accordingly. Sea bass and related species are examined form a comparative point of view, more generally for some of their requirements and data summed up in Table 4. A comparison is made from a qualitative and quantitative point of view for protein, lipid, starch, and mineral (phosphorus) but the level of acquisition forms our knowledge and our experience is more detailed with sea bass rather than with the two other species considered and more data will certainly benefit to the comparative and fruitful approach.

Table 4. Comparative values for some nutritional requirements in juvenile fish, (* on RT basis)

| | seabass | red drum | snook |
|---|---|---|---|
| relative % protein | 48 | 35 | 42 |
| DP g/100g fish/d | 1.5-1.7 | - | - |
| DE / 100g body mass | 55 kJ | | |
| crude lipid % | 6-10-13 | 2-13 | 12 |
| 18 :3n-3 % in the diet | 2-3 | 0.8-1.7 | - |
| 20 :5n-3 % in the diet | 0.3 | 0.3-0.67 | - |
| 22 :6n-3 % in the diet | 0.3 | | - |
| starch % | 12 | - | 6-24 |
| *ADC protein* | | | |
| fishmeal | 90 | 74-100 | - |
| soybean meal | 85 | | - |
| *ADC energy* | | 54-60 | |
| available phosphorus | 0.9 | - | 0.6 |
| calcium | - | - | - |
| methionin g % g diet | 0.8* | 1.0 | 1.0 |
| DP/DE mg/kJ | 26 mg | 23-27 | |
| proteinases | - | - | 10 |

An example of FM replacement is given (Fig.2) with a possibility to reach 30% inclusion of soybean meal (diet B with 23% SBM or 30% replacement of fishmeal) without significant reduction in growth performances (100-110g) of sea bass after 3 months trial; however, a trend is observed of a progressive reduction in weight gain (100 down to 90g) following a reduction in fishmeal content. Interestingly, feed intake maintained at 3% /day for the 3 diets at the difference with other species (red drum, rainbow trout, and striped bass with up top 75% replacement of FM protein with soybean protein.

Given the limited supply of fishmeal, a replacement with soybean meal for red drum juveniles (Davis & Arnold, 2002) reduced palatability of the feed. Provided that 20% poultry by-products meal was present, fishmeal was reduced to 5% of the diet and replacing it with solvent extracted soybean meal. Similar approach was reported on trout (Cho, 1992; pers. com.) and a combination of soy concentrate and CPSP$^{80}$ would overturn he problem of succulence of the diet in absence of regular fishmeal. On a 34% CP diet with protein supplied by soybean meal and fishmeal, diets containing only soybean meal protein were poorly consumed by red drum (Reigh & Ellis, 1992) which lost weight.

| | A | B | C | F |
|---|---|---|---|---|
| dry matter | 92 | 92 | 92 | 90 |
| CP | 49 | 49 | 49 | 49 |
| DP | **44** | **44** | **44** | **44** |
| CL | 10 | 10 | 11 | 11 |
| cbh | 20 | 15 | 14 | 5 |
| ash | 9 | 9 | 9 | 13 |
| DE MJ/kg | **15** | **15** | **15** | **15** |
| *fishmeal* | *50* | *33* | *27* | *33* |
| *soyabean meal* | *0* | *20* | *30* | *10* |

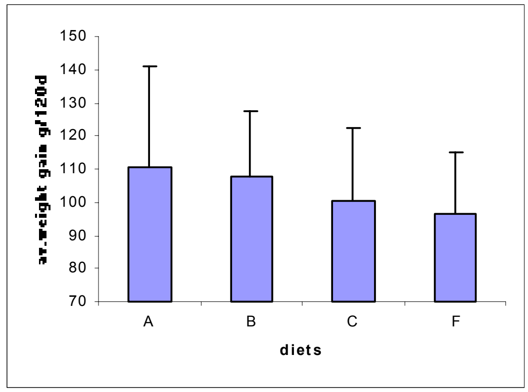



Fig.2. Practical diets with FM reduction for *L.calcarifer* juveniles and growth performances (weight gain/120 days) by incorporation of soybean meal (SBM 48). ( Boissin & Aquacop,1989; *unpublished results*).

- corn meal: it was chosen for salmonids feeds even if its protein quality is variable depending on the way of processing and a risk of overheated product, but it use was less frequent in marine rations.

