EUROPEAN COMMISSION DIRECTORATE-GENERAL FOR AGRICULTURE AND RURAL DEVELOPMENT

Directorate B. Multilateral relations, quality policy

Expert Group for Technical Advice on Organic Production EGTOP

Final Report on Aquaculture (part A)

The EGTOP adopted this technical advice at the 8th plenary meeting of 03-05 December 2013

About the setting up of an independent expert panel for technical advice

With the Communication from the Commission to the Council and to the European Parliament on a European action plan for organic food and farming adopted in June 2004, the Commission intended to assess the situation and to lay down the basis for policy development, thereby providing an overall strategic vision for the contribution of organic farming to the common agricultural policy. In particular, the European action plan for organic food and farming recommends, in action 11, establishing an independent expert panel for technical advice. The Commission may need technical advice to decide on the authorisation of the use of products, substances and techniques in organic farming and processing, to develop or improve organic production rules and, more in general, for any other matter relating to the area of organic production. By Commission Decision (EC) No 427/2009¹ of 3 June 2009, the Commission set up the Expert Group for Technical Advice on Organic Production.

EGTOP

The Group shall provide technical advice on any matter relating to the area of organic production and in particular it must assist the Commission in evaluating products, substances and techniques which can be used in organic production, improving existing rules and developing new production rules and in bringing about an exchange of experience and good practices in the field of organic production.

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Commission Decision (EC) No 427/2009 of 3 June 2009 establishing the expert group for technical advice on organic production (O.J. L 139, 5.6.2009, p. 29–31)

The report of the Expert Group presents the views of the independent experts who are members of the Group. They do not necessarily reflect the views of the European Commission. The reports are published by the European Commission in their original language only. http://ec.europa.eu/agriculture/organic/home_en

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All declarations of interest of Permanent Group members are available at the following webpage: http://ec.europa.eu/agriculture/organic/home_en

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1. EXECUTIVE SUMMARY

In consideration of:

- the lack of organic juveniles reported by some MS;
- the restriction on the movement of live animals between countries and regions based on the Council Directive (EC) No 88/2006²;
- the reluctance of farmers to introduce on their farms animals which could be unsuitable for the local (geographical) environment (e.g. genetic or population traits, resistance to different diseases, growth performances, reproductive cycle, behavioural characteristics, etc.); and
- the lack in the Commission Regulation (EC) No 889/2008³ of specific organic rules for managing the life cycle stage between the hatching and the weaning of juveniles,

the Group supports the use of non-organic juveniles, for on-growing purposes, when organic aquaculture juvenile animals are not available, subject to the following restrictions/recommendations:

- a) Organic juveniles should be used when available.
- b) At least the latter two thirds of the duration of the production cycle shall be managed under organic management (Article 25(e)(2) of Commission Regylation (EC) No 889/2008).
- c) After the approval of specific organic rules for the life cycle stage between hatching and weaning of juveniles, a transitional period may be established to allow farmers to comply with the new rules.

Furthermore, the Group supports the creation of a database on the availability of organic juveniles produced in each country, comparable to the seed database (Article 48 of Commission Regulation (EC) No 889/2008). Transparency of the use of non-organic juveniles should be ensured in such a database.

Some typical aquaculture practices of extensive fish farming in wetlands, such as brackish water ponds, tidal areas and costal lagoons, closed by levees and banks, have been carried out in Italy and Spain, as well as in other coastal areas of Europe, for many centuries.

The Group recognizes the high value of these extensive aquaculture practices, in terms of cultural heritage, biodiversity conservation and economic perspectives for the local communities. The Group also recognizes that moving wild fry from the sea into the lagoon does not necessarily affect the stock status of the species concerned.

Therefore, with the exception of eels, the Group considers that restocking of wild fry in the extensive aquaculture farming carried out inside wetlands, such as brackish water ponds, tidal areas and costal lagoons is in line with the objectives, criteria and principles of organic aquaculture production, subject to the following restrictions:

- a) A management plan, approved by the local or national authority, that ensures the sustainable exploitation of the species concerned, should be provided.
- b) In extensive aquaculture farming carried out inside wetlands, such as brackish water ponds, tidal areas and coastal lagoons, the fish shall be fed with feed naturally available in the environment.

Commission Regulation (EC) No 889/2008 of 5 September 2008 laying down detailed rules for the implementation of Council Regulation (EC) No 834/2007 on organic production and labelling of organic products with regard to organic production, labelling and control

Council Directive (EC) No 2006/88 of 24 October 2006 on animal health requirements for aquaculture animals and products thereof, and on the prevention and control of certain diseases in aquatic animals

In the Group's opinion, fish should cover their needs for amino acids and fatty acids primarily through the natural compounds of the feed. In order to comply with the general rules on feed (cfr. Article 25(j) of Council Regulation (EC) No 889/2008), namely: "... optimum performance, animal health, high product quality, including the nutritional composition which shall ensure high quality of the final edible product and low environmental impact", the diet for carnivorous fish should be characterized by a well-balanced proportion of amino acids, fatty acids and lipids.

The Group also recognizes the specific needs of animal protein and lipids in the diet of shrimps, although in different proportion according to life stages. Therefore, the Group supports a limited use of fishmeal and fish oil derived from sustainable fisheries, as a supplement of the feed naturally available in the rearing environment of shrimps. Such feed rations could be up to 10% for the fish oil and up to 25% for the fish meal.

The Group is concerned about the consequences of the listed priorities of sourcing feed as laid down in Council Regulation (EC) No 889/2008 Article 25(k). Indeed, with respect to fish meal derived from trimmings, the risk is that the levels of phosphorus contained in such fish meal might result in conflicts with national environmental legislation.

Since fish meal and fish oil are limited resources, finding alternative protein sources for fish meal is clearly a high priority for organic aquaculture. Similarly, developing production of aquafeeds that: a) satisfy aquatic organisms' nutritional requirements for specific amino acids and fatty acids, b) suit their feeding habits and c) result in a high retention of nutrients to maintain animal health and to achieve good quality final products is a high priority.

As a consequence, the Group supports the use of alternative protein sources in organic aquaculture, when available and appropriate for brood-stock, weaning, and on-growing diets. The development of organic alternative protein sources should be considered a priority.

Considering all these issues, the Group concluded that the following alternative options should be considered, in order of priority:

- a) Besides fish meal and fish oil derived from trimmings of fish, crustaceans and molluscs, also fish meal and fish oil derived from "whole fish not used for human consumption", caught in sustainable fisheries, should be allowed as ingredients in feed for organic carnivorous fish. This includes feed for fry and brood-stock, as well as for on-growing fish, until sufficient alternative sources of protein and oil are available.
- b) The use of other alternative feed materials consisting of whole micro or macro organisms with high content of essential amino acids and lipids, where possible produced organically, may be needed and are to be preferred to the use of purified or free amino acids as feed supplements/additives.
- c) If not available from organic procedures, essential amino acids and lipids obtained by fermentation or other similar procedures should be allowed as ingredients/additives in carnivorous fish feed only if specifically authorised.

In the case of histidine, the approval in the specific context recommended by the Group should not be seen as a precedent for the use of histidine as a feed additive outside aquaculture, nor for the use of other free amino acids as feed additives for any type of livestock. Other uses / substances should be evaluated separately.

2. BACKGROUND

Organic aquaculture is a relatively new addition to the scope of EU organic legislation having been added for the first time by Council Regulation 834/2007⁴. The implementing rules were introduced via Commission Regulation (EC) No 710/2009⁵ which amended the main implementing rules for organic farming introduced by Commission Regulation 889/2008. The rules for aquaculture have applied for almost three years, i.e. since 1 July 2010. The final paragraph of Article 2 of Commission Regulation (EC) No 710/2009 states: "This Regulation may be revised on the basis of relevant proposals from Member States, which are accompanied by a duly justified motivation, with a view of the modification of this Regulation from 1 July 2013."

The group is therefore requested to prepare a report with technical advice on the matters included in the terms of reference.

3. TERMS OF REFERENCE

In the light of the most recent technical and scientific information available to the experts, the group is requested to report on the following list of requests.

1. **The use of non-organic Juveniles** in the context of the sequential phasing out of their use by 31.12.2015 (except for the specific cases involving the natural influx of fish and crustaceans, and also European glass eels and wild mollusc seed).

France, Italy, Germany and Spain have pointed out the lack of organic juveniles and the consequent difficulty in meeting the requirement to ensure that at least half come from organic sources by the end of 2013 and by two years from this date that all juveniles will be from organic sources. France points to the difficulty which the sector is having in meeting these requirements for a wide range of organic aquaculture, including oysters, freshwater fish, marine fish and shrimps and links this to the early stage of development and lack of critical mass of organic production (grow-out) generally. Italy points out that some of the difficulty is related to the need to source from a compartment of equal status under the aquaculture animal health rules (Council Directive (EC) No 2006/88⁶) and that in exceptional cases Member States should be allowed to issue exemptions under the flexibility rules (Article 22(2)(b) of Council Regulation (EC) No 834/2007). Spain points to the lack of juveniles for seabass and seabream and Germany to trout. Sweden proposes, in the context of the difficulty experienced in phasing out non-organic juveniles in agriculture that EGTOP consider if there are species, in additional to eels, for which it may be necessary to accept non-organic juveniles for a long time yet (in addition to the exceptions listed in the heading above).

Council Regulation (EC) No 834/2007 of 28 June 2007 on organic production and labelling of organic products and repealing Regulation (EEC) No 2092/91.(O.J. L 189, 20/07/2007, p. 1.)

