Directorate-General for Agriculture
and Rural Development

Expert Group for Technical Advice on Organic Production

EGTOP

Final Report

On

Aquaculture (Part B)

The EGTOP adopted this technical advice by written procedure
in July 2014

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 2009/427/EC 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.

EGTOP Permanent Group

Keith Ball, Alexander Beck, Michel Bouilhol, Jacques Cabaret, Roberto Garcia Ruiz, Niels Halberg, Sonya Ivanova-Peneva, Nicolas Lampkin, Giuseppe Lembo, Lizzie Melby Jespersen, Robin Frederik Alexander Moritz, Bernhard Speiser, Fabio Tittarelli

Contact

European Commission
Agriculture and Rural Development
Directorate B. Multilateral relations quality policy
Unit B4 – Organics
L130 03/224A
B-1049 Brussels
Functional mailbox: agri-exp-gr-organic@ec.europa.eu

http://ec.europa.eu/agriculture/organic/eu-policy/expert-advice/documents/final-reports/index_en.htm

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Members of the Group are acknowledged for their valuable contribution to this technical advice. The members are:

Permanent Group members:
• Keith Ball
• Alexander Beck
• Michel Bouilhol
• Jacques Cabaret
• Roberto Garcia Ruiz
• Niels Halberg
• Sonya Ivanova-Peneva
• Nicolas Lampkin
• Giuseppe Lembo
• Lizzie Melby Jespersen
• Robin Frederik Alexander Moritz
• Bernhard Speiser
• Fabio Tittarelli

Sub-Group members:
• Giuseppe Lembo (chair)
• Bernhard Speiser (rapporteur)
• James Casey
• Alicia Estevez Garcia
• Alfred Jokumsen
• Nikos Papandroulakis
• Patrick Sorgeloos

External experts:
none

Observers:
none

Secretariat:
• João Onofre
• Luis Martín Plaza
• Dario Dubolino
• Eoin Mac Aoidh
• Louis Mahy
• Stefanie Noe

All declarations of interest of Permanent Group members are available at the following webpage:

http://ec.europa.eu/agriculture/organic/eu-policy/expert-advice/documents/declaration-of-interests/index_en.htm

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1. EXECUTIVE SUMMARY

The EGTOP (thereafter called ‘the Group’) has evaluated a number of topics relevant for organic aquaculture.

With respect to stocking densities, the Group concluded the following: (1) for Arctic charr, the Group recommends to increase the stocking density limit to 25 kg/m^3 per year. (2) For carp, the Group recommends to reduce the maximum limit of farming yield to 500 kg/ha per year. (3) For small-sized crayfish (<20 mm), the Group recommends a maximum stocking density of 100 individuals per m^2; for crayfish of intermediate size (20 - 50 mm) a stocking density of 20 – 30 individuals per m^2 and for adult crayfish (>50 mm) a maximum stocking density of 5 individuals per m^2. (4) Besides, the Group does not consider any further modifications of the figures in the Annex XIIIa of Regulation (EC) 889/2008 (as amended by Reg. (EC) 710/2009) to be appropriate.

With respect to intensive Recirculation Aquaculture Systems (RAS), the Group concluded that they should remain prohibited for on-growing purposes. However, re-use of water is clearly in line with organic principles of sustainable and responsible use of resources, and is to be encouraged and further explored.

With respect to eyestalk ablation, the Group concluded the following: (1) The techniques of pinching, enucleation/slittering, cautering and ligation all have to be considered as forms of eyestalk ablation, and are therefore currently prohibited. (2) In the Group’s opinion, all forms of eyestalk ablation should remain prohibited.

With respect to the use of hormones, the Group recommends not to allow the use of hormones for the production of caviar or juveniles in sturgeons.

With respect to the production of phytoplankton, the Group sees no possibility for applying the overall principle of fertilization with low solubility fertilizers (currently applied for terrestrial plants) to phytoplankton. Also, the Group considers that it would be difficult to define production of ‘organic phytoplankton’ which would be sufficiently different from conventional phytoplankton to justify its existence as a separate, organically certified product. In view of the necessity to use phytoplankton in hatchery, the Group recommends that for the time being, the use of phytoplankton should be authorized without requiring organic certification. GMO strains of algae must not be allowed.

With respect to the production of zooplankton, the Group recommends that in the absence of better alternatives, the use of non-organic zooplankton should be allowed.

With respect to the production of fish larvae, the Group recommends that, for larval rearing of marine species, methods such as the ‘mesocosm’ or ‘large volume rearing’ should be used. The specific requirements for such rearing systems include: (1) an initial stocking density below 20 eggs or larvae/litre, (2) a larval rearing tank volume of minimum 20 m^3, and (3) feeding of larvae on the natural plankton developing in the tank that is supplemented by externally produced phytoplankton and zooplankton.

With respect to disinfection and management of aquatic environment, the Group concluded the following:

- The use of tosylchloramide sodium (cloramine T) for disinfection is not in line with the objectives, criteria and principles of organic farming as laid down in Council Regulation (EC) No 834/2007. It should therefore not be included in Annex VII.
- The use of hydrogen peroxide/sodium percarbonate in the absence as well as in the presence of animals is in line with the objectives, criteria and principles of organic farming, and the Group recommends to include both substances in Annex VII, in the 'basic list of substances for management of aquatic environments'.
- The use of peracetic and peroctanoic acid in the absence as well as in the presence of animals is in line with the objectives, criteria and principles of organic farming, and the Group recommends to include both substances in Annex VII, in the 'basic list of substances for management of aquatic environments'.
- The use of hypochlorous acid produced from mixtures of potassium peroxomonosulphate and sodium chloride in the absence of animals is in line with the objectives, criteria and principles of organic farming. It should be included in Annex VII, along with sodium hypochlorite and calcium hypochlorite.
- The use of saltwater and freshwater is in line with the objectives, criteria and principles of organic farming. The Group recommends: (1) to include sodium chloride in Annex VII, in the 'basic list of substances for management of aquatic environments', and (2) to amend Art. 25s(6) as follows: "For biological control of ectoparasites preference shall be given to the use of cleaner fish, and to the use of freshwater, marine water and sodium chloride solutions".
- The use of slaked lime in the absence of animals, e.g. pre-treatment of water before it enters the rearing ponds/tanks, is in line with the objectives, criteria and principles of organic farming. In Annex VII, the entry 'lime' should be replaced by 'quicklime (calcium oxide) and slaked lime (calcium hydroxide)'.

The Group further recommends re-structuring section 2 of Annex VII of Reg. (EC) 889/2008 (as shown in chapter 4.6.2). Section 2 should be renamed to 'Substances for use in aquaculture and seaweed production'. It should be subdivided into three sub-sections. Section 2.1 should be named 'basic list of substances for management of aquatic environments'. Substances in this section may be used for all purposes authorized under general legislation. Section 2.2 should be named 'Substances for cleaning and disinfection of equipment and facilities, in the absence of aquaculture animals'. Substances in this section may be used for all purposes in the absence of aquaculture animals authorized under general legislation. Section 2.3 should be named 'Substances for limited use in aquatic environments'. Substances in this section may be used for very limited purposes indicated there.