- alkane - diets in early seventies were already used in replacement of fish meal with success. … At that time, fishmeal level was at least 40% of the diet. In Asia, the availability of marine sources: squid meal, squid liver powder, whole shrimp meal,….contributed to improve considerably feed efficiency. Such considerations led nutritionists to find the best mix of potent ingredients to meet such high growth rate.

- Algae meal is around 23% protein content and carbohydrates would represent 40-60% with starch, mannitol, laminarine, and cellulose. for example by the bio-availability of calcium source and trace elements.(iodine, zinc) Although preliminary studies need to be extended to species, this kind of product should be considered for future rations in the perspective of fishmeal and shrimp partial replacement , considering both growth rate and health status.

## BREEDERS

Broodstock for marine aquaculture species required a particularly up-graded diets where quality ingredients need to be selected (Wouters *et al.*, 2001). Breeders kept in round tanks can be handled, injected and spawned regularly (Nédélec *et al.*, 1997). Barramundi (sea bass) have been found easier to spawn voluntarily or induced tank spawning; maturation in captivity all year round is achieved under environmental control (T°C and photoperiod) in order to get 1-7 million eggs /month / pond.

For breeders nutrition, has been modified as far as animals reach 3-4 kg: instead of a regular grower diet, (48%CP 10%lipids) a mixture of fresh ingredients such as squid, mussel, and shrimp is prepared and added to wheat gluten, vitamins, fish oil to form what is called a moist extruded feed (30%moisutre), frozen and supplied on an automatic feeder (rolling carpet) placed above a $30m^3$ round fiberglass tank. So, in place of dry ingredients, the major change in view of reproduction is to bring fresh materials, increasing palatability and more prone to cover special requirement in terms of broodstock requirements (water, native protein, n-3 fatty acids, phospholipids, pigments,…) ; Feed ration is around 2% of live weight per day to sustain good maintenance conditions of breeders.

## CONCLUSIONS

Raw materials are selected for their nutritive value, balance in amino acids, digestibility of proteins, level of succulence, absence of anti-nutritional factors, availability, cost, regularity in composition over a year, etc., Raw materials will not be considered in a same way whether rearing conditions are intensive, semi-intensive, in low water renewal,… also, it will depend on stage of development of fish: for example at larval stage , raw materials are different in composition , quality, cost,.. from ingredients for a grower diet, and similarly, one will select high quality ingredients, native protein sources for a breeder diet. Another consideration is the balance of the diet in rearing conditions in earthen ponds for example, where at least during a certain period, natural productivity (biota) will represent a substantial nutrient input and then feed will become a "supplemental feed" more or less. In summary, high quality ingredients will be selected for larval stages and breeder diets, ingredients of prime quality will make the feed for intensive farming of many species, Basic ingredients (plant sources mainly) will serve in case of semi-extensive rearing system, and wastes of fish processing (for example with rainbow trout) can represent a solution in certain situations (Abdel-Warith *et al.*, 2000). At last, farm made feeds represent a solution for local ingredients (alfalfa, rice bran, copra,…) in particular context. This topic faced a strong evolution with the reduction of fishmeal in formulation, the possible utilization of transgenic lupine (Kissil *et al.*, 2000) , the drastic reduction in meat by-products, dioxin found in a small category of fish meals ,…and the general trend to go for mare and more plant sources with a corollary which is: (i) more anti-nutritional factors to cope with (ii) a great need for attractants , phagostimulating substances to compensate the lack of succulence with the retrieval of fishmeal. By and large, this trend should be viable from a nutritive point of view, meaning that a well-balanced diet could be found apart form the restriction mentioned above. The least cost formulation is set with new data, updated ones, with more technical constraints than before. Linear programming has limits and one should avoid too much simplification such as what happened with brewery yeasts (conferring a prophylactic aspect in tanks,..) whose properties are so much needed today with dramatic increase in bacterial or virus epizooties in many places where farming is under development.

## ACKNOWLEDGEMENTS

Authors are gratefully acknowledging colleagues of Aquacop and Dr. J. Guillame who took large contribution to the work on sea bass inside the AADCP component 4.

### REFERENCES

Abdel-Warith, A. A., Russell, P. M., Davies, S.J., 2001. Inclusion of a commercial poultry by-product meal as a protein replacement of fish meal in practical diets for African catfish *Clarias gariepinus* (Burchell 1822). Aquaculture Research, 32(1)296-305.

Anon., 1999. Artificial diet on performance of black sea bream *(Sparus macrocephalus*). Marine Science, 2 : 9-10.

