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Feed formulation, diet development and feed technology

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SUMMARY – The first and foremost criterion is that diets should supply all the essential nutrients and energy in adequate proportions to satisfy the requirements for bodily or gonadal growth, health and well being of the species concerned. In the absence of reliable data on the nutrient requirements of species that are new to aquaculture, the main question that arises is how far can we adopt data from other closely-related species. To obtain basic data on nutrient requirements, there is also a definite need for developing experimental diets with purified ingredients that are well accepted by the species concerned. In developing practical diets using "least-cost" formulations, the choice and level of incorporation of the different ingredients should be based on available nutrient/energy content of each ingredient rather than on the basis of crude chemical composition of the ingredients. These formulations should also recognise the importance of possible nutrient imbalances and interactions between nutrients or ingredients. Specific attention should be paid to the amount of any known anti-nutritional factors (ANF) in the individual ingredients and especially to the tolerable levels of ANFs in the final diet. Ingredient or feed processing techniques can contribute towards the reduction of ANFs. Processing techniques also affect nutrient availability and the physical characteristics of the final product. For instance, the texture, water stability or buoyancy of the diets can be modified both by the physical nature of the ingredients as well as by feed processing techniques. While feed ingredients used in aquaculture feeds should be finely ground, the final feed should have no fines. Additives of technological interest should not affect nutrient availability. The physical characteristics should also meet the behavioural needs of the animal in terms of palatability and acceptance by the species. The technology of feed manufacture and the inherent constraints can thus vary considerably depending upon the species or the stage of the life cycle. Feed technology has a major role in supplying diets suited to the different size classes of fish grown in different environments. The success in the development of adequate feeds for aquatic animals depends on close co-operation between the feed engineer and the nutritionist.

Key words: Aquaculture, feeds, formulation, technology.

RESUME – "Formulation des aliments, mise au point des régimes et technologie alimentaire". Le premier critère et le plus important est que les régimes devraient apporter tous les nutriments essentiels et l'énergie en proportions adéquates pour satisfaire les besoins pour la croissance corporelle ou des gonades, pour la santé et le bien-être des espèces concernées. En l'absence de données fiables sur les besoins nutritionnels des espèces qui sont nouvelles en aquaculture, la principale question qui se pose est jusqu'à quel point est-ce que nous pouvons adopter des données d'autres espèces étroitement liées. Pour obtenir des données fondamentales sur les besoins nutritionnels, il est absolument nécessaire de mettre au point des régimes expérimentaux avec des ingrédients purifiés qui soient bien acceptés par les espèces concernées. Dans le développement de régimes pratiques en utilisant des formulations "à moindre coût", le choix et le niveau d'incorporation des différents ingrédients devraient être basés sur la teneur disponible en nutriments/énergie de chaque ingrédient plutôt que sur la base de la composition chimique brute des ingrédients. Ces formulations devraient également reconnaître l'importance de possibles déséquilibres en nutriments et des interactions entre nutriments et ingrédients. Une attention spéciale devrait être accordée à la quantité de tout facteur antinutritionnel connu (ANF) dans les ingrédients individuels et particulièrement aux niveaux tolérables d'ANF dans le régime final. L'ingrédient ou les techniques de traitement des aliments peuvent contribuer à la réduction des ANF. Les techniques de traitement affectent également la disponibilité des nutriments et les caractéristiques physiques du produit final. Par exemple, la texture, la stabilité dans l'eau ou la flottabilité des régimes peuvent être modifiés par la nature physique des ingrédients ainsi que par les techniques de traitement de l'aliment. Tandis que les ingrédients alimentaires utilisés dans les aliments aquacoles devraient être finement broyés, l'aliment final ne devrait pas avoir d'impuretés. Les additifs d'intérêt technologique ne devraient pas affecter la disponibilité en nutriments. Les caractéristiques physiques devraient également répondre aux besoins comportementaux de l'animal en termes de palatabilité et d'acceptation par l'espèce. La technologie de la fabrication d'aliments et les contraintes inhérentes peuvent donc varier considérablement en fonction de l'espèce ou du stade du cycle de vie. La technologie alimentaire joue un rôle majeur en apportant des régimes adaptés aux différentes classes de tailles de poissons élevés dans différents environnements. Le fait de réussir à développer des aliments adéquats pour les animaux aquatiques repose sur une étroite coopération entre le formulateur d'aliments et le nutritionniste.