The Group was asked to reconfirm the advice on various substances given in 2008 by an ad-hoc expert group. Due to time constraints, the Group could not make full evaluations, but it has indicated in which areas clarifications are most needed.

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 834/2007. The implementing rules were introduced via Commission Regulation 710/2009 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 R. 710/2008 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. **Stocking Density** for the main species or groups of species, other than molluscs, is set out in Annex XIIIa of R.889/2008. Article 25f(2) of the Regulation states that "in considering the effects of stocking density on the welfare of farmed fish, the condition of the fish (such as fin damage, other injuries, growth rate, behaviour expressed and overall health) and the water quality shall be monitored." France has requested that the maximum stocking density for Brown trout and Rainbow trout grown in fresh water be increased from 25 kg/m^3 to 35 kg/m^3 (supported by Bulgaria in comments on the draft mandate) and that the maximum farming yield of freshwater species in fishponds (carp, perch, pike etc.) be reduced from 1 500 kg of fish per hectare per year to 500 kg (Bulgaria supports annual production below 1500 kg/ha). Italy has requested that the maximum allowed density for trout is reduced from 25 to 20 kg/m^3, and that the maximum density for the charr be increased from 20 to 25 kg/m^3. Sweden requests that the EGTOP mandate include advice on the possibility to regulate the stocking densities for the crayfish species, *Astacus astacus* and *Pacifastaccus leniusculus*, both in ponds and for larvae and breeding ponds indoors. Sweden also proposes that EGTOP evaluate the pros and cons of closed recirculation systems in relation to Articles 3 to 5 of Regulation 834/2007. Advice on this area should include reference to density.

2. **Substances for cleaning and disinfection in the presence [and absence] of animals** require particular care and measures to ensure that the application is not harmful (according to Recital 17 of R. 710/2009). Currently only two substances are listed in Annex VII(2.2) and several requests have been received to add the following substances:

   - **Chloramine T/ Tosylchloramide sodium** - France has submitted a dossier and the application has been supported by Italy
   - **Hydrogen peroxide** (liquid or powder **(Sodium percarbonate**) - France and Ireland have submitted dossiers and their applications have been supported by Italy and Denmark. This substance is currently permitted for use in the absence of animals
   - **Sodium chloride –** France and Denmark have applied for its inclusion. This substance is currently permitted for use in the absence of animals
   - **Peracetic acid [and peroctanoic acid] –** France, Italy and Denmark have applied for the inclusion of Peracetic acid and France has submitted a dossier with its application for peroctanoic acid. Both substances are currently permitted for use in the absence of animals.

- **Hypochlorous acid –** The UK has applied for inclusion of this bleach formulation (active ingredient from potassium monopersulphate in combination) use in absence of animals. It is likely to be safer than sodium hypochlorite bleach (currently permitted for use in the **absence of animals**) as it does not produce toxic chlorine and is considered to have greater efficacy against pathogens].

Regarding lime (calcium oxide) which is currently permitted in the absence of animals, Denmark has pointed to the need for clarification that this also applies to **slaked lime (calcium hydroxide),** formed when lime is mixed (slaked) with water. Denmark has also indicated an interest in having slaked lime listed under 2.2 (use in presence of animals) and is willing to prepare a technical dossier.

3. **Reproduction.**
   a. Germany has pointed to the need for harmonization of the interpretation of the **prohibition of eyestalk ablation** for reproduction in shrimp. EGTOP should clarify the term ablation in relation to hatchery practices such as ligation, incision, pinching etc. which do not directly remove the eyestalk.

   b. Spain requested a clarification on the use of hormones (natural or artificial) for certain species as sturgeon, turbot and eels to reach the sexual maturity needed for reproduction.

4. **Specific rules for juveniles, invertebrates and microalgae/plankton**. In the context of the lack of organic juveniles mentioned under point 1) above, Spain has pointed to the need to develop **specific rules for the production of juvenile fish** to ensure continuity of production.

   Spain has requested that rules be developed for zooplankton, rotifers, micro-crustaceans, worms and other aquatic feed organisms. Spain has also pointed to the need for rules concerning multicellular marine algae/phytoplankton and microalgae for use as feed and food with particular focus on the use of nutrients of plant or mineral origin listed in Annex 1.

5. **Reconfirmation of ad-hoc Expert advice of 2008.** For the issues not mentioned above it would be useful that the group re-examine the advice provided by the Ad-hoc group five years ago on the other topics with a view to reconfirming or updating it. It should be noted that in the exchanges regarding the EU proposal to include organic aquaculture in Codex Guideline 32-1999 on Organically produced foods, one country has questioned the suitability of potassium permanganate and iodophores for cleaning and disinfection in the absence of animals. This country has also queried the use of sodium chloride and humic acid for the same use (on account of not being familiar with their use for this purpose).

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

6. **Deadline**
   The deadline for adoption of the **Part B final report**: **30 June 2014**

4. CONSIDERATIONS AND CONCLUSIONS

4.1 Stocking density

Introduction, scope of this chapter

A considerable number of studies have investigated stocking densities in relation to fish welfare, i.e. performance (e.g. mortality, feed intake, feed conversion ratio, growth), condition (e.g. fin and gill condition), and stress levels (e.g. plasma cortisol, plasma glucose, hematocrit, energetic metabolism). It is generally concluded, that despite the lack of clear evidence, high stocking density may compromise fish welfare. However, a low density may also be detrimental to welfare, as it may result in extremely aggressive behaviour between conspecifics (Ellis et al., 2002).

Impact of stocking density and other factors on animal welfare and health

Trout: A range of studies have investigated the relationship between stocking density and rearing or environmental conditions on different aspects of growth performance and welfare in rainbow trout (*Oncorhynchus mykiss*). North et al. (2006) studied the impact of stocking densities of 10, 40 and 80 kg/m^3 on a variety of physiological and morphometric indicators. They demonstrated that being held at high density (80 kg/m^3) did not have consistent effects on growth rates or physiological indicators of welfare, but increased fin erosion. Furthermore, they found evidence for stronger dominance hierarchies at low density (10 kg/m^3). Consequently, it was concluded that both low and high stocking densities had the potential to compromise welfare. Two studies have investigated the combined effects of stocking density (~ 25 and ~ 100 kg/m^3, respectively) and sustained exercise (water current of 0.9 body length/sec.). The first study showed that high density, irrespective of water current, resulted in a lower growth performance. Furthermore, water current was shown to have a positive effect on energetic budgets, reducing metabolic rate irrespective of density, and was attributed to induce schooling behavior thereby reducing aggressive behavior and stress (Larsen et al., 2012). The second study showed that growth rates were reduced at high stocking density, irrespective of water current and this was attributed to high energy costs. The authors concluded that this was unlikely to be due to chronic stress, as cortisol values were low at all densities, but may have been due to an alteration in physiological state (McKenzie et al., 2012). Interestingly, what is considered low density and what is considered high density appears to be quite ambiguous, as these ‘definitions’ vary between studies. Furthermore, the results of these studies clearly illustrate the complex nature of the interaction between stocking density, fish welfare and several environmental factors, which may influence indicators of welfare, performance and stress resilience.