Commission Regulation (EC) No 710/2009 of 5 August 2009 amending Regulation (EC) No 889/2008 laying down detailed rules for the implementation of Council Regulation (EC) No 834/2007, as regards laying down detailed rules on organic aquaculture animal and seaweed production

⁶ Council Directive (EC) No 2006/88 of 24 October 2006 on animal health requirements for aquaculture animals and products thereof, and on the prevention and control of certain diseases in aquatic animals (*O.J. L 328*, 24.11.2006, p. 14–56

2. **Permitted feed sources and feed additives.** A number of countries (France, Italy and the UK) have questioned the requirement in Article 25k that the raw material for fishmeal and fish oil be restricted to (food) fish trimmings and have requested that this be reviewed and that fresh raw fish from sustainable fisheries be permitted, Spain has asked that the availability of trimmings from organic aquaculture and sustainable fisheries be assessed. France would like to clarify that trimmings from crustaceans and molluscs can also be used in addition to trimmings of fish. Likewise, in its comments on the draft mandate, Sweden supports a clarification that trimmings from mussels can be used.

In addition France and Italy have requested that the restrictions in Article 251 (maximum 10% fishmeal and fish oil in ration) be reviewed in order to increase that %, particularly as regards Penaeid shrimps.

Linked to the above mentioned restriction on the use of fresh raw fish, the UK has submitted a dossier for the use of the amino acid histidine as a feed additive for aquaculture feeds; this has been supported by Ireland. Italy has expressed an interest in the issue of ensuring supply of essential amino acids (histidine and others which are not specified) and linked to this refers to the alternative protein sources permitted in conventional aquaculture feed under Commission Regulation (EU) No 56/2013⁷. The opinion of EGTOP on the use of such sources in organic aquaculture feeds it therefore requested. France has requested that conventional sources of lecithin and purified cholesterol be permitted if organic sources are not available.

In preparing its report the group is invited to examine technical dossiers provided to the Commission by the Member States.

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Commission Regulation (EU) No 56/2013 of 16 January 2013 amending Annexes I and IV to Regulation (EC) No 999/2001 of the European Parliament and of the Council laying down rules for the prevention, control and eradication of certain transmissible spongiform encephalopathies (*O.J. L 21, 24.1.2013, p. 3–16*)

4. CONSIDERATIONS AND CONCLUSIONS

4.1. The use of non-organic juveniles

4.1.1 The lack of organic juveniles

State of the art

The Group recognizes the information provided by the MS delegations of Bulgaria, France, Germany, Italy, Romania, Spain and Sweden, on the lack of organic juveniles, as expressed in the Terms of Reference.

Although there are no official data on the number of certified organic hatcheries in Europe, the Group has information on a few hatcheries (e.g. a trout hatchery in Denmark) that have recently converted or are in the process of conversion to organic production (www.eurofishmagazine.com, June 3/2013).

Therefore, the present production of organic juveniles seems inadequate to supply the growing demand of the organic aquaculture industry.

Main difficulties

Besides the lack of organic juveniles, due to the few hatcheries certified as organic, one of the main difficulties experienced by the sector is the restriction on the movement of live animals between countries and regions based on the "Directive 2006/88/EC on animal health requirements for aquaculture animals and products thereof, and on the prevention and control of certain diseases in aquatic animals".

This Directive established five categories of health status in which countries, zones and compartments have to be classified, and rules to be followed for introducing or dispatching animals among areas with different health status classification.

A second barrier to the movements of seed or juveniles among farms is due to the reluctance of farmers to introduce on their farms animals which could be unsuitable for the local (geographical) environment (e.g. genetic or population traits, resistance to different diseases, growth performances, reproductive cycle, behavioural characteristics, etc.).

Furthermore, the Group fully supports the view put forward by Spain concerning the lack in Commission Regulation (EC) No 889/2008 of specific organic rules for managing the life cycle stage between the hatching and the weaning of juveniles. This lack of organic regulation concerns fresh water species (e.g. stocking density, husbandry environment) and, even more, marine species (e.g. phytoplankton and zooplankton production, essential nutrients in the trophic chain, stocking density during larval rearing and weaning, husbandry environment).

The Group considers that production rules for the phase of the life stage between hatching and weaning of juveniles would have a strong influence in determining the characteristics of the adult (e.g. skeletal and pigmentation anomalies, immune resistance, etc.).

Because of the lack of detailed organic rules in this area it is difficult to distinguish organic and non-organic hatcheries.

Conclusions

Owing to the lack of organic juveniles reported by some MS and the main difficulties pointed out above, the Group supports the inclusion of specific rules for the life cycle stage between hatching and weaning of juveniles in the organic regulation, as soon as possible. This would give higher credibility of the rules for both farmers and consumers.

Furthermore, the Group supports the creation of a database on the availability of organic juveniles produced in each country as for the Seed database (ref. Article 48 of Commission Regulation (EC) No 889/2008). Transparency of the use of non-organic juveniles should be ensured in such a database.

Commission Implementing Regulation.(EU) No 1030/2013⁸ of 24 October 2013 postponed to January 1, 2015 the compliance deadline in Paragraph 11 of Article 95 of Commission Regulation (EC) No 889/2008, for those aquaculture and seaweed production units which were established and produced under nationally accepted organic rules before 1 January 2009.

Considering the above issues and the new regulation, the Group supports the use of non-organic juveniles, for on-growing purposes, when organic aquaculture juvenile animals are not available, subject to the following restrictions/recommendations:

- a) Organic juveniles should be used when available.
- b) At least the latter two thirds of the duration of the production cycle shall be managed under organic management (Article 25(e)(2) of Commission Regulation (EC) No 889/2008).
- c) After the approval of specific organic rules for the life cycle stage between hatching and weaning of juveniles, a transitional period should be established to allow farmers to comply with the new rules.

4.1.2. Restocking in lakes, earth ponds of tidal areas and costal lagoons

Historical and traditional extensive aquaculture

Some typical aquaculture practices of extensive fish farming in wetlands, such as brackish water ponds, tidal areas and costal lagoons, closed by levees and banks, have been carried out in Italy and Spain, as well as in other coastal areas of Europe, for many centuries. These traditional extensive farming systems involve the control of the hydraulic circulation and water renewal in lagoons as well as selective fish "seeding".

The management of these areas is mainly aimed at the exploitation of euryhaline species migrating into the coastal lagoons from the sea and backwards, in particular with capture systems placed at the communication channels between the lagoon and the open sea, which enable the selective capture of the different species and, in the more complex systems, of different sizes of the same species.

Nowadays, due to the significant decrease of fish immigration from the sea, specific restocking actions are carried out in several coastal lagoons with wild fry, mainly sea bass, sea bream, mullets and eels. Article 25(e) of Commission Regulation (EC) No 889/2008 does not allow such restocking practices, preventing farmers obtaining organic certification for their production.

Extensive aquaculture inside coastal lagoons has been traditionally developed, over a period of three centuries, especially in Northern Italy, along the Adriatic sea coast, as well as along the Mediterranean and Atlantic coasts of Almeria, Cádiz and Huelva in Spain, the Greek coast and the Black Sea coast of Bulgaria.

Coastal lagoons can be regarded as being relevant also from a historical perspective and as a cultural heritage for coastal lagoon communities concerned. For these communities, extensive aquaculture represents also an activity of common interest in terms of preservation of traditional knowledge and biodiversity conservation.

Nevertheless, the Group is concerned about the catching of juvenile eels for organic eel farming because of the declining population due to over-fishing and environmental pollution and because eels are a critically endangered red list species and captured eels cannot breed.

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Commission Implementing Regulation (EU) No 1030/2013 of 24 October 2013 amending Regulation (EC) No 889/2008 laying down detailed rules for the implementation of Council Regulation (EC) No 834/2007 on organic production and labelling of organic products with regard to organic production, labelling and control (O.J. L 283, 25.10.2013, p. 15–16)

Conclusions

The Group recognizes the high value of these extensive aquaculture practices, in terms of cultural heritage, biodiversity conservation and economic perspective for the local communities.

The Group also recognizes that moving wild fry from the sea into the lagoon does not necessarily affect the stock status of the species concerned.

Therefore, with the exception of eels, the Group considers that restocking of wild fry in the extensive aquaculture farming carried out inside wetlands, such as brackish water ponds, tidal areas and costal lagoons is in line with the objectives, criteria and principles of organic aquaculture production, subject to the following restrictions:

- a) A management plan, approved by the local or national authority, that ensures the sustainable exploitation of the species concerned, should be provided.
- b) In extensive aquaculture farming carried out inside wetlands, such as brackish water ponds, tidal areas and costal lagoons, the fish shall be fed with feed naturally available in the environment.

4.2 Permitted feed sources and feed additives

4.2.1. Dietary requirements of carnivorous fish

Sustainable and environmentally-efficient use of aquaculture feed

Fishmeal of high quality provides a balanced amount of all essential amino acids, minerals, phospholipids and fatty acids reflected in the normal diet of fish (Hardy, 2010; Lund et al., 2012). In particular, a diet based on marine sources secures optimum development, growth and reproduction, especially of farmed larvae and brood-stock. Fish oil is a major natural source of the omega-3 fatty acids eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), which cannot be synthesized by the fish, but are produced by marine phyto- and zooplankton, which are consumed by the wild fish larvae (Baron et al, 2013).

However, the global supply of fish meal and of fish oil is no longer able to meet the increasing demand from an expanding aquaculture industry and, due to the increasing prices of fish meal and fish oil, the aquaculture sector has been forced to look for alternative ingredients including plant products (cf. section 4.2.4) (Gatlin et al., 2007, Hardy 2010).

Replacing fish meal and fish oil is not straightforward due to their unique contents of protein, excellent amino acid profile, high nutrient digestibility, high palatability, adequate amounts of micronutrients, as well as general lack of anti-nutrients in fish meal and the high contents of long-chained polyunsaturated fatty acids in fish oil (Gatlin et al., 2007; Kaushik and Seiliez, 2010; Krogdahl et al., 2010; Lund et al., 2012).