Mots-clés : Aquaculture, aliment animal, formulation, technologie.

Introduction

A diet should supply all essential nutrients and energy in tune with the animal's needs for the maintenance of vital physiological functions such as growth, reproduction and health. Besides, in aquaculture as in other animal production systems, another major issue is that of ensuring flesh and environmental quality, both of which are related to nutrition. Since the nutrient requirements for all the new species under aquaculture are not known, it is rather a common practice to extend data from more or less closely related species. In the formulation of diets, it is essential that, even when the diets are formulated theoretically to contain all the essential nutrients in adequate quantities, the availability of these nutrients from the raw materials used can vary significantly. The diet should be supplied in a form which is easily accepted by the cultivated animal and should have little adverse environmental impact.

Choice and quality of ingredients

Despite much research, both intensive and semi-intensive aquaculture relies upon a relatively small number of feed ingredients. Under semi-intensive culture conditions, cereal bran-oilcake mixture remains the major aquafeed. In intensive aquaculture, the diets are formulated to be nutrient and energy dense, based mainly on ingredients of marine origin. Since most teleosts are known to utilise dietary carbohydrates rather poorly, the chosen ingredients are necessarily protein and energy-rich. When it comes to finding alternatives to fishmeals as a protein and amino acid source, several other agricultural by-products such as animal by-products, cereals (wheat, corn), pulses (lupin, peas, faba beans), oil seeds (soybean, rapeseed) hold potential interest (Table 1), depending upon local availability and cost.

Table 1. Crude protein (CP) levels of different alternatives to fishmeal

CP levels	Ingredients
<25%	Whole cereals, pulses, oil seeds
25-50%	Oil seed meals
>50%	Animal by-products (meat meal, blood meal), plant protein concentrates, isolates, extractives, single cell proteins

Value of ingredients as protein and energy sources

For marine finfish species, published data on nutrient availability from commonly used ingredients are relatively scarce. In order to obtain reliable data on apparent digestibility coefficients (ADC) of ingredients, a standardised methodology is necessary. The fecal collection method should avoid any possible leaching of nutrients. In European sea bass, Spyridakis *et al.* (1989) found that data obtained with a fecal collection method based on continuous screening (Choubert *et al.*, 1982) was more comparable to that obtained by immediate siphoning of fecal material. Recent work by Sugiura *et al.* (1998) also indicate that the methods of calculation of ADC of ingredients need refinements when dealing with ingredients having extremely variable levels of a given nutrient.

For practical purposes, it is very much tempting to use *in vitro* methods to determine protein digestibility and new methods have been developed recently (Bassompierre, 1997), although a clear correlation between data obtained *in vitro* and *in vivo* is rather poor (Gomes *et al.*, 1993) and needs confirmation using the same species of fish concerned. Similarly, the values based on digestibility measured in mink have been found to correlate well with the biological values in sea bream (Aksnes *et al.*, 1997) but no systematic work on correlation between digestibility data obtained in mink as compared to marine fish has been done so far. Besides, such methods provide information only on protein digestibility and not for other nutrients or energy which are also of interest to the feed formulator. Some literature data on the digestibility of some ingredients in European sea bass and gilthead sea bream are summarised in Table 2.