Sea bass: A number of studies have been carried out with sea bass reared at different stocking densities. In a study with sea bass of approximately 135 g, four densities (10, 40, 70, 100 kg/m^3) were tested over a 63-day period (Sammouth et al 2009). Fish performance, stress indicators and water quality were compared. Up to a density of 70 kg/m^3, no significant differences in daily feed intake were observed. Density above 70 kg/m^3 showed a negative impact on growth performance, and at 100 kg/m^3 specific growth rate was decreased by 14 %. Santos et al. (2010) showed that high densities may act as a chronic stressor to the fish, leading to a reduced feed intake and growth. Carbonara et al. (2014) studied the relationships between stocking density and fish welfare. Adult sea bass were reared at either low (10 kg/m^3) or high (50 kg/m^3) stocking density for 84 and 116 days. In the higher density, the activity level (energetic expenditure), measured by U_{crit} and electromiograms (EMG) was about twofold higher than that in the lower density. Furthermore, the higher density group exhibited a decrease in the reserve of metabolic energy. In conclusion, the authors highlighted that EMG can better represent the integrated response of the whole fish organism to stress conditions. In other words, the amount of energy reserves (anaerobic metabolism) that fish could use to cope with stress conditions (Lembo et al. 2007).

Arctic charr: A relevant number of studies have also been carried out for assessing the suitable stocking density for breeding the arctic charr (*Salvelinus alpinus*). The general conclusion is that arctic charr tolerate relatively high stocking densities and, providing that water quality is secured, there is no evidence of stress conditions which may compromise fish welfare (Wallace et al., 1988; Brown et al., 1992; Christiansen et al., 1992; Jörgensen et al., 1993; Metusalach et al., 1997; Brännäs and Linnér, 2000; Gunnarsson et al., 2011; Dalsgaard et al., 2012).

Carp: Carp is mainly farmed in central European countries, where the most suitable farming conditions for carp have been identified; i.e. high fecundity, good growth rate, tolerance to unstable environmental conditions, good ability to utilize available natural food, as well as low-protein feeds (Adamek et al., 2012). The main producers are the Czech Republic and Poland, each producing about 18000 t in 2010, followed by Germany and Hungary each producing about 10000 t in 2010 (Adamek et al., 2012). The main farmed species of carp is Common carp (*Cyprinus carpio*). It is mainly produced extensively and semi-intensively in ponds based on natural food – zooplankton and zoobenthos. About 25 % of the carp production is semi-intensive, using barley and wheat as supplementary feed (Adamek et al., 2012). The average farming yield under traditional farming conditions is about 500 kg/ha (Adamek et al., 2012; Adamek, pers. comm.). However, organic carp can also be produced intensively with artificial feed (i.e. cereals of organic origin), which increases the production capacity to more than 1000 kg/ha. There seems to be a public opinion among Czech consumers that extensively/semi-intensively farmed carps have a better flesh quality than intensively farmed carps (Adamek, pers. comm.).

Crayfish: According to the existing literature, the signal crayfish (*Pacifastacus leniusculus*) and the noble crayfish (*Astacus astacus*) are cultured in Europe, mainly in Finland, Sweden, Poland, Bulgaria, United Kingdom, Germany and Spain. Crayfish is cultured either for direct human consumption or for restocking purposes, combining intensive and semi-intensive/extensive techniques. In intensive culture, densities of up to 1000 juveniles per m^2 have been used in the case of *P. leniusculus*, with relatively good results (Savolainen et al., 2004; González et al., 2010, 2011a), although the authors recommend lower number of juveniles (100 – 200 per m^2). For *A. astacus*, densities up to 500 juveniles per m^2 have been cultured using recirculating aquaculture systems (Abeel et al., 2012). In semi-intensive/extensive aquaculture, densities of 10 – 150 juveniles of 20 – 50 mm size and 5 – 30 individuals for adolescent and adult crayfish per m^2 are commonly used (Ackefors, 2000). Due to the moulting process during growth of crayfish and the vulnerability of the animals to cannibalism, it is necessary to provide the animals with refuges/shelters i.e. PVC pipes (Ackefors, 2000; González et al., 2011b; Savolainen et al, 2003, 2004) and keep low densities (Wolf, 2004).

Reflections of the Group / Balancing of arguments in the light of organic farming principles

It is a challenge to identify appropriate density limits that promote optimal welfare in fish. This is in part due to a lack of understanding of how the different environmental factors interact with each other and with stocking density to affect welfare (Ashley, 2007). Another reason is that the effect of density measures on welfare may vary greatly between studies, due to the study-specific nature of experiments. For example, studies vary in experimental duration, water quality, density levels used, feeding methods, size of the fish, life history of the fish, level of domestication, type of rearing system used and environmental conditions. A density threshold for one set of conditions may, therefore, not be relevant for another (Ashley, 2007) and makes comparison of the results between studies difficult. High stocking density potentially increases the risk of

prevalence of diseases, but incidence of disease may as well be related to water quality, environmental and management conditions.
Therefore, the Group considers stocking density as an appropriate fish welfare indicator only when it is considered in a holistic approach and linked to environmental conditions, water quality, feeding quality, life history of the fish, level of domestication, type of rearing system used, etc.
However, the Group recognizes the need to establish a synthetic indicator of the fish welfare, which could be easily understood, communicated and monitored (according to the Reg. EC 889/2008, Art. 25f, paragraph 2) together with fish conditions such as fin damage, other injures, growth rate, behaviour expressed, overall health and water quality.
The figures in Annex XIIIa are threshold values which, on average, represent safer fish welfare conditions. These limits are based on practical experience. On the whole, these limits have been successfully applied over the last few years in EU organic aquaculture. Even if slightly higher stocking densities might be possible under specific, local conditions, this does not mean that this would be possible for the whole sector. In the Group's opinion, in the case of the arctic charr there is no scientific evidence for setting the limit of the stocking density at a lower level than for the rainbow trout.
Regarding carp, very little scientific evidence is available to the Group. However, the Group thinks that the maximum yields for organic production should not exceed the yields under extensive and semi-intensive conventional production. The Group therefore recommends to reduce the maximum yields for carp from 1500 kg/ha to 500 kg/ha per year.
In the case of crayfish, densities should be adapted to the developmental stage and the rearing system used. For small (<20 mm) juveniles, a density of 100 individuals per m^2 is recommended, for animals of 20 – 50 mm size, density should be kept at 20 – 30 individuals per m^2 whereas for adult animals (>50 mm), the maximum density should be 5 per m^2.