Organic aquaculture is an alternative production approach driven by the growing interest in sustainable utilization of resources (Mente et al., 2011). There is increasing concern about the consumption of fish meal and fish oil. Indeed, the current European regulation on organic aquaculture (Commission Regulation (EC) No 889/2008) does not allow fish meal and fish oil derived from whole fish, but only from trimmings of fish from organic aquaculture or from trimmings of fish already caught for human consumption in sustainable fisheries, in order to reduce the risk that fishing primarily to produce fishmeal will further contribute to fish stock depletion. Further, the regulation does not allow balancing the dietary amino acid profile by supplementing with synthetic free amino acids to fulfil the dietary requirements of the specific organically produced species.

A large number of studies have investigated the effects of replacing fish meal with various plant protein ingredients (Borquez et al., 2011; Glencross et al., 2011; Pratoomyot et al., 2010; Torstensen et al., 2008; Yang et al., 2011). Complete replacement is usually not successful due to problems related to the factors mentioned above and the altered patterns of amino acid uptake

when replacing fish meal with plant based protein ingredients (Bendiksen et al., 2011; Borquez et al., 2011; Espe et al., 2006; Francis et al., 2001; Gatlin et al., 2007; Larsen et. al., 2012; Lund et al., 2011).

High replacement ratios require that anti-nutrients and indigestible substances are efficiently removed from alternative protein ingredients to meet the high protein requirement of fish. Furthermore, it is necessary to ensure that the dietary amino acid profile is optimised, for example by adding free amino acids, and/or by combining several plant protein sources with different amino acid composition (Francis et al., 2001; Kaushik and Seiliez, 2010; Wilson, 2002).

However, as mentioned above supplementation with synthetic amino acids is not allowed according to Council Regulation (EC) No 834/2007 Article. 15 1d. (IV) and currently no amino acids are listed in Annex VI of Commission Regulation (EC) No 889/2008. Furthermore, procedures for the removal of anti-nutrients have to follow organic rules. Finally, there is less availability of relevant organic plant sources to optimize the amino acid profile in comparison to conventional plant sources (Lund et al., 2011; Rembiałkowska, 2007).

Lysine and methionine are often the most limiting amino acids when fish meal is replaced by plant protein sources (Mai et al., 2006). The amino acids which are in excess when the first limiting amino acid runs out will be broken down producing energy and nitrogen (mainly excreted as ammonia with potential adverse environmental impacts) instead of being converted to fish meat. Therefore, a carefully balanced amino acid profile is important for the growth of the fish, as well as the minimization of nitrogen discharge.

The replacement of fish meal by vegetable proteins is further complicated because not only the overall dietary amino acid profile is important for efficient utilisation of amino acids, but also the timing by which amino acids from different protein sources appear in the blood stream after a meal (Larsen et al., 2012)

Larsen et al. (2012) investigated plasma free amino acid concentration patterns in juvenile rainbow trout (*Oncorhynchus mykiss*) fed either a fish meal based diet (FM) or a diet (VEG) where 59% of fish meal protein (corresponding to 46% of total dietary protein) was replaced by a mixture of plant proteins from wheat, peas, field beans, sunflower and soybean. Results showed that the appearance of most amino acids (essential and non-essential) in the plasma was delayed in fish fed the VEG diet compared to those fed the FM diet. Essential and non-essential amino acids furthermore appeared more or less synchronously in the plasma in fish fed the FM diet, while the appearance was less synchronised in fish fed the VEG diet. Further there were 2.7 times more indigestible carbohydrates in the VEG diet than in the FM diet, which suggested that the uptake of amino acids was affected by dietary carbohydrates. In conclusion, the study showed that amino acid uptake patterns were affected when replacing fish meal with plant based protein ingredients.

Further considerations

High quality fish meal with an optimal amino acid profile has a high nutrient digestibility and hence high utilization by the fish that results in minimum discharge of nutrients to the environment.

For larvae and juveniles it is critical to secure optimum feed quality for survival and growth. Similarly for brood stock it is essential to secure optimum egg quality.

Hence, fish meal and fish oil are strategic ingredients to be used at critical stages of the life-cycle, when optimum performance is required.

It should be ensured that the marine ingredients are obtained from sustainable sources, and that the fisheries in question are being managed in compliance with the FAO Code of Conduct for Responsible Fishing. However, the availability and increasing prices of fish meal and fish oil will counteract and limit the inclusion rates of these limited resources and increase the pressure for alternative sources to balance the specific amino acid requirements of farmed fish species.

The issue of how the dietary requirements of carnivorous fish, can be met is addressed in subsequent sections 4.2.3. and 4.2.4.

4.2.2. Dietary requirements of shrimps

Feeding habits

The most important shrimp species in aquaculture are *L. vannamei* (White shrimp), *P. monodon* (Giant tiger shrimp), *M. rosenbergii* (Freshwater prawn) (Valderrama 2011).

Although they are all benthivore species, they have different diets in their natural habitats:

- *M. rosenbergii* is an omnivorous benthivore, and mainly feeds on algae (FAO-FIMA 2011);
- *L. vannamei* is an omnivorous benthivore, and mainly feeds on living preys and detritus (FAO 2011);
- *P. monodon* is a carnivorous benthivore and mainly feeds on worms, crustaceans and molluscs (Tacon 2002, Piedad-Pascual 1984).

These differences in feeding habits are due to the amount of enzymes in the digestive tract of the different shrimps. Carnivorous shrimps have proteolytic enzymes like trypsin and chymotrypsin whereas herbivorous species have more glucolytic enzymes like amylase. This is why carnivorous shrimp have a greater ability to digest protein and herbivorous shrimp have greater ability to digest plant material.

Nutritional requirements

The need for protein varies among species. The life stage of the animal is also important, younger stages have higher needs than older stages (sub-adults and adults) due to the different growth rate (Weir 1998).

According to the available scientific literature, the needs for protein can vary for:

- *P. monodon* between 35 and 50% of the dry matter in feed (Fox et al., 1998; Cousin, 1995; FAO 2011; Dayal et al., 2003; McVey, 1993).
- L. vannamei between 20 and 30% of the dry matter in feed (Velasco et al. 2000; Cruz-Suarez et al. 2000; Kureshy and Davis 2002).
- *M. rosenbrergii* between 30 and 38% of the dry matter in feed (Freuchtnicht et al. 1988; Reed and D'Abramo 1989).

Lipids are also essential components of the diet of shrimps. Lipids are mainly used for direct energy production and cell membrane building.

For *P. monodon*, *M. rosenbergii* and *L. vannamei* the optimal lipid level is between 6 and 8% of the feed dry matter (Alday Sanz 2011; Tiwari and Sahu, 1999), but should not be above 10% (Glencross 2002) or below 2% (Chen, 1998).

Some lipids are more important than others because they cannot be synthesized *de novo* or not in sufficient amounts by shrimps. Phospholipids (e.g. lecithin) and cholesterol are the two main categories of essential lipids for shrimps. They are also used as emulsifiers for lipid digestion (Glencross and Smith, 1998). Without phospholipids in their diet, shrimps are unable to digest lipids properly.

According to the available scientific literature, the need for phospholipids is as follows:

M. rosenbergii – around 5% of the diet (Tiwari and Sahu, 1999);

 $P.\ monodon-1\%$ of the diet for post-larvae (Paibulkichakul et al., 1998) and 1.25% for juveniles (Chen 1993);

L. vannamei, – the requirements for lecithin and cholesterol are linked together.

Cholesterol is a ring compound, which is part of cell membranes and is also necessary in the moulting process (see chapter on cholesterol).

According to the literature, the need for cholesterol varies among the different species of shrimps and according to the different life stages.

- For *M. rosenbergii*, cholesterol need is quite high at 0.3-0.6% of the diet (Sahu, 2004), but this species is able to use phytosterols contained in plants instead of cholesterol as ecdysone precursors, so the amount added in the diet can be reduced significantly (Mitra et al., 2005).
- For *P. monodon*, cholesterol need is lower, but it is crucial and cannot be replaced. Requirements are 1% of the diet for post-larvae (Paibulkichakul et al.,1998) and 0.17% of the diet for juveniles (Smith et al., 2001).
- For *L. vannamei*, there is a relationship between cholesterol and phospholipids. A diet with no phospholipids requires 0.35% cholesterol, whereas a diet with 5% phospholipids requires only 0.05% cholesterol (Gong et al., 2000). A good combination seems to be 0.15% of cholesterol for 1% or more phospholipids.

Conclusions

The Group recognizes the clear differences between shrimp species, their feeding habits and their nutrient requirements. Furthermore, all the above considerations show the need for animal protein and lipids in the diet of shrimps, although in different proportions according to their life stages. Therefore the Group recognises the need for the use of fish meal and fish oil in the diet of shrimps.

Article 25.1 of Commission Regulation (EC) No 889/2008 states: "1. Aquaculture animals as referred to in Annex XIIIa, Section 6, Section 7 and Section 9 shall be fed with feed naturally available in ponds and lakes. ... 3. Where natural feed is supplemented according to paragraph 2 the feed ration of species as mentioned in section 7 and of siamese catfish (Pangasius spp.) as mentioned in section 9 may comprise a maximum of 10 % fishmeal or fish oil derived from sustainable fisheries".

With reference to the above article, the Group supports a limited use of fishmeal and fish oil derived from sustainable fisheries, as a supplement to the feed naturally available in the rearing environment. In the case of shrimps only, such feed rations should not be above 10% for fish oil, as in the current regulation, but could be up to 25% for fish meal.