Table 2. Some data on available nutrient and energy from different ingredients for sea bream and sea bass[†]

	DM (%)	CP (%)	CF (%)	Ash (%)	P (%)	Fibre (%)	NFE	DP (%)	DE (kJ/g)	Available P (%)
Gilthead sea bream										
Poultry by-product meal	87.8	43.9	17.4	8.9		2.9	14.8	35.9	18.2	
Feather meal A	89.0	54.1	3.2	6.0		3.6	22.2	13.5	1.3	
Feather meal B	94.0	57.5	6.0	5.2		2.9	22.3	33.1	15.1	
Meat and bone meal C	86.9	50.9	2.4	18.4		4.8	10.3	36.8	11.4	
Meat and bone meal E	95.1	43.9	6.2	19.1		3.2	22.6	15.6	2.5	
Meat and bone meal L	95.0	45.8	7.3	20.3		2.8	18.8	20.1	5.7	
Poultry meat meal	91.3	52.3	8.0	10.5		3.3	17.2	47.0	14.5	
Lipomel	90.7	49.9	17.2	7.2		3.3	13.2	32.7	9.2	
Skimmed milk	89.2	29.8	3.1	8.5		2.6	45.2	28.5		
Blood meal	92.1	61.0	3.2	5.1		2.8	19.9	28.3	13.7	
Herring meal	94.3	53.8	9.1	10.3		2.4	18.7	51.6	20.9	
Sunflower meal	87.3	34.6	3.2	7.3		12.8	29.5	29.8		
Cottonseed meal	87.4	33.6	2.9	6.7		4.7	39.5	25.3	7.5	
Soybean meal	84.2	38.7	3.5	6.7		4.7	30.6	35.2	8.5	
Tomato pulp meal	88.3	26.8	7.1	7.9		20.9	25.7	5.4	1.9	
Corn gluten meal	88.2	49.6	5.2	6.3		4.2	23.0	44.7	17.1	
Flaked maize	86.9	20.7	3.4	4.7		10.6	47.5	12.5	6.2	
Full fat soybean meal	87.0	33.9	14.4	6.3		4.6	27.8	25.6	12.8	
Corn gluten feed	84.7	25.6	6.6	7.8		14.5	30.3	16.7	6.4	
European sea bass										
Fishmeal, Norway	92.5	74.9	13.7	11.4	1.8			71.9	21.5	1.4
Fishmeal, Portugal	89.5	70.1	11.5	18.3	3.1			62.7	18.3	2.3
Soybean meal (toasted, dehulled, solvent extracted)	87.7	51.6	2.5	7.5	0.6			46.3	14.2	0.2
CPSP G	96.7	71.7	21.3	4.5	0.8			69.2	26.0	0.7
Blood meal	90.4	97.1	1.0	1.9				88.0	22.5	
Dextrin, yellow, corn	88.3			0.3					16.2	
Meat meal, defatted, steam cooked	96.5	75.1	14.7	10.2	3.5			69.1	19.4	2.8

[†]Data for gilthead sea bream are from Nengas *et al.* (1995); data for European sea bass are from da Silva and Oliva-Teles (1998).

Lipid sources

With regard to fatty acid sources, given the essential nature of highly unsaturated fatty acids for marine finfish, the natural choice is marine oils rich in docosahexaenoic acid (22:6n-3; DHA). New data indicate the importance of the relative proportions of DHA as well as other fatty acids of the w-6 series in marine finfish nutrition. The choice of oils should be more closely controlled in terms of their fatty acid composition, giving special attention to prevent oxidative damage of such unsaturated fatty acid sources during storage and feed processing. One should also keep in mind that there is no single indicator of oxidative damage and that one might have to resort to a combination of methods to determine the antioxidant status of raw materials.

Premixes and additives

It is common practice that vitamins, minerals and trace elements are supplied as a premix which is added to the major ingredient mixture in small amounts. Based on data available on the nutritional

requirements, such premixes should be formulated in order to avoid any possible interaction between potent components (e.g. choline and trace elements) within the premix itself. The concentration of individual nutrients within the premix should be such as to enable uniform distribution of each of these in the finished feed. Leaching of water-soluble vitamins and especially of ascorbic acid is a major concern in the aquaculture feed industry. As regards ascorbic acid, ascorbyl phosphates are found to be more water stable even after extrusion than simple coated forms (Gadient and Fenster, 1994). In a recent study, Marchetti *et al.* (1999) observed that fat coating was required to increase the water stability (over 60 to 120 minutes) of water-soluble vitamins in both pelleted and extruded feeds, although the rationale behind search for such a long stability might be questionable.