Conclusions

- For arctic charr, the Group recommends to increase the stocking density limit from 20 kg/m^3 to 25 kg/m^3 per year.
- For carp, the Group concluded that the maximum limit of farming yield should be reduced from 1500 kg/ha per year to values which are typical for extensive and semi-intensive production (i.e. 500 kg/ha per year).
- For small-sized crayfish (<20 mm), the Group recommends a maximum stocking density of 100 individuals per m^2. For crayfish of intermediate size (20 – 50 mm), the Group recommends a maximum stocking density of 20 – 30 individuals per m^2. For adult crayfish (>50 mm), the Group recommends a maximum stocking density of 5 individuals per m^2.
- For the time being, the Group does not consider any further modifications of the figures in the Annex XIIIa to be appropriate.

4.2 Recirculation Aquaculture Systems for on-growing

Introduction

According to Art. 2j of Reg. 889/2008 a 'closed recirculation aquaculture facility' is defined as 'a facility where aquaculture takes place within an enclosed environment on land or on a vessel involving the recirculation of water, and depending on permanent external energy input to stabilize the environment for the aquaculture animals'. In a *closed recirculation aquaculture system (RAS)* new water is mainly supplied for filling up and to replace water lost by evaporation. The degree of recirculation can be of about 95 % (Jokumsen & Svendsen, 2010).

Description of Recirculation Aquaculture System (RAS)

Intensive Recirculated Aquaculture Systems (RAS) are used in aquaculture production to minimize water consumption, as well as the environmental impact of the water discharge. RAS can use the same water many times and hence includes a wide range of waste water treatment devices (Martins et al., 2010; Dalsgaard et al., 2012). As a matter of fact, the use of RAS disconnects the production from the external environment. A sketch of a RAS is given below:

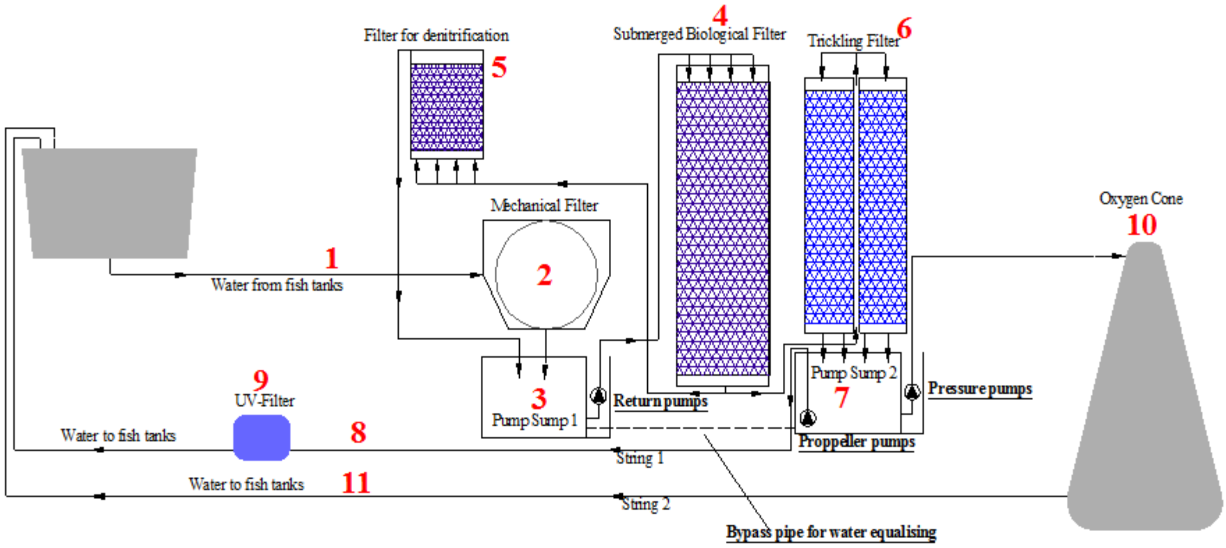

Figure 1: Sketch of a Recirculated Aquaculture System (RAS). The numbers in the figure are referred to (in brackets) in the text. Source: Billund Aquakulturservice ApS, Denmark.

The water supply for an intensive RAS freshwater farm is typically ground water, in the case of marine farms the water is pumped directly from the sea by means of submersible pumps. The production water from the fish tanks (1) passes through a mechanical filter (2), i.e. a microsieve (mesh size of about 60 µm). The microsieve separates particulate matter, which is flushed as sludge to a sludge storage tank until it can be used as agricultural fertilizer or for production of biogas. From the microsieve (2) the water is pumped (3) to the biofilters (4), where the dissolved fractions, especially ammonia (NH_4^+), are converted into nitrate (NO_3^-). In a separate biofilter (5) with anoxic (no oxygen) conditions (a denitrification filter), the NO_3^- is anaerobically converted into N_2 gas under consumption of easily degradable organic matter (Van Rijn et al. 2006; Suhr et al., 2013). The recirculated water passes on to a trickling filter (6) for degassing (N_2, CO_2) and aeration before it enters (7-8) the fish tanks. Before entering the fish tanks, the water passes an UV radiation device (9) to kill micro-organisms, especially bacteria. However, a portion of the aerated water from the trickling filter is pumped through an oxygen cone (7-10) for oxygenation before it enters (11) the fish tanks. In addition, pure oxygen may be added at each tank/section (Chen et al., 2006, Pedersen et al., 2012; Van Rijn, 2013) and the temperature can be adjusted using devices for heating or cooling the water. The amount of new water consumed in the RAS corresponds to the amount required to flush the microsieves (2) and the biofilters (4), to compensate for evaporation, and to keep the temperature at an appropriate level. The water consumption in RAS is more than 100 times less, i.e. less than 500 l/kg feed fed to the fish than in traditional flow through systems (Jokumsen & Svendsen, 2010). Obviously, RAS requires input of external energy for pumping water around, water treatment, and aeration of the water, as well as that required in the buildings. The advanced technologies, management, comprehensive surveillance systems, working processes, and hygienic procedures in a RAS farm requires well-educated and trained personnel with the competence required to achieve optimum productivity. The high degree of recirculation makes it critical to continuously monitor and control the water quality within narrow limits, and the extensive use of alarm systems is necessary for several parameters (Jokumsen & Svendsen, 2010).

Comparison of Recirculation Aquaculture Systems (RAS) and flow-through systems

In the following table, a comparison has been set up between a traditional flow through system in organic farming and an intensive recirculated aquaculture system (RAS).