4.2.3. Fish meal and fish oil from trimmings

According to Commission Regulation (EC) No 889/2008, feeding regimes shall be designed with the following priorities: (a) animal health, (b) high product quality, including the nutritional composition which shall ensure high quality of the final edible product; (c) low environmental impact.

However, the Group is concerned about the consequences of the listed priorities of sourcing feed as laid down in Commission Regulation (EC) No 889/2008 Article 25k.

The Group considers that the levels of phosphorus in the fish meal derived from trimmings might conflict with national environmental legislations, because this may result in too high P-concentrations. Fish meal from trimmings is lower in protein and higher in phosphorus content compared with high quality fish meal (Eurofins; www.ffskagen.dk). The presence of carcass remnants (head, skin, bones) in trimmings also increases the phosphorus content of the fish meal. Using this meal for feeding fish puts limitations on the inclusion level so as to comply with environmental legislation. Danish environmental legislation only allows the phosphorus content of fish feed to be max. 0.9% (max. 1% on dry weight basis) (www.retsinformation.dk/Forms/R0710.aspx?id=140333).

Eurofins (<u>www.eurofins.dk/dk/f0devarer-agro.aspx</u>) has found phosphorus content of traditional fish meal up to 2.2%, while the phosphorus content of trimmings was 2.4%. Based on these findings, using 41% of traditional fish meal in the diet will theoretically result in 0.9% of phosphorus in the diet, not taking into account other potential phosphorus sources. Under the same conditions, using the same amount of trimming-meal would result in a phosphorus content

of 0.99% in the feed for organic aquaculture. Thus, to comply with the environmental legislation, the diet could contain 37.5% trimming meal, while, conventional fish feeds contains about 25% fish meal, and for conventional feeds a long list of alternatives exists, with the diets balanced by supplementing free amino acids.

The challenges are much higher for producing feeds for organic aquaculture because the list of available ingredients is limited and supplementation with synthetic amino acids is not allowed according to Council Regulation (EC) No 834/2007 Article 15(1)(d) (IV) and currently no amino acids are listed in Annex VI of Commission Regulation (EC) No 889/2008.

Fish meal and fish oil from organic aquaculture trimmings are also not allowed in the feed for aquaculture animals of the same species. As a result, only limited quantities of trimmings from organic farming are available. The current organic fish production (excluding shellfish and others) is about 25,000 t (Zubiaurre, 2013). About 50% of this is sold as whole fish from the farm itself, fish shops etc. and the remaining 50% is processed into fillets, yielding about 50-60%, leaving about 40–50% trimmings. The amount of trimmings available for manufacturing of fish meal and fish oil may therefore be about 5–6,000 t. Assuming a yield of fish meal and oil of max. 20% and 6% respectively, this means a production of approximately 1,000 t of fish meal and 300 t of fish oil. Taking the needs of different species into account, these amounts are only sufficient for a very limited organic production and are below the critical level needed for sustainable manufacturing processes.

The manufacturing process to obtain fish meal and oil from trimmings is similar to that of wild caught industrial fish (Sand eel, blue whiting etc.). However, due to the carcass remnants and the little remaining meat, the protein content of the meal from trimmings is 67–70% and the ash content is about 15%. Further, the digestibility is below 90% (pers. com. Claus Christoffersen, FF, Skagen, Denmark), whilst it should be at least 90% in a high quality fish meal.

Carnivorous fish requires relative high dietary protein content, i.e. 38-48% of the diet, depending on fish size, with the highest requirement and quality for fry and brood-stock. This means that, to produce an adequate feed, the inclusion rate of fish meal from trimmings should be high, which conflicts with the limitations of max. 0.9% dietary phosphorus content. Furthermore, the available organic plant sources are limited and their amino acid profiles are not adequately balanced to make an optimum fish feed (Lund et al., 2011). As discussed previously, the breakdown of surplus amino acids is likely to result in increased environmental impact and reduced growth, health and welfare of the fish.

With reference to the specific question asked by some MS, as regards the eligibility of the use of trimming from crustacean and molluscs, the Group's opinion is that such use is not prohibited by the Commission Regulation (EC) No 889/2008.

Conclusions

In the Group's opinion, the animals' need for amino acids and fatty acids should be met primarily through natural feed compounds. Fishmeal and fish oil are important components of this, particularly for carnivorous aquaculture animals, which have specific amino acid and other nutritional requirements.

However, the Group concluded that, for carnivorous fish, this is not possible using fish trimmings alone, due to its lower quality.

In conclusion, the Group's opinion is that in order to comply with the general rules on feed (cfr. Article 25(j) of Commission Regulation (EC) No 889/2008), namely: "... optimum performance, animal health, high product quality, including the nutritional composition which shall ensure high quality of the final edible product and low environmental impact", the diet for carnivorous fish should include fish meal derived not only from trimmings but also from whole fish, not used for human consumption, caught in sustainable fisheries. This includes feed for fry and broodstock, as well as for on-growing fish, until sufficient alternative sources of proteins and oils are available.

4.2.4. Alternative protein sources

State of the art

Since fish meal and fish oil are limited resources, finding alternative protein sources for fish meal is clearly a high priority for organic aquaculture. Similarly, developing production of aquafeeds that: a) satisfy aquatic organism's nutritional requirements for specific amino acids and fatty acids, b) are suited to their feeding habits and c) result in a high retention of nutrients to maintain animal health and produce a good quality final product, is a high priority. Improvements in protein retention efficiency of farmed aquatic animals are needed to reduce any potential environmental impacts of organic aquaculture and, also, to make more efficient use of dietary protein, the most expensive component of diet formulations for fish. Research into alternatives to fish meal is now an international research priority. A "metabolically" optimized protein and lipid diet formulation per species and per life cycle of farmed aquatic animal is the focus of current aquatic animal nutrition research.

Alternative proteins sources are needed to replace fishmeal, especially for diets of carnivorous species. Plant proteins (soybean, rapeseed, corn gluten, wheat gluten, pea and lupin meals) can replace fishmeal up to 25–35% (Pereira and Oliva-Teles, 2003; Hardy 2010; Enami 2011). The feed ration may comprise a maximum of 60% of organic plant products (Commission Regulation (EC) No 889/2008, Article 25(k)(3)). Mente et al., (2011) reviewed nutrition in organic aquaculture and organic diets. The review showed that an organically certifiable yeast-based protein source could replace up to 25% of the fishmeal without affecting growth rates, feed efficiency or biological indices in cobia fish. Substitution levels above this resulted in decreased performance in all measured parameters. Fishmeal and soybean meal were replaced with an organic diet (yeast), and there was no difference in growth rates in tilapia. Another study replaced fishmeal with a plant-based diet (algal fermentation), in shrimps Litopenaeus vannamei, and showed that there were no significant difference in final production, survival and food conversion ratios (FCR). However, fishmeal diet deposited more 22:6n-3 (docosaheaxaenoic acid, DHA) in shrimp's tissues in comparison with the plant-protein diet. The effect of organic fertilization and organic diets on production of channel catfish in earthen ponds was also investigated.

Organically cultivated seaweed or sustainably harvested wild seaweed, including all multicellular marine algae or phytoplankton and microalgae, may be used as feed ingredient.

Other potential sources of proteins, such as wild-harvested and/or cultured annelid worms, insect larvae/pupae, gastropods (e.g. golden apple snail) is also considered promising in order to replace fishmeal in the future (Bergleiter et al. 2009).

Processed animal protein (PAP) is an important ingredient in feeds and provides a valuable source of animal by-product utilization. PAP has a high nutritional value making it an excellent alternative to imported proteins such as soya. It has a significantly higher protein value (45-90% on a fed basis) than plant feed ingredients. PAP contains 10 % phosphorus, which is low in relation to the content of amino acids. While there may be consumer and producer concerns about the feeding of PAP to fish, due to the potential transmission of prions, the scientific panel opinion published by the European Food Safety Authority (EFSA) in 2011 concluded that processed animal protein in feed for food producing non-ruminants, respecting the proposed ban on intra-species recycling, presents a negligible risk to human health (EFSA, 2011).

The use of insects as a source of protein in fish diets is also being explored. The nutritive value of insects as feeds for fish, poultry and pigs has been recognised for some time in China, where studies have demonstrated that insect-based diets are cheaper alternatives to those based on fish meal. The insects used are the pupae of silkworms (*Bombyx mori*), the larvae and pupae of house flies (*Musca domestica*) and the larvae of the mealworm beetle, *Tenebrio molitor*. Silkworm pupae are an important component of cultured carp diets in Japan and China. Dried ground

soldier fly larvae have been fed to chickens and pigs with no detrimental effects (Newton et al., 1977; Hale, 1973). In recent years there has been some interest in the use of housefly maggot meal as a substitute for fish meal in tilapia and African catfish diets (Adesulu and Mustapha, 2000; Fasakin et al., 2003; Ajani et al., 2004; Ogunji et al., 2006). Bondari and Shepherd (1987) observed that channel catfish and blue tilapia fed on soldier fly larvae for 10 weeks were acceptable as food by consumers. Growth and organoleptic quality were not affected when common carp were fed on non-defatted silkworm pupae, a major by-product of the sericulture industry in India (Nandeesha et al., 2000). Ng et al. 2001demonstrated that *T. molitor* larvae meal was highly palatable to the African catfish (*Clarias gariepinus*) and could replace up to 40% of the fish meal component without reducing growth performance.