Additives include those having a potential biological or physiological significance such as antioxidants, probiotics, antibiotics, attractants and enzymes as well as those having technological significance such as emulsifying, binding or preserving agents. A number of inorganic or organic binders ranging from cement (Rumsey *et al.*, 1981) to alginates (Storebakken and Austreng, 1987) have been tested in aquaculture feeds. From a nutritional point of view, such non-nutrient binders are advantageously replaced by raw materials such as wheat gluten, extruded peas or gelatinised starch having at least some nutritional significance. A recent study by Dias *et al.* (1998) indicates that sea bass fed diets containing such non-nutrient fillers tend to compensate by increasing their overall feed intake.

In the general context of replacing fishmeal with other protein sources, some of the studies have suggested a possible decrease in palatability of such diets (Gomes *et al.*, 1995). For the feed formulator or manufacturer, an a priori, objective knowledge of the palatability of the finished diet is difficult to obtain. The possible role of specific feed attractants (Mackie and Mitchell, 1985; Hara, 1994) in improving nutrient intake is a field which requires also more attention.

Physical characteristics of ingredients

Since most fish do not possess the necessary grinding appendices in the mouth, dietary particle size should be sufficiently small to be ingested whole and available for digestive processes. Generally, it is recommended that the particle size of all ingredients be below 250 microns (NRC, 1993) but this should naturally be much lower when dealing with diets for larval or juvenile stages of fish. The significance of reducing particle size on improving physical characteristics of the finished feed is related to improved digestibility. Reduction in particle size improves gelatinisation ratio, increases pellet durability and water stability, but is economically rather expensive (Obaldo *et al.*, 1998). Studies by Jeong *et al.* (1991) and our own unpublished work have shown that starch digestibility is improved with increased gelatinisation ratios. Even in pigs, it has been shown that fine grinding improves ileal digestibility (Hess *et al.*, 1998). Since most ingredients used for terrestrial animal production are rather coarse, fish feed manufacturers resorting to such ingredients have to pay special efforts to reduce the particle size of such ingredients.

Another aspect which has practical significance for the feed processing engineer is the possible differences in physical characteristics of the ingredients such as pack density, specific gravity, compressibility, viscosity, heat capacity and conductivity, etc. The adaptability of the feed manufacturer and his machinery to such changes in the physical characteristics of ingredients is important in fine-tuning the final product still meeting the nutritional standards.

Quality control

For formulating diets for experimental purposes, it is necessary that all ingredients are controlled for all essential nutrients. But, under practical conditions, such a control is difficult to set forth and mostly restricted to rapid proximate composition analyses. Specific attention should however be given to obtain guarantees for absence of anti-nutritional factors, to avoid adulteration of products and for homogeneity between batches. Besides such quality control of incoming ingredients, a strict control of material flow within the factory is of utmost importance to avoid deterioration of the nutritional value (oxidation, potency of vitamins) of ingredients and premixes.

Feed formulation

Diet formulation represents translation of nutrient and energy requirements of a given species for a given response into an acceptable diet using a balanced mixture of ingredients which is economically sustainable. The reliability of knowledge on the quality of ingredients and the constraints retained both have an impact on the quality of diet formulation.

Importance of a reliable data base

For any end user and especially the feed formulator, a reliable, updatable database on chemical composition, physical characteristics and bioavailability information on feed ingredients is necessary. Although there are a number of generic databases such as INFIC or io7 either in printed form or in computerised form (Halver, 1995; Tran and Lapierre, 1997), the value of a database depends upon the convenience with which they can be updated and maintained on a regular basis. In this context, it is highly recommended that feed manufacturers constitute their own database depending upon the sources of raw materials that they commonly use. Most importantly, the feed formulator should have at his disposal, information on the bioavailability of nutrients and energy of each ingredient and for each species concerned. Hence, data on the levels of digestible protein, available amino acids, available phosphorus, digestible energy for each ingredient should be obtained and regularly incorporated in such a database. Further, such databases should also contain information on physical characteristics as mentioned above. One must admit that currently, such data are not available from a single public source for the aquafeed formulator. Some information on chemical composition and nutrient/energy availability as applicable to finfish is provided in the NRC (1993) and also in some text books devoted specifically to fish nutrition and feeding (see Guillaume *et al.*, 1999).