Aquacop, 1983. Production of feeds to support shrimp production in Tahiti. *Proc. 1st Biennal Conf. Warm Water Aquaculture, Crustacea, Brigham Young University, Hawaii*: 382-387.

Aquacop, Cuzon, G., Thouard, E., 1993. Feeding of cultured *Lates calcarifer*. Replacement of fish meal by greaves meal in a grow-out diet. Ifremer, internal report, 30pp.

Cahu, C. L., Zambonino-Infante, J. L., Quazuguel, P., Le Gall, M. M., 1999. Protein hydrolysate vs. fish meal in compound diets for 10-day old seabass *Dicentrarchus labrax* larvae. Aquaculture, 171(1-2)109-119

Chaimongkol, Boonyaratpalin, M., 2001. Effects of ash and inorganic phosphorus in diets on growth and mineral composition of sea bass *Lates calcarifer* (Bloch). Aquaculture Research, 32 (suppl.1): 53-59.

Chamberlain, G. W., Miget, R. J., Haby, M. G., (comps.), 1990. Red drum Aquaculture. Rep. Texas. A&M Univ. Sea Grant Program.Tamu-SG-90-603:pp. 109-112

Chen, J. C., Lee, W. C., 1999. Growth of Taiwan abalone *Haliotis diversicolor supertexta* fed on *Gracilaria tenuistipitata* and artificial diet in a multiple-tier basket system. Journal Shellfish Resources. 18(2) 627-635.

Chu, F.- L. E., Ozkizilcik, S., 1999. Acceptability of complex microencapsulated diets by striped bass (*Morone saxatilis*) larvae. Journal Experimental Marine Biology and Ecology, 237(1) 1-9.

Cuzon, G., Chou, R., Fuchs, J., 1990. Nutrition of the seabass *Lates calcarifer* .Barret, J. (ed.) Advances in Tropical Aquaculture, Tahiti (French Polynesia), 20 Feb - 4 Mar 1989. Actes Colloque Ifremer. No. 9 pp. 757-763.

Davis, A. D., Arnold, C. R., 2002. Red drum production diets: replacement of FM by SBM. Proceedings World Aquaculture Society, Pekin, Abstract, p.167.

Davis, D. A., Lazo, J. P., Arnold, C. R., 1999. Response of juvenile red drum (*Sciaenops ocellatus*) to practical diets supplemented with medium chain triglycerids, Fish Physiology Biochemistry. 21(3) 235-247.

Duray, M. N., Juario, J. V., 1987. Broodstock management and seed production of *S.guttatus*(Bloch) and *L.calcarifer*(Bloch). In: perspectives in aquaculture development in SEA and Japan. Proceedings seminar on aquaculture development on SEA, Iloilo, Philippines, Juario and Bonito eds., 195-210.

El-Sayed, A. F. M., 1994. Evaluation of soybean meal, spirulina meal and chicken offal meal as protein sources for silver sea bream (*Rhabdosargus sarba*) fingerlings. Aquaculture 127 /169-176.

Fagbenro, O. A., 1999. Comparative evaluation of heat-processed winged bean (*Psophocarpus tetragonolobus*) meals as partial replacement for fish meal in diets for the African catfish (*Clarias gariepinus*). Aquaculture, 170(3-4) 297-305.

Fredette, M., Batt, J., Castell, J., 2000. Feeding stimulant for juvenile winter flounders, North American Journal Aquaculture. 62(2) 157-160.

Gatlin, D. M. III, Gaylord, T. G., Keembiyehetty, C. N., 1996. Nutrition and feeding of red drum and hybrid striped bass in intensive recirculating systems; ,Honolulu, HI (USA),11-14 Jun 1995 Proceedings of the PACON Conference on sustainable aquaculture '95.p. 173.

Gara, B., Shields, R. J., Mc Evoy, L., 1998. Feeding strategies to achieve correct metamorphosis of Atlantic halibut, *Hippoglossus hippoglossus* L., using enriched *Artemia.* Aquaculture Resources, 29(12) 935-958.

Garcia-Galano, T., Pérez, J. C., Gaxiola, G., Sanchez, A., 2000. Effect of feeding frequency on gastric evacuation and growth of juveniles of *C. undecimalis* (Bloch). V Symposium Intl. de Nutrición Acuícola, 19-22 nov.2000, Merida,Yucatán.