St-Hilaire et al. (2007) describe a study in which they determined if black soldier fly (*Hermetia illucens*) pre-pupae and housefly pupae could be used as a partial replacement for fish meal and fish oil in rainbow trout (*Oncorhynchus mykiss*) diets. Their data suggest that a rainbow trout diet in which black soldier fly pre-pupae or housefly pupae constitute 15% of the total protein has no adverse effect on feed conversion efficiency over a 9-week feeding period. However, rainbow trout fed on black soldier fly diets low in fish oil had reduced levels of omega-3-fatty acids in the muscle. According to the researchers, modifying the diet of the fly larvae could improve digestibility and fatty acid content of the pre-pupae, which in turn could enhance the fatty acid profile of the fish fed on the fly pre-pupae. The use of the black soldier fly in manure management, yields abundant numbers of fly pre-pupae. The authors of the study suggest that fly pre-pupae may be an economical and sustainable feed ingredient for carnivorous fish diets. However, before fly pre-pupae can be used commercially in rainbow trout diets, a larger trial over a longer period should be conducted to confirm their preliminary results. The CAB Abstracts database contains some 700 records describing research on alternative protein sources for use in aquafeeds.

Main difficulties

Plant proteins are probably the most widely used alternative to fishmeal, but they can cause problems, including lower crude protein levels, palatability issues, amino acid deficiencies and the occurrence of anti-nutritional factors such as trypsin inhibitors, as well as phosphorus and nitrogen release to the environment (Hardy and Tacon 2002). The quality of the plant ingredients that can be used in the organic diets can have an effect on the final product quality like fat content, colour and texture (Lunger et al. 2007). Furthermore, research has not determined clearly the proportions of protein of animal and plant origin that should be used in organic feed. The formulation of "ideal protein contents" and how to increase dietary energy levels also need to be explored. The potential use of alternative proteins in feeds for organic aquatic animals should be further investigated over larger trials and longer periods per species and per life cycle to confirm these preliminary results.

Conclusions

PAP, insects and plant ingredients could be used in organic aquaculture up to different species-specific percentages, but consideration on the final product quality and taste is important and needs further investigation. The difficulties in formulation of test diets to secure acceptability to fish, maximize growth of fish and nutrient retention efficiency are the most important factor in using alternative protein sources. New organic feeds with alternative organic protein sources, as well as evidence on the effects of these on fish growth and fish physiology and health performance are needed for organic aquaculture.

The Group supports the importance of considering the use of alternative proteins sources in organic aquaculture, whenever they are available and appropriate for brood-stock, weaning, and on-growing diets. These alternative organic protein sources should be considered a priority. In general the use of feedstuffs consisting of whole micro or macro organisms with high contents of

essential amino acids is to be preferred to the use of purified or free amino acids as feed supplements/additives.

4.2.5. Histidine

Identification of substance, terminology, synonyms

Histidine (His) is an essential amino acid ($C_6H_9N_3O_2$) which cannot be synthesized by fish or any other aquatic animal and therefore must be supplied in the diet.

His is a very important precursor for the synthesis of proteins, vitamins (e.g. Vit C) and enzymes and it plays a vital role in the structure and binding functions of haemoglobin.

Synonyms of L-Histidine: 2-amino-3-(3H-imidazol-4-yl) propanoic acid; Imidazole alanine; α-amino-1H-imidazole-4-propanoic acid; 4-(2-amino-2-carboxyethyl)imidazole; α-amino-1H-imidazole-4-propionic acid; α-amino-4-imidazolepropionic acid; Glyoxaline-5-alanine (Ref: http://www.chem.gmul.ac.uk/iupac/AminoAcid/).

The original request is to use His of 98% purity and produced by a specific strain of bacteria as an additive in the nutrition of organic salmonid fish species. The Group does not want to restrict its findings to a specific commercial product, and has therefore widened the scope of this discussion to "histidine produced by fermentation", without restrictions on purity or microbial species.

Authorization in general agriculture/aquaculture or feed/food processing

The use of His as an additive in the nutrition of salmonid fish is authorized under EU legislation (Commission Regulation (EC) No 244/2007⁹). His is also positively evaluated as a feed ingredient in salmonid diets by EFSA (2005).

Technological or physiological functionality for the intended use

Amino acids are the building block of proteins. The various proteins are built up of amino acids in specific patterns and sequences. All amino acids resemble each other to a certain extent, some of them so much that fish can synthesize them from others. Some, however, are essential as they cannot be synthesized by the fish itself. They must therefore be supplied in the feed, since each amino acid has its own specific function in the fish (Wilson, 2003).

The amino acid profile of the feed, e.g. the proportions and the amounts of various amino acids, must be balanced with the protein requirements of the fish to be utilized efficiently.

Therefore the essential amino acids in the diet must be in adequate amounts. As discussed previously, if one amino acid is limiting, other surplus amino acids will be broken down and excreted

Therefore, a carefully balanced amino acid profile is critical for the growth of the fish, as well as the minimization of nitrogen discharge as ammonia.

An unbalanced diet recipe for *conventional* aquaculture feed may be balanced by adding specific external amino acids, e.g. His, to ensure optimum utilization of the other dietary amino acids present. As an essential amino acid His has to be provided in adequate amounts in any feed for aquaculture. The provision of His has been shown to prevent cataracts in salmonids. Cataract is permanent lens opacity of both eyes (Waagbø et al, 2010).

Commission Regulation (EC) No 244/2007 of 7 March 2007 concerning the authorisation of L-histidine monohydrochloride monohydrate as a feed additive (O.J. L 73, 13.3.2007, p. 6–8)

Necessity for intended use and known alternatives

His is an essential amino acid, which is very important for fish diets. Salmonids have a particular need for His, in order to prevent cataracts. Klein and Halver (1970) indicated that coho salmon, has a minimum His requirement of 0.7% of a dry diet or 1.7% of the dietary protein or 7 g His/kg diet (40% dietary protein) whereas Waagbø et al. (2010) indicated that 17.6 g His/kg of the diet mitigated cataract formation in salmon smolts (37% dietary protein). However, at present, it is still unclear how His prevents or mitigates cataract development and the molecular basis of cataractogenesis in salmon.

Marine raw materials vary significantly in composition and quality according to species and season, as well as to the production, processing and storage conditions. This variation in quality affects both extruded feed quality and fish performance. Certain types of fish meal from South America also contain high levels of His but this is only available from whole fish, which is banned by the Article 25(k) of Commission Regulation (EC) No 889/2008.

Up to 1995, animal by-products have been used widely in fish feeds, and supplied sufficient levels of His (e.g. blood meal from non-ruminants contains sufficient levels of His). However, the use of PAP (Processed Animal Protein) has been banned in the EU and only recently has been re-admitted with the Commission Regulation (EC) No 56/2013. In addition, at present, the availability of PAP originated from organic farming is not known. Nevertheless, many consumers may be reluctant to accept the use of blood meal as a fish feed ingredient. Therefore further research is needed which aims: a) to identify and study protein feed resources e.g. insect meal in accordance with the organic principles and the animal's health, b) to identify differences between fish genotypes and c) to study consumer preferences in relation to feeds used in organic aquaculture.

Origin of materials, methods of manufacture

L-histidine monohydrochloride monohydrate can be produced in a fermentation process using a natural strain of Escherichia coli. Production of the raw material takes place in fermenters, and then undergoes several steps of purification. The fermentation medium used is a mixture of glucose syrup, ammonium sulphate, corn steep liquor and minerals. His is isolated from the fermentation broth, acidified, purified (including an ion exchange procedure) and de-colourised. After addition of hydrochloric acid the solution is concentrated and L-Histidine·HCl·H₂O is crystallized and dried (EFSA 2005). His produced by fermentation can be considered as a natural substance of microbial origin.

The strain ATCC-21318 of the bacterium *Escherichia coli*, which is currently used for the production of His, is not a GMO (EFSA 2005). In addition, it is possible to produce His with a non-GM fermentation medium, although this would not be required according to Article 9 of Council Regulation (EC) No 834/2007.

However, other potential commercial production approaches should be evaluated separately.

Environmental issues

A balanced fish diet is crucial for nutrition, growth, health and welfare, as well as for environmental reasons. Unbalanced diets, i.e. amino acid profile of the diet which is not balanced to the requirements of the fish species, will result in bad protein utilization, which means break down of proteins and excretion of nitrogen as ammonia. Therefore, a balanced amino acid profile of fish diets is crucial for the organic aquaculture. EFSA (2005) identified no adverse influence on the environment by production and feeding with His of microbial origin.

Animal welfare issues

Fish health and welfare is largely dependent on a well-balanced diet with appropriate proportions of nutrients, including amino acids. His deficiency causes disease, mainly cataract. His supplementation has been shown to prevent cataract in salmon (Waagbø et al., 2010). Supplementation of His is, therefore, clearly positive from the point of view of animal health and welfare.

Human health issues

His is an essential amino acid in human nutrition as well. EFSA (2005) identified no risks for consumers, and no specific risks for workers and users.

Food quality and authenticity

Food quality and authenticity, in the sense of final fish product for human consumption, is not supposed to be affected by supplementation of His in the fish diet.

Traditional use and precedents in organic production

Materials of microbial origin are widely authorized in organic production. This includes their use as feed materials (Annex V) and as feed additives (Annex VI) in many areas of organic production. The following items are listed in the Commission Regulation (EC) No 889/2008: phaffia yeast Article 25(K); mushroom culture wastes, composted or fermented mixture of vegetable matter (Annex I); spinosad (Annex II); Saccharomyces cerevisiae and S. carlsbergiensis (yeasts) (Annex V); enzymes, yeasts and bacteria (Annex VI); yeasts (Article 27(1)(b) and Annex VIIIa).

Aspects of international harmonization of organic farming standards

At the moment, neither the Codex Alimentarius Guidelines for the production, processing, labelling and marketing of organically produced foods (GL 32-1999, last amended 2013) nor the National Organic Program (USA) cover aquaculture.