Flow-through organic system	RAS
Advantages • Production in common with nature • Favours biological diversity and animal welfare • Natural temperature and light conditions • Lower stocking density • Behavioural needs can be met • Renewable energy use, e.g. for aerators • Environmentally sustainable **Disadvantages** • Dependent on external conditions (weather, temperature fluctuations, water quality) • Risk of escape • Risk of ingress of pathogens	**Advantages** • Low water consumption • Recycling of water • Stable farming conditions/water quality • Control of water temperature • No environmental impact • Prevents ingress of pathogens • Prevents escapes • Recycling/collection of waste nutrients (fertilizer) • Easy to disinfect/clean **Disadvantages** • Energy consuming • Use of pure oxygen • Higher stocking density • In case of disease, risk of boosting prevalence

Re-use of water

An alternative strategy is re-use of water which, to some extent, combines the advantages of both flow through systems and RAS, without compromising organic principles. Re-use of water means a kind of *extensive* recirculation in *out-door* systems with up to 70 % of reuse of the water (Colt, 2006). Instead of being discharged, the water is pumped back to the inlet and re-used in the fishponds, tanks or raceways after passing waste water treatment devices such as natural-filter beds, settlement ponds, mechanical or biological filters to collect waste nutrients, and/or using seaweeds and/or bivalves and algae, which contribute to improving the quality of the effluent. The type(s) and capacity of waste water treatment device(s) depend(s) on the specific conditions on the specific farm – related to production capacity/intensity approved and fulfilment of water quality criteria.

To comply with the species-specific physiological requirements of the fish, the proper oxygen saturation in the aquatic environment shall be achieved only by using mechanical aerators. This means that there should be a well- balanced equilibrium between the stocking density, the efficiency of the waste water nutrients removal and the amount of water re-used for the proper operation of the organic farm.

Reflections of the Group / Balancing of arguments in the light of organic farming principles

Most of traditional organic farms are open-air flow through systems. However, due to the limitations of water resources, national regulations in some countries require that farms are only allowed to take a limited amount of new water from the water courses. In such cases the re-use of water could be a solution in line with the principles of organic production.

Closed recirculated systems (RAS) have several environmental advantages, but require significant input of external energy, high stocking densities (for economic reasons), advanced waste water treatment devices, use of UV radiation and use of pure oxygen. All the above, together with the disconnection of the aquaculture production from the external natural aquatic environment, makes the closed recirculated systems (RAS) not in line with the principles of organic production.

Conclusions

The Group concluded that RAS should remain prohibited for on-growing purposes (see glossary). However, re-use of water is in line with organic principles of sustainable and responsible use of resources, and is to be encouraged and further explored.

4.3 Reproduction

4.3.1 Eyestalk ablation in shrimps

Introduction, scope of this chapter

Eyestalk ablation is currently prohibited in organic production (Reg.889/2008, Annex XIIIa, section 7). The Group was asked for clarification regarding the interpretation of the prohibition of eyestalk ablation for reproduction in shrimp. The Group was also asked to clarify the term ablation in relation to hatchery practices such as ligation, incision, pinching etc.

Necessity for eyestalk ablation, known alternatives

The crustacean eyestalk is the location for the X-organ sinus gland that contains a heat-stable factor which inhibits gonadal maturation (Quackenbush & Herrkind, 1981), a gonad inhibitory hormone (GIH) that occurs in nature in the non-breeding season and is absent or present only in low concentrations during the breeding season (Bray and Lawrence, 1992). The reluctance of most shrimp to routinely develop mature ovaries in captivity is a function of elevated levels of GIH, and eyestalk ablation lowers the high haemolymph titer of GIH. The effect of eyestalk ablation is not on a single hormone such as GIH, but rather affects several physiological processes. Besides the GIH evidence, another hypothesis suggests that eyestalk ablation also reduces light sensitivity and thereby induces ovarian maturation. In the banana prawn (*P. merguiensis*), dim light favours ovarian maturation and spawning. There are several direct and indirect effects of eye ablation in female shrimps, including;

- increases total egg production by producing more frequent spawning, but not larger spawns
- moult cycle duration is shorter
- increases mortality
- deteriorates female condition
- in some instances, produces lower hatch rate of eggs
- leads to changes in ovarian colour
- increases energetic demands
- leads to eventual loss in egg quality

Without ablation, shrimp hatcheries would have to rely on natural breeding. This is slow and unpredictable, especially for species like *Penaeus monodon*, therefore it would lead to shortages of the small shrimp needed to stock ponds. The aim of ablation, under these circumstances, is to stimulate the female shrimp to develop mature ovaries and spawn. Even in conditions where a given species will develop ovaries and spawn in captivity, use of eyestalk ablation may increase total egg production and increases the percentage of females in a given population that will participate in reproduction. Once females have been subjected to eyestalk ablation, complete ovarian development often ensues within as little as 3 to 10 days.

Many researchers are looking into ways to reduce or inhibit the hormones preventing breeding by using either molecular substances such as GIH-dsRNA and anti-GIH antibody that can be injected into female broodstock of shrimp to deplete GIH and neutralize its activity (Treerattrakool et al., 2008, 2014) or Serotonin (5HT) injections (Wongprasert et al., 2006). Also, some breeding programmes (Benzie, 2009; Preston and Clifford, 2002) are trying to develop shrimp breeds that are able to breed more efficiently without needing ablation.

Techniques of eyestalk ablation

There are four main techniques used for eyestalk ablation: pinching, enucleation/slitting, cauterisation and ligation.

Pinching is the most common technique used for ablation. One eyestalk is pinched between the thumb and index finger and squeezed. This destroys one of the glands producing the hormone that prevents breeding. This type of ablation is practical, as it is quick, cheap and can be done by one person. If a razor blade is used in conjunction with this technique, it speeds up the process. This method may leave an open wound.

Enucleation is the method of slitting one eye with a razor blade, then crushing the eyestalk, with thumb and index fingernail, beginning one-half to two-thirds down the eyestalk and moving distally until the contents of eyes have been removed. This leaves behind the transparent exoskeleton, so that clotting of haemolymph and closure of the wound, may occur more rapidly.

Cauterisation uses either an electrocautery device or an instrument such as a red-hot wire or forceps that are applied to the base of the eyestalk. This is a relatively low-stress method as the wound is sealed quickly and shrimp usually resume eating soon after ablation. If performed correctly, this method closes the wound and allows scar tissue to form more readily. A variation of this technique is to use scissors or a sharp blade to sever the eyestalk, and then to cauterise the wound.

Ligation means tying off the eyestalk tightly with surgical or other thread. This method also has the advantage of immediate wound closure. The thread is then tightened to limit the blood supply to the eyestalk. After ligation, the eyestalk falls off after a couple of days. The recovery rate for ligation is good and the shrimp are active soon after the tie is attached, and spawning and maturation are observed as normal.

Animal welfare issues

Shrimp have a very simple nervous system and there is no scientific evidence that they have feelings corresponding to the feeling of ‘pain’ in humans. However, in aquaculture facilities it is important to consider the animal’s state of health and the amount of stress it faces. Thus, a more comprehensive welfare definition should comprise a) the animal’s physiological and psychological capability to cope with its environment, b) the integrated response of the whole organism to stress conditions.