Garcia-Ortega, A., Huisman, E. A., Sorgeloos, P., Verreth, J., 2001. Evaluation of protein quality in micro bound starter diets made with decapsulated cysts of *Artemia* and fishmeal for fish larvae, Journal World Aquaculture Society 32(3) 317-329.

Gaxiola, G., Gomez, L. M., Garcia, T. J., Garcia-Galano, T., Gracia, V. Y., Sánchez, A., 2000. Adaptación enzimática a tres frecuencias de alimentación en juveniles del *C. udecimalis.*V Symposium Int[l] de Nutrición Acuícola, 9-22 nov. 2000, Mérida,Yucatán.

Gracia-López, V., García-Galano, T., Gaxiola-Cortés, G., Pacheco-Campos, J., 2001. Preliminary results on dietary protein level and use of commercial feeds for juvenile of common snook, *Centropomus undecimalis* (Bloch, 1792).

Glencross, B. D., Van Barneveld, R. J., Carter, C. G., Clarke, S. M., 1999. On the path to a manufactured feed for farmed southern blue fin tuna. World Aquaculture. 30(3)42-46.

Hamre, K., Naess, T., Espe, M., Holm, J. C., Lie, O., 2001. A formulated diet for Atlantic halibut (*Hippoglossus hippoglossus, L.*) larvae. Aquaculture Nutrition, 7(2) 123-132.

Hardy, R. W., 1998. Fish nutrition: increasing aquaculture production. Aquaculture. Magazine: 24(3)86-90.

Hardy, R. W., 1999. Fish, feeds, and nutrition: grains and their by- products. Aquaculture. Magazine: 25 (6) 54-58.

Holt, G. J., 2001. Red drum *Sciaenops ocellatus* culture. Aquaculture 2001, Lake Buena Vista, FL (USA),21-25 Jan 2001, Aquaculture 2001: Book of Abstracts, 754 pp.

Izquierdo, M. S., Socorro, J., Arantzamendi, L., Hernandez-Cruz, 2000. Recent advances in lipid nutrition in fish larvae. Fish Physiology and Biochemistry Special issue: Advanced Biochemical and Molecular Approaches to Sea Bream. Aquaculture and its Impact on the Environment. 22: 2 (97-107).

Kaushik, S. J., Cravedi, J. P., Lalles, J. P., Sumpter, J., Fauconneau, B., Laroche, M., 1995. Partial or total replacement of fish meal by soybean protein on growth, protein utilization, potential estrogenic or antigenic effects, cholesterolemia and flesh quality in rainbow trout, *Oncorhynchus mykiss*. Aquaculture, 133 (3-4) 257-274.

Kissil, G. W., Lupatsch, I., Higgs, D. A., Hardy, R. W., 2000. Dietary substitution of soy and rapeseed protein concentrates for fish meal, and their effects on growth and nutrient utilization in gilthead seabream *Sparus aurata* L. Aquaculture Research 31 (7) 595-601.

Knauer, J., Southgate, P. C., 1999. A review of the nutritional requirements of bivalves and the development of alternative and artificial diets for bivalve aquaculture, Review Fisheries Science 7(3/4) 241-280.

Krogdahl, A., Bakke- Mckellep, A. M., Roed, K. H., Baeverfjord, G., 2000. Feeding Atlantic salmon *Salmo salar* L. soybean products : effects on disease resistance (furunculosis), and lysozyme and IgM levels in the intestinal mucosa, Aquaculture Nutrition 6(2) 77-84.

Lee, H. B., Guillaume, J., Chou, R., Heng, H. H., Cuzon, G., 1994. Asian sea bass (*Lates calcarifer*, Bloch) nutrition. A review of the results from AADCP component 4. In second fish nutrition workshop, 25-27/10/1994, Singapore.

Lochmann, R. T., Gatlin, D. M. III., 1993. Essential fatty acid requirement of juvenile red drum (*Sciaenops ocellatus* ). Fish Physiology Biochemistry 12(3) 221-235

Lovell, T. R., 1979. Formulating diets for aquaculture species. Feedstuffs, 51(27)29-32.

McGoogan, B. B., Reigh, R. C., 1996. Apparent digestibility of selected ingredients in red drum (*Sciaenops ocellatus*) diets. Aquaculture 141( 3-4) 233-244.

Métailler, R., Cadean-Roa, M., Person-LeRuyet, J., 1983. Attractive chemical substances for the weaning of dover sole (*Solea vulgaris*): qualitative and quantitative approach. Journal World Aquaculture Society, 14:679-684.