Further considerations

All the above considerations show how His supply is important in the nutrition of salmonid fish. By definition, the same is true for all essential amino acids. Lysine (Lys) and arginine (Arg) requirements in fish nutrition are considered to be the first limiting amino acids in fish and the level of Lys, Arg and methionine (Met) in plant-protein meals are often low, compared to fish meal. In fish, it has been demonstrated that Lys and Arg supplementation may enhance protein synthesis and deposition, growth and reduce nitrogen losses (Kaushik, S.J. and Seiliez, I., 2010). However, the metabolic pathways by which dietary essential amino acids influence growth performance require further investigation. Fish and crustaceans have high dietary protein requirements (30–65% DM; NRC, 1993). Significant inter-species differences with regard to protein and amino acid requirements exist in fish. The variations in amino acid requirements can be attributed to a number of factors, such as differences in basal diet composition, size and age of fish, genetic differences, feeding rate and culture conditions and experimental design and choice of response criterion, all of which affect the overall growth rate, health and welfare.

The application of natural compounds derived from a variety of sources including yeasts, botanical peptides and animal by-products is at the top of the agenda of the researchers in the different fish species. Indeed, natural compounds can contribute to bactericidal action, digestive stimulation, immune stimulation, anti-oxidants, anti-parasitic effects in aquatic species and their use as alternatives to antibiotics and synthetic products is growing in the feed industry.

Finally, the Group considers that a better supply of essential amino acids would also improve the welfare and health of other aquatic species and that the supplementation of His should not be restricted to salmonids.

However, it should be outlined that the Group does not support the use of His to neutralize inadequate management practices. In the Group's opinion, animals should cover their need for amino acids primarily through the feed. Therefore, the Group recommends that supplementation with a preferably un-purified histidine source, additive or feed material, should only be used to balance the dietary composition of amino acids to avoid deficiency diseases (mainly cataracts in salmonids). The Group does not accept this practice in cases where the only justification is faster growth and/or a lower price of the feed.

Conclusions

The Group concluded that the essential amino acid histidine is very important for fish diets, particularly for salmonids. Therefore, if natural sources of histidine, such as certain types of fish meal, are not allowed/available, the use of supplementary histidine sources in the fish diet is considered in line with the objectives, criteria and principles of organic aquaculture production. Histidine should therefore be included in Annex VI of Commission Regulation (EC) No 889/2008 with the following priorities:

- 1. Use of un-purified fermentation products as a feed stuff, produced by organic procedures where possible.
- 2. Use of purified histidine as a feed additive only if option 1 is not possible.

4.2.6. Cholesterol

Identification of substance, terminology, synonyms

Cholesterol ($C_{27}H_{45}OH$) is a lipid (a sterol), composed of four carbon rings. In addition to its importance within cells, cholesterol also serves as a precursor for the biosynthesis of steroid hormones, bile acids, and vitamin D. Cholesterol is the principal sterol synthesized by animals. The Group was asked whether conventional cholesterol could be used.

Authorization in general agriculture or feed/food processing

Cholesterol is considered to be a raw material for feedstuffs and is therefore not in the list of feed additives. Cholesterol is also used in cosmetic products, like makeup, skin and hair care products. It is used as an emulsifier, as well as to increase the viscosity of personal care products (www.cosmeticsinfo.org; http://www.dishmangroup.com/index.asp; http://www.solvayvitamins.nl/).

Technological or physiological functionality for the intended use

Moulting is an essential process for growth of crustaceans. Cholesterol is a precursor of ecdysone, a steroid prohormone needed for moulting. For some species, it is the only precursor and hence it is very important to meet the cholesterol requirements for shrimps and other crustaceans. Dietary cholesterol deficiency may cause reduced growth rates leading to low survival and even death. The process involving cholesterol in the growth of shrimp is similar to vitamins in mammals (Williams et al., 2004). Cholesterol is crucial for many physiologically active compounds including adrenal corticoids, bile acids and vitamin D (Akiyama et al., 1992; Sheen et al., 1994). However, it does not increase growth rates.

Necessity for intended use and known alternatives

To the Group's knowledge, shrimps are the only aquatic animal species that require dietary cholesterol (see section 4.2.2). Crustaceans are not able to synthesize cholesterol *de novo*. Cholesterol is necessary for the nutrition of crustaceans because, it has a crucial role in the moulting process and it is important in maintaining the integrity and chemical permeability of

cell walls. Therefore it is crucial to have cholesterol in their feed (Teshima and Kanazawa, 1971). A dietary cholesterol deficiency is most commonly manifested as a reduced growth rate (reviewed by Teshima, 1997). According to the literature, the requirements of cholesterol vary among the different species of crustaceans and according to their life stage.

- For *M. rosenbergii*, cholesterol needs are high: 0.3-0.6% of the diet (Sahu, 2004) but this species is able to use phytosterols contained in plant instead of cholesterol as ecdysone precursors, so the amount added in the diet can be reduced significantly (Mitra et al., 2005).
- Cholesterol requirement for *P. monodon* is lower, but it is crucial and cannot be replaced. Needs are 1% of the diet for post-larvae (Paibulkichakul et al.,1998) and 0.17% for juveniles (Smith et al., 2001). The survival rate of sub-adult shrimps fed with 0.02% and 0.95% of cholesterol in the diet was only 13.3% and 33.3%, respectively, after two months. While, survival rate for shrimp fed diets containing amounts of cholesterol between the two above mentioned levels was between 83.3% and 93.3% (Sheen et al.,1994).
- For *L. vannamei*, there is a relationship between cholesterol and phospholipids. Indeed, a diet with no phospholipids requires 0.35% of cholesterol, whereas a diet with 5% of phospholipids requires only 0.05% of cholesterol (Gong et al., 2000). A good combination seems to be 0.15% of cholesterol for 1% or more phospholipids.

For those shrimp species that require a cholesterol-rich diet, it may be necessary to supplement the feed. A feed with 10% of fishmeal (the maximum allowed by the Commission Regulation (EC) No 889/08) contains an average of only 0.05% cholesterol. With such a low level, there is a high risk of mortality for sub adults shrimps and even higher for post-larval stages. Furthermore, it is very difficult to predict the quality of the natural feed produced by the environment where shrimps are reared (e.g. ponds) and the quantity consumed. Farmers, on the other hand, cannot only rely on annual fluctuations of the natural productivity of the rearing shrimp ponds.

Origin of materials, methods of manufacture

Natural cholesterol is produced from only one source so far, i.e. it is extracted from sheep wool grease, but the yields are very limited. Other potential sources of cholesterol are egg yolk, animal by-products, shellfish (Dong, 2009).

Cholesterol is extracted from lanolin, which comes from sheep wool grease, in a multistep extraction procedure. The lanolin can be refined to produce pure cholesterol (91%), which is added into the feed for shrimp.

Environmental issues

Cholesterol is a by-product of wool industry and it is not considered harmful to the environment in its production process. Its inclusion in the feed for shrimps is not considered to cause any changes in the pond water quality.

Animal welfare issues

Dietary cholesterol is considered an essential nutrient for good growth, including moulting and high survival in crustaceans. Only adequate dietary contents will secure growth performance and welfare of the animal. In fact, less than 0.02% and more than 0.95% of cholesterol in the diet of sub-adults caused mortality (Paibulkichakul et al 1998; Smith et al 2001; FAO-FIMA 2011; Alday-Sanz V. 2011; Piedad-Pascual, F. 1984; Chen 1998).

Human health issues

Supplemented cholesterol in the feed for crustaceans is necessary for the moulting process and it is not supposed to be detected in the final product. Therefore in this context cholesterol is not considered to have any impact on human health.

Food quality and authenticity

Supplementation with cholesterol in the shrimp diet does not affect food quality of the final product for human consumption.

Traditional use and precedents in organic production

According to the French National organic regulation, cholesterol is authorized as follows:

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Annex P-1

PART A – Raw materials or simple feeds

4) Specific products for shrimp diet: -Cholesterol.

Cholesterol can be utilised under the following conditions

- Cholesterol is purified at 85% and comes from wool grease;
- Guarantee to provide: Flocks are free of scrapie.

This has been extended by the Commission Implementing Regulation (EU) No 1030/2013 until January 2015 to those aquaculture and seaweed production units which were established and produced under nationally accepted organic rules before 1 January 2009. The ad-hoc expert group on "Use of certain fish feed additives and cleaning substances in organic aquaculture" held in Brussels on 19 and 20 November 2008 concluded that cholesterol may be used in organic crustacean farming, preferentially using organic sources.

Aspects of international harmonization of organic farming standards

At the moment, neither the Codex Alimentarius Guidelines for the production, processing, labelling and marketing of organically produced foods (GL 32-1999, last amended 2013) nor the National Organic Program (USA) cover aquaculture.

Further considerations

The Group acknowledges that a feed with an adequate supply of cholesterol would secure the growth rate, the welfare and health of the crustaceans. However, it should be outlined that the Group does not support the use of cholesterol to neutralize inadequate management practices. In the Group's opinion, animals should cover their need for cholesterol primarily through the feed. Therefore, the Group recommends the supplementation of cholesterol only to secure the quantitative dietary needs in the feed for crustaceans. By contrast, the Group does not accept this practice in cases where the only justification is faster growth and/or a lower price of the feed. However, establishment of an organic procedure for the extraction of cholesterol from wool is needed.

The ad-hoc expert group on "Use of certain fish feed additives and cleaning substances in organic aquaculture" held in Brussels on 19 and 20 November 2008 concluded: Cholesterol may be used in organic crustacean farming. The source of cholesterol should follow the priority list given below:

- a) For preference, sustainable marine sources or organically certified sources such as highly purified cholesterol from sheep's wool should be used.
- b) If unavailable, non-organic natural sources may be used.