Ablation appears to be a relatively minor discomfort, as ablated shrimp might begin to behave and feed normally quite soon after the operation is completed. The speed with which the shrimp begins to feed is used as an indicator of the stress levels (if the ablation is done well, feeding will begin very soon after the procedure). Anaesthesia has been tried, but does not seem to improve recovery. It has been reported that in the tiger prawn (*Penaeus monodon*), the eyestalks fully regenerate in less than 6 months.

Nevertheless, physiological responses to stress should be regarded, first and foremost, as a condition of adaptive defense of the organism, that has the fundamental function of preserving the individual life.

Reflections of the Group / Balancing of arguments in the light of organic farming principles

Without eyestalk ablation, production of juveniles is unpredictable and does not allow a guaranteed production cycle. The alternative of collecting breeders in the wild, in absence of a well-documented management plan, is not desirable.

Although a lot of research activities have been made so far, most of the breeding programmes still have to rely on eyestalk ablation. To date, domesticated strains have played a dominant role in seed production for only *P. vannamei* and *P. stylirostris* (Benzie 2009), while for *P. monodon* real alternatives still need to be developed (e.g. varieties which do not need eyestalk ablation).

The Group accepts that ablation appears to be a relatively minor discomfort, as ablated shrimp might begin to behave and feed normally quite soon after the operation is completed. However, organic principles, and consumer expectations, are that organic animal husbandry avoids

mutilations in all animals. The Group thinks that for the sake of integrity of organic production, this fundamental principle should be uniformly applied for all animals.
Furthermore, the Group recognizes that there are different techniques for eyestalk ablation. If eyestalk ablation is to be authorized, the Group thinks that the technique of ligation would be more acceptable than pinching, enucleation/slittering, cauterisation or other methods.

Conclusions
The Group considers the techniques of pinching, enucleation/slittering, cauterisation and ligation as forms of eyestalk ablation, which are not in line with the principles of organic production.
In the Group's opinion, all forms of eyestalk ablation in shrimps should remain prohibited.

4.3.2 Use of natural/artificial hormones

Introduction, scope of this chapter
The Group was asked to clarify whether natural or artificial hormones can be used in fish such as sturgeon, turbot and eels. In this chapter, the question is mainly discussed for sturgeon. In the case of turbot, current aquaculture practices do not use hormones for reproduction. In eel, there is not yet any commercial breeding and/or production of elvers.
Many sturgeon species are threatened with extinction, being listed in CITES since 1998. The first trials in sturgeon farming started in the middle of the 19^{th} century, mostly to help in the conservation of wild populations through restocking. However, great advances in its culture have only been achieved in the last decades. Nowadays some sturgeon species are successfully being reproduced and raised in captivity (Coppens International, 2007). They are considered as slow growers in the wild, although under culture conditions some species have proven to have very high growth rates and to be tolerant to extremely high stocking densities (60-70 kg per m^3 with a survival rate of 50-80% from fry stage to marketable size) (Mims et al, 2002). There are several challenges for their production such as:

- supplies of brood stock and fry are very limited;
- there is a very long maturation period before females produce ripe eggs for reproduction;
- they need moderate temperature and ample supply of water;
- high initial investments;
- feed quality and proper management are requisites for proper reproduction.

Caviar (unfertilized eggs) and meat are the main products in sturgeon aquaculture. Nowadays, most caviar comes from aquaculture.

Necessity of using hormones, known alternatives
Sturgeon are slow to reach sexual maturity; some females cannot produce eggs until they reach an age of 30 years or more in the wild. Sometimes brood stock in the fish farms comes directly from the wild, although nowadays capture of wild fish is regulated by law and an increasing amount of brood stocks are reared in captivity. Under optimal culture conditions, and fed with high nutritional quality feeds, sturgeons reared in captivity can reach sexual maturatity in 1/3 – 1/2 of the time span needed in the wild. Gonad maturation requires 1 – 2 months at a water temperature below 10°C and final ripening is mediated by an increase in water temperature above 14°C using a slight increase in day length. Once they reach maturatity males and females are selected for spawning by determining the stage of gonadal maturity. In the case of males sperm is obtained by stripping, and in the case of females the maturatity of the eggs can be checked either by stripping or cannulation from the urogenital opening.
Certain fish, such as sturgeon or grouper, do not spawn in captivity and need supplementary hormones (via injection, slow-release pellets or water supplies) for final spermiation and final

egg maturation and ovulation (Mims et al, 2002; Coppens International, 2007). Hormonation can be done using, sturgeon (SP) and common carp (CCP) pituitaries, LH-RHa or a combination of both (Mohler and Fletcher, 1999). Optimum dosing is critical and depends on the species and body weight. 80 – 90% of females respond to the hormone injection with ovulation. Females are then checked for behavioral changes (they swim at the wall of the tanks, rubbing the walls) and when free adhesive eggs are observed in the tank, the eggs are removed from the female. There are several techniques to remove the eggs without sacrificing the female, such as caesarean section and minimal invasive surgical technique (MIST). MIST method is more rapid and consists in performing a small section in the posterior-ventral area of the oviduct that permits ovulated eggs to pass from the body cavity through the gonopore without going through the oviducts. With caesaran or MIST, only 50-90% of the eggs can be removed (Mims et al., 2002)
According to the information available to the Group, almost all conventional caviar producers currently use hormones. However, a few producers claim to produce caviar without hormones. Without hormones, the production of caviar is possible. However, the use of hormones does increase capacity and profitability.

Origin of raw materials, methods of manufacture
Carp and sturgeon pituitary extracts are from natural origin.

Animal welfare issues
Fish are anesthetized using MS-222, before they are treated with hormones.

Reflections of the Group / Balancing of arguments in the light of organic farming principles
The request is in conflict with Art. 25i of Reg. 889/2008, which states "*The use of hormones and hormone derivate is prohibited*". The Group is concerned about other possible uses of hormones in other species, and does not want to set a precedent by recommending their use in sturgeons. Consumers are worried about the use of hormones in general, regardless of amounts or purpose.

Conclusions
The Group recommends not to allow the use of hormones for the production of caviar in sturgeons, nor for production of juveniles.

4.4 Specific rules for production of juveniles and their feed

Introduction, scope of this chapter

Larval rearing is one of the most critical stages for the successful propagation of any species and represents one of the major bottlenecks of the whole aquaculture process. Most fish larvae, particularly the marine ones, are very small (total length of approximately 3 – 4 mm) at first feeding and thus are sensitive to rearing environment and to feed quality. Furthermore, these small larvae require live plankton for their first feeding and thus hatcheries include facilities for plankton production (both phytoplankton and zooplankton), the actual larval rearing zone and also for weaning – nursery. The majority of the hatcheries have also brood stock facilities, although in some cases transported eggs are used to initiate a production cycle.