Moon, H. Y., Gatlin, D. M. III., 1990. Amino acid nutrition of the red drum (*Sciaenops ocellatus* ): Determination of limiting amino acids and development of a suitable amino acid test diet. Takeda, M., Watanabe, T. (eds.)3. Int. Symp. on Feeding and Nutrition in Fish, Toba (Japan),28 Aug-1 Sep 1989 The proceedings .pp. 201-208.

Mukhopadhyay, N., Ray, A. K., 1999. Utilization of copra meal in the formulation of compound diets for rohu, *Labeo rohita,* fingerlings. Journal Applied Ichthyology 15(3) 127-131.

Nédélec, G., Thouard, E., Aquacop., 1997. Controlled year-round spawning of tropical sea bass *L. calcarifer* in captivity in French Polynesia. Abstracts island tropical aquaculture, Martinique, p.221.

Obaldo, L. G., Dominy, W. G., Terpstra, J. H., Cody, J. J., Behnke, K. C., 1998. The impact of ingredient particle size on shrimp feed. Journal Applied Aquaculture 8(4) 55-66.

Obaldo, L. G., Dominy, W. G., Ryu, G. H., 2000. Extrusion processing and its effect on aquaculture diet quality and shrimp growth. Journal Applied Aquaculture 10(2)41-53.

Ohs, C. L., D'Abramo, L. R., Buddington, R. K., Robinette, H. R., Roethke, J. M., 1998. Evaluation of a spray- dried artificial diet for larval culture of freshwater prawn, *Macrobrachium rosenbergii*, and striped bass, *Morone saxatilis.* Aquaculture Nutrition 4(2)73-82.

Oliva-Teles, A., Gouveia, A. J., Gomes, E., Rema, P., 1994. The effect of different processing treatments on soybean meal utilization by rainbow trout, *Oncorhynchus mykiss.* Aquaculture. Special issue: Fish Nutrition and Feeding. 124( 1-4) 343-349.

Partridge, G. J., Southgate, P. C., 1999. The effect of binder composition on ingestion and assimilation of micro bound diets (MBD) by barramundi *Lates calcarifer* Bloch larvae. Aquaculture. Resources 30(11/12) 879-886.

Pearce, C. M., Daggett, T. L., Robinson, S. M. C., 2002. Effect of binder type and concentration on prepared feed stability and gonad yield and quality of the green sea urchin, *Strongylocentrotus droebachiensis*, Aquaculture, 205(3/4) 301-323.

Pérez-Camacho, A., Albentosa, M., Fernandez- Reiriz, M. J., Labarta, U., 1998. Effect of micro algal and inert (cornmeal and cornstarch) diets on growth performance and biochemical composition of *Ruditapes decussatus* seed, Aquaculture 160(1/2) 89-102.

Reigh, R. C., Ellis, S. C., 1992. Effects of dietary soybean and fish-protein ratios on growth and body composition of red drum (*Sciaenops ocellatus* ) fed isonitrogenous diets. Aquaculture. 104 (3-4)279-292.

Rimmer, M. A., Reed, A., 1989. Effects of nutritional enhancement of live food organisms on growth and survival of barramundi (sea bass) *L.calcarifer* (Bloch) larvae. Advances in tropical aquaculture, Tahiti, Feb.20-March 4,1989. Aquacop, Ifremer. Actes de colloque, 9 pp.611-623.

Robinson, E.H., 1991. Nutrition and feeding of red drum. Handbook of nutrient requirements of finfish. Wilson, R.P. (ed.)196 pp

Rout, R. K., Bandyopadhyay, S., 1999. A comparative study of shrimp feed pellets processed through cooking extruder and meat mincer, Aquaculture Engineering. 19 (2) 71-79.

Seiffert, M. E. B. Cerqueira, V. R., Madureira, L. A. S., 2001. Effect of dietary (n-3) highly unsaturated fatty acids on growth and survival of fat snook (*Centropomus parallelus,* Pisces: Centropomidae) larvae during first feeding. Brazilian Journal of Medical and Biological Research . 34( 5) 645-651

Sener, E., Yildiz, M., 2000. Aquaculture potential and fish feed production in Turkey. World Aquaculture 31(3)52-56.

Shimeno, S., Matsumoto, M., Hosokawa, H., Masumoto, T., Matsumoto, T., 1999. Effect of dietary types on the utilization of practical diet by yellowtail, Nippon Suisan Gakkaishi 65(5) 866-871.