To clarify the availability of organic cholesterol, a market analysis should be carried out. This would provide the data necessary for an evidence-based decision that organic cholesterol is not available in sufficient quantities.

Conclusions

The Group concluded that the use of cholesterol as raw material in the feed for supplementing the diet of shrimps is in line with the objectives, criteria and principles of organic production. It should, therefore, be admitted by the Commission Regulation (EC) No 889/2008 with the following priorities:

- 1. If organic cholesterol is available, this source should be used as priority.
- 2. As an alternative, cholesterol derived from shellfish and other sources can be used when no organic or wool-derived cholesterol is available.

4.2.7. Lecithin

Introduction

The Group was asked whether conventional sources of lecithin could be permitted, if organic sources are not available. This topic has been addressed previously by an ad-hoc expert group on the "use of certain fish feed additives and cleaning substances in organic aquaculture" (Brussels, 19 and 20 November 2008). Because of this, the Group only dealt with selected aspects.

Authorization in organic production

Lecithin is currently authorized by Commission Regulation (EC) No 889/2008, as reported in the following Annexes:

ANNEX II (Pesticides - plant protection products referred to in Article 5(1)), as a Fungicide; ANNEX VI (Feed additives used in animal nutrition referred to in Article 22(g), Article 24(2) and Article 25(m)(2)), (c) Emulsifying and stabilising agents, thickeners and gelling agents, only if derived from organic raw material. Use restricted to aquaculture animal feed; ANNEX VIII A as additive for all plant based products and for animal products restricted to Milk products. Based on Article 27(1)(a) of Commission Regulation (EC) No 889/2008.

Aspects of international harmonization of organic farming standards

At the moment, neither the Codex Alimentarius Guidelines for the production, processing, labelling and marketing of organically produced foods (GL 32-1999, last amended 2013) nor the National Organic Program (USA) cover aquaculture.

In the rules for food processing, the National Organic Program states that organic forms of lecithine must be used. As an exception, non-organic de-oiled lecithin may be used only when an organic form of de-oiled lecithin is not commercially available (NOP Rule 205.606). The details are as follows:

Lecithin de-oiled

Status: Allowed with Restrictions

Class: Processing Agricultural Ingredients and Processing Aids

Origin: Non-synthetic

Description: Non-organic de-oiled lecithin may be used in processed products labelled as 'Made with Organic [specified ingredients]' provided that the de-oiled lecithin is not produced or handled with the use of sewage sludge, genetic engineering, genetically modified organisms (GMOs), or ionizing radiation. Nonorganic de-oiled lecithin may also be used in or on processed products labelled as 'organic' only when the certifier determines that the ingredient is not commercially available in an organic form and that it meets the requirements of 205.301(b) and 205.301(f).

Necessity for intended use

Lecithin is one of the two main categories of essential lipids, and is therefore necessary for shrimp aquaculture. Details are described in section 4.2.2.

Reflections of the Group

The ad-hoc expert group on "Use of certain fish feed additives and cleaning substances in organic aquaculture" held in Brussels on 19 and 20 November 2008 concluded: "... For preference, lecithin from organically certified sources such as organic soybean, may be used following mechanical extraction. If unavailable, non-organic natural sources may be used provided they are of non-GMO origin".

The applicants did not provide detailed quantitative documentation about a shortage of organic lecithin. As lecithin is a by-product of the oil industry, the potential availability of organic lecithin can be estimated from the trade volume of organic oils. Based on the information available to the Group, global production of organic lecithin is about 2000 t/y, and greatly exceeds the demand.

To further clarify the availability of organic lecithin, a market analysis should be carried out. This would provide the data necessary for an evidence-based decision on the availability of organic lecithin in sufficient quantities.

The use of lecithin for organic flavourings in the food industry will be discussed in the EGTOP report on food (II). In the Group's opinion, the use of conventional vs. organic lecithin should be regulated in the same way in food and in feed.

Conclusions

On the basis of the above information, the Group reconfirms the advice provided by the ad-hoc expert group in 2008.

4.2.8. General conclusions and recommendations

Article 25(k) of Commission Regulation (EC) No 710/2009¹⁰ states "Feed for carnivorous aquacultural animals shall be sourced with the following priorities:

- a) Organic feed products of aquaculture origin;
- b) Fishmeal and fish oil from organic aquaculture trimmings;
- c) Fishmeal and fish oil and ingredients of fish origin derived from trimmings of fish already caught for human consumption in sustainable fisheries;
- d) Organic feed materials of plant origin and of animal origin as listed in Annex V and the restriction laid down therein complied with.

In the Group's opinion, aquaculture animals should cover their needs for amino acids and fatty acids primarily through the natural compounds of the feed. In order to comply with the general rules on feed (cfr. Article 25(j) of Commission Regulation (EC) No 889/2008), namely: "...animal health, high product quality, including the nutritional composition which shall ensure high quality of the final edible product and low environmental impact", the diet for carnivorous fish should be characterized by a well-balanced proportion of amino acids, fatty acids and lipids.

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Commission Regulation (EC) No 710/2009 of 5 August 2009 amending Regulation (EC) No 889/2008 laying down detailed rules for the implementation of Council Regulation (EC) No 834/2007, as regards laying down detailed rules on organic aquaculture animal and seaweed production (O.J. L 204, 6.8.2009, p. 15–34)

The Group also recognizes the specific needs of animal protein and lipids in the diet of shrimps, although in different proportion according to life stages. Therefore, the Group supports a limited use of fishmeal and fish oil derived from sustainable fisheries, as a supplement of the feed naturally available in the rearing environment of shrimps. Such feed rations could be up to 10% for fish oil and up to 25% for fish meal.

The Group is concerned about the consequences of the listed priorities of sourcing feed as laid down in Commission Regulation (EC) No 889/2008 Article 25(k), reported at the beginning of this page. Indeed, with respect to fish meal derived from trimmings, the risk is that the levels of phosphorus contained in such fish meal might result in conflicts with national environmental legislation.

Since fish meal and fish oil are a limited resource, finding alternative protein sources to replace fish meal is clearly a high priority for organic aquaculture. Similarly, developing production of aquafeeds that: a) satisfy aquatic organisms' nutritional requirements for specific amino acids and fatty acids, b) suit their feeding habits and c) result in a high retention of nutrients to maintain animal health and to achieve good quality final products is a high priority. As a consequence, the Group supports the use of alternative proteins sources in organic aquaculture, when available and appropriate for brood stock, weaning, and on-growing diets. The development of organic alternative protein sources should be considered a priority.

Considering all these issues, the Group concluded that the following alternative options should be considered, in order of priority:

- a) Besides fish meal and fish oil derived from trimmings of fish, crustaceans and molluscs, also fish meal and fish oil derived from "whole fish not used for human consumption", caught in sustainable fisheries, should be allowed as ingredients in feed for organic carnivorous fish. This includes feed for fry and brood-stock, as well as for on-growing fish, until sufficient alternative sources of fish meal and fish oil are available.
- b) In addition, the use of other alternative feed materials consisting of whole micro or macro organisms with high contents of essential amino acids and lipids, where possible produced organically, may be needed and are to be preferred to the use of purified or free amino acids as feed supplements/additives.
- c) If not available from organic procedures, essential amino acids and lipids obtained by fermentation or other similar procedures should be allowed as ingredients/additives in carnivorous fish feed only if specifically authorised.

In the case of histidine, the approval in the specific context recommended by the Group should not be seen as a precedent for the use of histidine as a feed additive outside aquaculture, nor for the use of other free amino acids as feed additives for any type of livestock. Other uses / substances should be evaluated separately

5. MINORITY OPINION

In the conclusions of the paragraph 4.1.2. Restocking in lakes, earth ponds of tidal areas and costal lagoons is stated: "the Group recognizes the high value of these extensive aquaculture practices, in terms of cultural heritage, biodiversity conservation and economic perspective for the local communities."

Later in the paragraph, is stated: "Therefore, with the exception of eels, the Group considers that restocking of wild fry in the extensive aquaculture farming carried out inside wetlands, such as brackish water ponds, tidal areas and costal lagoons is in line with the objectives, criteria and principles of organic aquaculture production, subject to the following restrictions:

a) A management plan, approved by the local or national authority, that ensures the sustainable exploitation of the species concerned, should be provided. ..."

The Group did not find, however, an agreement in relation to the <u>exclusion of the eels</u> from the restocking practices considered in line with the objectives, criteria and principles of organic aquaculture production. The majority of the Group supported the need for such an exception because the eel is considered an overfished/endangered species.

The following is the minority opinion expressed by the Chair of the EGTOP Group Mr Giuseppe Lembo.

Considering that everyone in the Group agrees "... that restocking of wild fry in the extensive aquaculture farming carried out inside wetlands, such as brackish water ponds, tidal areas and costal lagoons is in line with the objectives, criteria and principles of organic aquaculture production, subject to the following restrictions: a) A management plan, approved by the local or national authority, that ensures the sustainable exploitation of the species concerned, should be provided.". Furthermore, considering that there is a general agreement on the overfished/endangered condition of the eel, it is likely that the reasons for the disagreement among the Group arises from a lack of a thorough knowledge about the role assigned to the "Management Plans" by the European Regulations.

Indeed, specific provision on management plans are included in the European regulations, both in general terms (in the context of the CFP), and with specific reference to the eel.

Management plan in the CFP

Management plans were previously regulated by the Council Regulation (EC) No 2371/2002¹¹ on the conservation and sustainable exploitation of fisheries resources under the Common Fisheries Policy.