Note: in larviculture, the term 'food' is often used for live prey and 'feed' for formulated rations. In this report, however, the term 'feed' is used throughout.

4.4.1 Phytoplankton

Mass-production of phytoplankton

Phytoplankton is of major importance in the hatchery process, having a double role. It is used in the rotifer cultures either as feed or as enriching media and also as medium for improvement of the rearing environment of larvae. Its role for larval rearing includes antibacterial properties but also shading effect that improves larvae predation or as trigger for feeding behavior or physiological processes (Scott & Baynes 1979; Naas *et al*. 1992; Tamaru *et al*. 1993; Reitan *et al*. 1993; Cahu *et al*. 1998; Van der Meeren 1991).

In all cases, the cultures are started from selected strains followed by an upscale in production (increase in volume) and are based on three operations: (i) strain maintenance, (ii) pre-cultures and (iii) mass cultures. The mass culture is usually performed in plastic bags or more recently in photobioreactors at high cell density (Tredici & Materassi 1992; Pulz 2001).

Nutrients needed for mass-production of phytoplankton

Commercial nutrient solutions contain all necessary macro- and micronutrients, silicates and vitamins in easily soluble, mineral form (Vonshak, 1986; Smith et al., 1993; Lavens & Sorgeloos, 1996).

There is a potential conflict with the principles of organic production. In the organic production of terrestrial crops, it is an overall principle that plants must not be fertilized with easily soluble nutrients. Art. 4(b)(iii) of Reg. 834/2007 limits the use of fertilizers to '*low solubility* mineral fertilizers'. In the implementing rules, hydroponic production is prohibited (Art. 4 of Reg. 889/2008). It is clear that this principle was developed for terrestrial plants, and does not hold for aquatic production, i.e. phytoplankton, where the nutrients are only available in soluble form. In the case of vitamins and other substances, the same rules concerning GMO risk should apply as for feed of terrestrial animals.

Inputs and technologies needed

Carbon dioxide is regularly supplied for phytoplankton cultures (especially in reactors) as a nutrient source. In the context of greenhouse production, carbon dioxide has been previously discussed by the Group (see EGTOP report on greenhouse production).

Conclusions on phytoplankton

The Group sees no possibility for applying the overall principle of fertilization with low solubility fertilizers (as given in Art. 4(b)(iii) of Reg. 834/2007 and currently applied for

terrestrial plants) to phytoplankton. Also, the Group considers that it would be difficult to define production of 'organic phytoplankton' which would be sufficiently different from conventional phytoplankton to justify its existence as a separate, organically certified product.
In view of the necessity to use phytoplankton in hatchery, the Group recommends that, for the time being, the use of phytoplankton should be authorized without requiring organic certification. However, GMO strains of algae must not be allowed.

4.4.2 Zooplankton

Mass-production of zooplankton as live feed for larvae of marine fish
Two species of zooplankton are mass cultured due to their appropriate size and easiness of mass culture. These are (i) the rotifer *Brachionus sp.* and (ii) the nauplius of the branchiopod crustacean, *Artemia sp.* Rotifers are the initial prey for the majority of marine fish larvae and are later replaced by *Artemia sp.* during the larval rearing process. Appropriate methods have been developed also for the culture of some ciliate species and for some copepods (Lavens & Sorgeloos, 1996, Marcus 2005, A. Tandler pers. comm.).
Rotifers are an excellent first feed for fish larvae because of their small size and slow swimming speed, their habit of staying suspended in the water column and their ability to be cultured at high densities due to a high reproductive rate (Dhert et al., 2001). As with microalgae, there are many recognized techniques for culturing rotifers. Production may be extensive in large 50 to 150 m^3 tanks, or intensive in small tanks of 1.0 to 2.0 m^3. Culture methods are classified as either batch, semi-continuous, or continuous.

Nutrients needed for mass-production of rotifers
For the feeding of rotifers several products are used (sometimes in combination), such as baker's yeast, different algal species (locally produced or purchased as algal paste) and formulated feeds.

Artemia
Artemia sp. is collected as dehydrated embryos or cysts from salt lakes and salt works. It is used either as instar I nauplii (400-600 micro-meters) hatched from cysts or as instar II-III nauplii (800-1000 micro-meters), reared with specially enriched feed. Frequently, cysts are de-capsulated with hypochlorite prior to hatching, in order to allow both preliminary disinfection of prey and better hatching rates (Lavens & Sorgeloos, 1996). Recently, other methods are applied which do not require de-capsulation: *Artemia* cysts are coated with non-toxic ferro-magnetic material (SepArt). After hatching the cysts, drain or siphon the nauplii and unhatched cysts into a separator that contains a magnet. Thus, unhatched cysts are trapped by the magnet, while nauplii are ready to use. Hatching and culture is performed in columns with high aeration at temperatures of about 26 °C.

Omega-3 fatty acids enrichment
Rotifers and *Artemia* need to be enriched in highly unsaturated fatty acids (EPA and DHA) and vitamins (C and A) and this can be done with microalgae (local cultures, algal pastes or powders of *Thraustrochytrids* single cell products) as well as oil emulsions. Commercial products are made up with synthetic antioxidants and emulsifiers, and do not comply with organic standards.

Reflections of the Group / Balancing of arguments in the light of organic farming principles
Unlike phytoplankton, the Group sees the technical possibility of an organic production of zooplankton, which would differ from conventional zooplankton in several aspects. Rules for organic *production* would need to be based on: Use of organic yeast and other microorganisms

(e.g. *thraustrochytrids*), only natural antioxidants and emulsifiers. For organic *enrichment*, only antioxidants, emulsifiers and vitamins, as allowed for terrestrial animals, should be used.
There are no organic enrichment diets available at the moment, and the Group is not able to evaluate whether their production would be commercially viable. The economic feasibility should be explored and the sector encouraged to consider organic production of zooplankton. Meanwhile, the Group sees no other possibility than to allow the use of non-organic zooplankton until better alternatives have been developed.

Conclusions on zooplankton
In the absence of better alternatives, the use of non-organic zooplankton should be allowed.