Least-cost formulation

Least-cost formulations should meet the nutritional requirements for a given production objective. It is the overall biological outcome of the product which should be kept in mind rather than the feed cost per unit weight. In this context, the nutritional constraints should be based on available nutrient content (digestible protein, digestible energy, available phosphorus, amino acid composition, digestible protein to digestible energy ratio, etc.) rather than on the basis of proximate analyses of the finished diet. For instance, in evaluating alternatives to fishmeal, more than unit protein cost, available amino acid cost per unit weight should be retained. In order to have an ideal amino acid balance, criteria such as the essential amino acid index has to be incorporated as a necessary constraint correcting possibly for changes in the digestible energy levels.

Linear programming is generally used with various degrees of sophistication and a number of commercial softwares are available for feed formulation. The choice of one over another should be based mainly on how far a given software allows for updating the data base as well as the constraints be it nutritional or technological. Development of quadratic analysis (second order polynomial equation) incorporating nutrient-response curves has been reported to be ideal than simple linear programming based on nutrient compositions alone (Lovell, 1987). The success of the latter depends however upon systematic compilation of reliable data on the biological response of a given species (body weight or protein gain, reproductive performance) corresponding to a given available nutrient supply in a given set of environmental conditions. For ease of purposes, some spread sheet softwares already incorporate solver modules which can be easily used for least-cost formulations, provided reliable data on ingredient composition including availability data are provided.

Diet development

Different kinds of diets

Even when the formulated diets are nutritionally well balanced, the form of supply of a diet bears special significance. The acceptance of dry diets in the case of marine fish larvae, broodstock fish or poorly domesticated species is a practical concern. The demand for different kinds of diets is rather

high in aquaculture, ranging from floating, sinking, soft or moist feeds. This demand attempts both to meet the inherent feeding behaviour (benthic or pelagic) of the species as well as the needs corresponding to different husbandry practices (e.g., control of feed intake and wastage). During the initial stages of salmonid culture, the use of moist or semi-moist diets was common. For some marine species, this practice still appears to prevail, although the adverse effects on feed stability and environmental quality are generally recognised. For species such as the yellowtail, extruded "soft-dry" diets have been developed (Watanabe *et al.*, 1991) and successfully used even for broodstocks (Watanabe *et al.*, 1996). Much experimental work has also dealt with the possible use of acid preserved fish silage for making semi-moist diets, but the economic viability of such practices remains doubtful in intensive aquaculture (Ives, 1991). Of late, the direct use of fish offals or trash fish combining micro-wave and extrusion technologies has regained some interest in order to decrease the pressure on marine fishery resources and as a measure of waste recycling with possibilities of formulating diets containing up to 47% fat (Hemre and Sandnes, 1999).

Another point which needs mention is that, in aquaculture, the feed particle diameter can vary considerably, ranging from less than 100 µ to even more than 20 mm, corresponding respectively to first-feeding larvae often weighing less than a mg to broodstock fish weighing several kilograms. For developing larval diets, a number of techniques have been employed: microencapsulation, protein or elastin coating, microbinding, microflakes, bound liposomes. In developing such diets, extreme attention should be paid to ingredient particle size, homogeneity, nutrient stability, density and attractiveness. Some progress has been made in the culture of marine finfish larvae with dry diets (Cahu *et al.*, 1998) and more is awaited in the years to come in order to fully and rationally exploit the potential of marine finfish culture.

Extrusion feed technology

Significant differences exist in the feed processing techniques used in fish nutrition research and in practical feed trials. For determination of nutrient requirements, for instance, often, diets made with purified ingredients are blended with water made into spaghettis and then either kept frozen or dried and crumbled before use. Dry or steam pelleting is also used for experimental purposes. Pelleting, which is easily applicable in the animal feed industry is done either with or without pre-conditioning, but cannot allow fat levels above 13 to 15%. Some double pelleting techniques can be used to reduce fines, increase pellet stability and fat content. Extrusion and to a lesser extent the expansion techniques which are used for aquaculture feed manufacture differ significantly in terms of thermal as well as mechanical treatments (Table 3).