In particular, the following provisions were included in the Article 6 of Council Regulation (EC) No 2371/2002:

- "... 2. Management plans shall include conservation reference points such as targets against which the maintenance of stocks within such limits shall be assessed. ...
- 3. Management plans shall be drawn up on the basis of the precautionary approach to fisheries management and take account of limit reference points recommended by relevant scientific bodies. They shall ensure the sustainable exploitation of stocks and that the impact of fishing activities on marine eco-systems is kept at sustainable levels. ... The management plans shall be multi-annual and indicate the expected time frame for reaching the targets established".

Council Regulation (EC) No 2371/2002 has been then replaced by the Regulation (EU) No 1380/2013 of the European Parliament and of the Council of 11 December 2013¹². This new

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Council Regulation (EC) No 2371/2002 of 20 December 2002 on the conservation and sustainable exploitation of fisheries resources under the Common Fisheries Policy (O.J. L 358, 31.12.2002, p. 59–80)

Regulation, however, does not change the previous direction in favour of conservation reference points.

In particular, the following provisions are included in the Article 9 of the Regulation (EU) No 1380/2013:

- "1. Multiannual plans shall be adopted as a priority, based on scientific, technical and economic advice, and shall contain conservation measures to restore and maintain fish stocks above levels capable of producing maximum sustainable yield in accordance with Article 2(2)." The provision of article 2 are:
- "1. The CFP shall ensure that fishing and aquaculture activities are environmentally sustainable in the long-term and are managed in a way that is consistent with the objectives of achieving economic, social and employment benefits, and of contributing to the availability of food supplies.
- 2. The CFP shall apply the precautionary approach to fisheries management, and shall aim to ensure that exploitation of living marine biological resources restores and maintains populations of harvested species above levels which can produce the maximum sustainable yield. In order to reach the objective of progressively restoring and maintaining populations of fish stocks above biomass levels capable of producing maximum sustainable yield, the maximum sustainable yield exploitation rate shall be achieved by 2015 where possible and, on a progressive, incremental basis at the latest by 2020 for all stocks.
- 3. The CFP shall implement the ecosystem-based approach to fisheries management so as to ensure that negative impacts of fishing activities on the marine ecosystem are minimised, and shall endeavour to ensure that aquaculture and fisheries activities avoid the degradation of the marine environment. ... "

Management plan with specific reference to the eel

Council Regulation (EC) No 1100/2007 of 18 September 2007¹³ establishes specific measures for the recovery of the stock of European eel. In particular, Article 2 of the Regulation states: "Establishment of Eel Management Plans.

- 1. Member States shall identify and define the individual river basins lying within their national territory that constitute natural habitats for the European eel (eel river basins) which may include maritime waters. If appropriate justification is provided, a Member State may designate the whole of its national territory or an existing regional administrative unit as one eel river basin.
- 2. In defining eel river basins, Member States shall have the maximum possible regard for the administrative arrangements referred to in Article 3 of Directive 2000/60/EC of the European Parliament and of the Council¹⁴.
- 3. For each eel river basin defined under paragraph 1, Member States shall prepare an Eel Management Plan.
- 4. The objective of each Eel Management Plan shall be to reduce anthropogenic mortalities so as to permit with high probability the escapement to the sea of at least 40 % of the silver eel biomass relative to the best estimate of escapement that would have existed if

Regulation (EU) No 1380/2013 of the European Parliament and of the Council of 11 December 2013 on the Common Fisheries Policy, amending Council Regulations (EC) No 1954/2003 and (EC) No 1224/2009 and repealing Council Regulations (EC) No 2371/2002 and (EC) No 639/2004 and Council Decision 2004/585/EC (O.J. L 354, 28.12.2013, p. 22–61)

Council Regulation (EC) No 1100/2007 of 18 September 2007 establishing measures for the recovery of the stock of European eel (O.J. L 248, 22.9.2007, p. 17–23)

Directive 2000/60/EC of the European Parliament and of the Council of 23 October 2000 establishing a framework for Community action in the field of water policy (O.J. L 327, 22.12.2000, p. 1–73)

- no anthropogenic influences had impacted the stock. The Eel Management Plan shall be prepared with the purpose of achieving this objective in the long term.
- 5. The target level of escapement shall be determined, taking into account the data available for each eel river basin, in one or more of the following three ways:
 - a) use of data collected in the most appropriate period prior to 1980, provided these are available in sufficient quantity and quality;
 - b) habitat-based assessment of potential eel production, in the absence of anthropogenic mortality factors;
 - c) with reference to the ecology and hydrography of similar river systems.
- 6. Each Eel Management Plan shall contain a description and an analysis of the present situation of the eel population in the eel river basin and relate it to the target level of escapement laid down in paragraph 4.
- 7. Each Eel Management Plan shall include measures to attain, monitor and verify the objective set out in paragraph 4. The Member States may define the means depending on local and regional conditions.
- 8. An Eel Management Plan may contain, but is not limited to, the following measures:
 - reducing commercial fishing activity,
 - restricting recreational fishing,
 - restocking measures,
 - structural measures to make rivers passable and improve river habitats, together with other environmental measures,
 - transportation of silver eel from inland waters to waters from which they can escape freely to the Sargasso Sea,
 - combating predators,
 - temporary switching-off of hydro-electric power turbines,
 - measures related to aquaculture.
- 9. Each Eel Management Plan shall contain a time schedule for the attainment of the target level of escapement laid down in paragraph 4, following a gradual approach and depending on an expected recruitment level; it shall include measures that will be applied as of the first year of application of the Eel Management Plan.
- 10. In the Eel Management Plan, each Member State shall implement appropriate measures as soon as possible to reduce the eel mortality caused by factors outside the fishery, including hydroelectric turbines, pumps or predators, unless this is not necessary to attain the objective of the plan.
- 11. Each Eel Management Plan shall include a description of the control and enforcement measures which will apply in waters other than Community waters in accordance with Article 10.
- 12. An Eel Management Plan shall constitute a management plan adopted at national level within the framework of a Community conservation measure as referred to in Article 24(1)(v) of Council Regulation (EC) No 1198/2006 of 27 July 2006¹⁵ on the European Fisheries Fund (1)".

Commission Regulation (EC) No 1198/2008 of 1 December 2008 establishing a prohibition of fishing for Greenland halibut in NAFO 3LMNO by vessels flying the flag of Estonia (O.J. L 323, 3.12.2008, p. 24–25)

In conclusion:

- 1) In light of the above information provided on the role assigned to the "Management Plans" by the European Regulations;
- 2) bearing in mind that the above mentioned regulations provide for control procedures, alongside Member States, for the compliance with the measures adopted in the management plans;
- 3) considering that the recalled regulations seem properly detailed and effective to achieve the intended purposes;
- 4) in view of the restriction established by the Group, for which: "A management plan, approved by the local or national authority, that ensures the sustainable exploitation of the species concerned, should be provided" in order to consider that "... restocking of wild fry in the extensive aquaculture farming carried out inside wetlands, such as brackish water ponds, tidal areas and costal lagoons is in line with the objectives, criteria and principles of organic aquaculture production";
- 5) with reference to the general principle that protection should be ensured to all overfished/endangered species, not only to the eel.

All the above considered, it seems that there are no reasons to believe that a "Management Plan" is effective in protecting any overexploited or endangered species, with the only exception of the eel.

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7. GLOSSARY

Weaning

Weaning is the time when the juveniles shift from natural feed to pellet feed.

Extensive aquaculture

Extensive aquaculture is a practice where the feeding is only provided by the natural carrying capacity of the natural environment.

Anti-nutrient

Anti-nutrients are substances present in vegetable protein sources, which impair protein digestion and utilization in the fish. Indeed, the digestive system of carnivorous fish has not evolved to deal with the wide variety of anti-nutritional factors (ANFs) that are present in most plant-based feedstuffs, and which may interfere with fish performance and health due to impaired nutrient utilization.

Phaffia yeast

Phaffia yeast consists of the cells of the yeast *P. rhodozyma* that are produced by pure culture fermentation and subsequently killed by heat and dried. The major components of phaffia yeast are proteins, carbohydrates, and lipids produced by the yeast cells. The primary colouring substance in phaffia yeast is astaxanthin. Astaxanthin is the carotenoid responsible for the orange-red colour of marine seafood, such as trouts, salmons and crustaceans. The high conjugated carbon-carbon double bonds give to astaxanthin both the properties of a potent antioxidant and a colorant.

PAP

Processed Animal Protein (PAP) means animal proteins (entire bodies or parts of animals or products of animal origin not intended for human consumption) derived entirely from category 3 material, which have been treated in accordance with Chapter II of annex V of the Commission Regulation (EC) No 1774/2002¹⁶. The Commission Regulation (EU) No 56/2013 allows the use of processed animal protein, derived from non-ruminants, for feeding aquaculture animals.

Retention efficiency

Improvements in protein retention efficiency is decisive to maintain animal health and welfare. A "metabolically" optimized protein and lipid diet formulation, for feeding farmed aquatic animals, allows a higher retention of nutrients, a higher feed conversion ratio and a reduced environmental impacts due to the lower amount of nitrogen discharge.

Euryhaline species

Euryhaline fish species are able to adapt to a wide range of salinities. An example of a euryhaline fish is the sea bream (*Sparus aurata*) which can live in brackish or salt water. Euryhaline organisms are commonly found in habitats such as estuaries and tide pools or lagoons, where the salinity changes regularly. However, some organisms are euryhaline because their life cycle involves migration between freshwater and marine environments because of reproductive needs. The salmon (anadromous fish) is one which migrate from the sea into fresh water to spawn and the eel (catadromous fish) is one which migrate from fresh water into the sea to spawn.

Commission Regulation (EC) No 1774/2004 of 14 October 2004 amending Regulation (EC) No 1623/2000 laying down detailed rules for implementing Regulation (EC) No 1493/1999 on the common organisation of the market in wine with regard to market mechanisms (O.J. L 316, 15.10.2004, p. 61–63)