Soletchnik, P., Thouard, E., Goyard, E.,Yvon, C., Baker, P., 1989. First larval rearing trials of red drum in Martinique. IN. DE Wohlschlag. Eds. In red drum symposium 22-24 June. Corpus Christi,Texas, USA.

Spisni, E., Tugnoli, M., Ponticelli, A., Mordenti, T., Tomasi, V., 1998. Hepatic steatosis in artificially fed marine teleosts. Journal Fish Diseases 21(3) 177-184.

Steffens, W., 1994. Replacing fishmeal with poultry by-product meal in diets for rainbow trout, *Oncorhynchus mykiss*. Aquaculture 127: 27-34.

Sugiura, S. H., Dong, F. M., Rathbone, C. K., Hardy, R. W., 1998. Apparent protein digestibility and mineral availabilities in various feed ingredients for salmonid feeds, Aquaculture 159 (3/4) 177-202.

Sugiura, S. H., Dong, F. M., Hardy, R. W., 1998. Effects of dietary supplements on the availability of minerals in fish meal; preliminary observations. Aquaculture 160 (3/4) 283-303.

Takii, K., Kawamura, K., Urushihara, K., Hanayama, M., Nakamura, M., Miyashita, S., Kumai, H., Ukawa, M., 2001. Digestive function of tiger puffer compensates for dietary soybean trypsin inhibitors and extruded soybean meal. Suisan Zoshoku, 49 (1) 67-76.

Teshima, S., Ishikawa, M., Koshio, S., 2000. Nutritional assessment and feed intake of microparticulate diets in crustaceans and fish. Aquaculture Resources 31 (8/9) 691-702.

Thoman, E. S., Davis, D. A., Arnold, C. R., 1999. Evaluation of grow out diets with varying protein and energy levels for red drum (*Sciaenops ocellatus*). Aquaculture 176 (3/4) 343-353.

Tookwinas, S., 1989. Larviculture of *L.calcarifer* and *E.malabaricus*. Advances in tropical aquaculutre, Tahiti, Feb.20-March 4, 1989. Aquacop, Ifremer. Actes de colloque, 9 pp.645-659.

Tucker, J. W. Jr., Lellis, W. A., Vermeer, G. K., Roberts, D. E., Jr., Woodward, P.N., 1997. The effects of experimental starter diets with different levels of soybean or menhaden oil on red drum (*Sciaenops ocellatus*). Aquaculture 149( 3-4) 323-339.

Tucker, J. W., Kennedy, Jr., Blain S., 2001. Snook culture .Aquaculture 2001, Lake Buena Vista, FL (USA), 21-25 Jan 2001 Aquaculture 2001: Book of Abstracts World Aquaculture Society, 143 J. M Parker Coliseum Louisiana State University Baton Rouge LA 70803 USA :754 pp.

Tucker, W. J., 1987. Snook and tarpon snook culture and preliminary evaluation for commercial farming. The Progressive Fish Culturist 49: 57-59.

Walford J, Lim T. M., Lam T. J., 1991. Replacing live foods with microencapsulated diets in the rearing of seabass (*Lates calcarifer*) larvae: do the larvae ingest and digest protein membrane microcapsules?. Aquaculture 92. 225-235.

Watanabe, K., Ura, K., Yada, T., Kiron, V., Satoh, S., Watanabe, T., 2000. Energy and protein requirements of yellowtail for maximum growth and maintenance of body weight, Fisheries Sciences. 66 (6) 1053-1061.

Watanabe, T., Aoki, H., Shimamoto, K., Hadzuma, M., Maita, M., Yamagata, Y., Kiron, V., Satoh, S., 1998. A trial to culture yellowtail with non-fishmeal diets, Fisheries Science. 64 (4) 505-512.

Webster C. D., Tidwell, J. H., Yancey, D. H., 1991. Evaluation of distillers'grains with solubles as a protein source in diets for channel catfish. Aquaculture, 96:179-190.

Williams, K. C. Barlow, C. Rodgers, L. Hockings, I. Agcopra, C., 1994. Abstracts from Australian barramundi farming workshop. Dept of Primary Industries, Brisbane, QLD. (Australia) pp. 13-15.

Wouters, R., Lavens, P., Nieta, J., Sorgeloos, P., 2001. Penaeid shrimp broodstock nutrition: an updated review on R and D. Aquaculture 202 (1/2) 1-21.