Table 3. Thermal treatments and duration of treatment between different feed processing methods (from Melcion and van der Poel, 1993)

Process	Temp. (°C)	Duration (sec)
Pelleting		
Short conditioning	60-90	25-35
Long conditioning	60-95	70-250
Expansion	80-140	5-15
Extrusion	80-200	30-150

Nowadays, extrusion technology, although more expensive and technically demanding than conventional dry pelleting process, has become more and more common in the aquaculture feed industry. From a nutritional point of view, extrusion or expansion processes are known to improve starch (and consequently energy) digestibility of cereals (Bergot and Brèque, 1983) as well as pulses such as peas (Table 4; Kaushik *et al.*, 1993) or lupin (Burel *et al.*, 1998) and to decrease the levels of anti-nutritional factors and bacterial counts in the finished feeds (Melcion and van der Poel, 1993). This technology is considered flexible and versatile and as having the advantage of improving the physical characteristics of the diets by decreasing the levels of fines and improving water stability

(Kearns, 1993). However, depending on the raw material matrix, significant differences can be observed in terms of physical characteristics of the diets (Table 5). Some of the improvements achieved through such processes appear also to have significant beneficial effects in terms of fish growth and feed efficiency (Akimoto *et al.*, 1992; Aksnes *et al.*, 1997) as well as on reduction of N excretion (Robaina *et al.*, 1999). A simple reduction in the amount of fines in the feed significantly improves suspended matter release in the aquatic environment, while improving feed efficiency values.

Table 4. Apparent digestibility coefficients (ADC, %) of peas as affected by treatment in rainbow trout

Treatment	Raw	Dehulled	Dehulled, extruded and micronised
Dry matter	9.8	24.7	71.6
Protein	81.9	84.3	89.8
Energy	14.4	28.8	71.7
Starch	–	–	96.2

Table 5. Some physical characteristics of pelleted and extruded fish feeds made with identical ingredient mixtures (Kaushik and Melcion, unpublished data)

Feed mixture	A [†]		B ^{**}	
	Pelleted	Extruded	Pelleted	Extruded
Feed processing technology				
Mass/volume (g/l)	612	580	633	504
Durability (mechanical, pfost) (%)	87	100	93	99
Durability (pneumatic, holmen) (%)	25	97	70	94
Buoyancy (% residues at 30 sec)	0	0	0	10
Sinking rate (cm/sec)	8	6.2	9.7	4
Water stability (% residues at 10 min and 1 hr)	30/89	0/4	17/37	11/92
Slope of particle breakdown (10-60 min)	0.0114	0.0097	0.0052	0.0167
Oil absorbing capacity (%)	16	18	16	31

[†]A: basal diet containing fishmeal, fish oil, gelatinised starch, vitamin and mineral mixtures.

^{**}B: 80% A + 20% wheat gluten.

Conclusions

Currently, a limited number of ingredients are used in the formulation of feeds used in intensive aquaculture. The choice of these ingredients should be based on the nutritional value taking into account potential anti-nutritional factors rather than merely on the basis of cost per unit weight alone. Formulations should be based on available nutrient and digestible energy from the ingredients meeting constraints based on latest reliable data on nutrient requirements. It is worth keeping in mind that poor feed formulations cannot be improved by sophisticated feed processing technologies. Nutritional and technological improvements have an impact not only on the economics but also on the environment. There is a strong need for diversification of feed ingredients used in aquaculture. Research data indicate that even with marine finfish, dietary fishmeal levels can be significantly reduced, provided care is taken to inactivate or decrease the levels of anti-nutritional factors. Nevertheless, knowledge is needed on the possible long-term physiological consequences. Similarly, to what extent marine oils can be substituted even partially by other sources over the full life cycle of marine finfish remains another important area for further research. For larval stages of marine fish, development of artificial diets is a priority in order to obtain quantitative data on nutrient requirements.

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