4.4.3 Production of larvae, post-larvae and juveniles

Introduction, scope of this chapter
A variety of hatchery techniques are available (Divanach & Kentouri, 1999), all sharing a common characteristic i.e. the use of plankton (phyto- and zooplankton) during the period of larval first feeding. The main classifications are based on the rearing density (intensive, semi-intensive, extensive) and the use of phytoplankton in the water (clear, green, pseudo-green) (Papandroulakis *et al.* 2002).
Independently from the applied method, there are three distinct phases during larval rearing: (i) egg hatching and autotrophic phase when larvae consume their yolk sac reserves, (ii) heterotrophic phase when larvae are fed on zooplankton, and (iii) the weaning to artificial diets. During these phases larvae complete their transformation to juveniles. Juveniles usually remain in the hatchery, for pre-growing, until reaching 2 – 5 g in weight. In cases where on-growing is performed in open sea conditions, the pre-growing period is extended until individuals reach a weight of 10-30g. During this period several procedures are commonly applied including grading, vaccination and quality control. This general scheme applies for both marine and freshwater larvae. A more detailed description of the applied techniques is presented in the following paragraphs

Intensive rearing systems for marine larvae
In intensive hatcheries, larvae are reared at high densities under controlled conditions and success is highly depending on the level of knowledge of the larvae’s specific biological needs. Intensive rearing is characterized by high stocking densities, controlled conditions of water quality, light intensity, photo-phase and feeding. The most commonly applied method are (i) the ‘clear water’ technique (Coves & Gasset 1993; Papandroulakis 2000), with no use of phytoplankton in the rearing medium, (ii) the ‘green water’ technique that is based on the creation of optimum conditions for endogenous phytoplankton bloom of specific organisms in the larval tanks (Saroglia *et al.* 1989), and (iii) the so-called ‘pseudo-green water’ technology (Papandroulakis *et al.* 2002), which is based on the frequent addition of phytoplankton and zooplankton in the larval rearing tanks, where phytoplankton is not produced, nor bloom, but its concentration remains constant by daily addition. The pseudo-green method is applied during the most critical segment of the rearing process: at the beginning of larval rearing (until the 20th to 30th day post hatching), when the larvae are still extremely weak, sensitive to alterations in the rearing environment, easily stressed and difficult to feed. After this period, the ’clear water’ methodology is applied.

Extensive and semi-intensive rearing systems for marine larvae

In extensive hatcheries, larvae are reared at low densities in large tanks or ponds under more natural conditions, feeding on endogenous blooms of wild marine zooplankton, but there is no industrial application due to the low productivity. As an intermediate approach between the intensive and extensive method, semi-intensive techniques, like the so called 'mesocosm technology' (Divanach & Kentouri 1999), have been developed and are applied for the rearing of several species. The actual form of the mesocosm technology was defined after studying the originally applied models of extensive rearing (Grice & Reeve 1982; Bever *et al.* 1985; Divanach 1985; Kentouri 1985; Lalli 1990). The most important characteristic of the infrastructure required is the size of the larval tanks which should range between 20 to 60 m^3. The conditions of rearing are independent from any climatic and/or seasonal changes. There is a partial control of the light conditions (intensity and photo-phase) and a minimal control of the temperature. The initial egg density in the mesocosm ranges from 4 to 7 eggs/l, depending on species, and should never exceed 20 eggs/l. Tanks are filled with natural seawater filtered mechanically, and wild plankton is thus introduced in the system offering a capacity for endogenous production. Phytoplankton is added daily to maintain the green medium for a period of 2 – 4 weeks after hatching. Exogenously produced enriched rotifers, enriched instar II *Artemia* sp. and artificial diet is added when required. The technology has been successfully used for the mass production of several species (Papandroulakis et al. 2004; Kentouri & Divanach 1983; Ben Khemis 1997; Koumoundouros et al. 1999; Papandroulakis et al. 2003; Papandroulakis et al. 2005). The mesocosm methodology results in high survival rates and low percentage of individuals with developmental abnormalities while, in general, larval growth performance is better than in the classical intensive systems. Similar semi intensive methods, like the above described, are also applied in different parts of the world, under different names such as 'large volume rearing' (Prestinicola et al 2013; Dhert et al., 1998) where the size of tanks, the rearing density and the presence of wild plankton are critical factors of the process. Recent studies (Prestinicola et al 2013) concluded that large volume rearing leads to a significant improvement of the morphological quality (i.e., lowered incidence of severe skeletal anomalies and meristic count variability) of gilthead seabream juveniles reared under semi-intensive conditions. Furthermore, there is evidence that the rearing conditions during the early life stages do have an impact on the behavioral response of sea bass during on-growing, and the individuals reared with the mesocosm method are more sensitive to human presence, presenting behavior closer to wild individuals (Papandroulakis et al., 2012).

Larval rearing of fresh water species (percid)

The larval rearing of pike-perch is very similar to that of marine fish larvae due to the size of the individuals at first feeding. The temperature is maintained constant at about 18 – 19 °C throughout the larval rearing phase, and gradually increased up to the time of transfer of juveniles to the on-growing tanks. The optimal temperature during on-growing is around 23 – 25 °C. Initial stocking density usually ranges between 20 and 50 larvae/l, but fish density must be reduced after the weaning phase. Feeding is based on live preys, similar to marine larvae, i.e. rotifers and *Artemia* nauplii. First feeding is composed of enriched rotifers (either the brackish water species *Brachionus plicatilis* or the freshwater species *B. calyciflorus*) or of small size *Artemia* nauplii (350-380 μm) for a period of 3 days. Afterwards, larvae are fed enriched *Artemia* nauplii (420-450 μm) (Lund, pers. comm.). At 25 – 30 days after hatching (body weight of 50 – 60 mg), the pikeperch are gradually weaned to appropriate dry feed, by replacing progressively the live prey with a high quality compound feed (300 – 500 μm) within 4-5 days.

Larval rearing of carp

Common carp are mainly omnivorous, with animal prey representing more than 75 % of the diet. A few days after hatching, the fish larvae feed mainly on small zooplankton, such as rotifers (not enriched) and copepod nauplii. After a short period, however, they shift to larger organisms such as cladocerans and copepods (Dulic et al., 2011; Nunn et al., 2012) or, seldom, to non-enriched *Artemia* nauplii. This change occurs gradually, largely depending on the size of the fish mouth, that is also correlated with body size. The size at which individuals shift from planktivorous to benthivorous feeding habit, however, depends on many factors, such as the availability of planktonic and benthic food, as well as the ratio between both types of food. Crustaceans will form the main component of the feed until individuals reach 100 – 150 mm. The amount of zooplankton ingested increases with fish size. From the juvenile stage onward, carp is primarily a bottom feeder, and aquatic insects (mainly benthic larvae of chironomids) form the main component of the diet (Adamek, 2014, pers. comm).

Larval rearing of molluscs and crustaceans
Mollusc larvae and start-feeding shrimp larvae are filter feeders and consequently feed on phytoplankton, which should be produced following the above recommendations (see section 4.4.1). Later stages of shrimp (mysis and postlarval stage), and most other crustacean larvae are first fed on *Artemia* and later on microdiets, therefore recommendations of section 4.4.2 should be applied, especially considering enrichment on polyunsaturated fatty acids.
For mollusc rearing, low densities are commonly used. For crustaceans, stocking density has variable effects: shrimp postlarvae can be kept at higher densities, while this is not the case for prawns and even less for crab and lobster.

Reflections of the Group / Balancing of arguments in the light of organic farming principles
For marine fish, there is evidence that juveniles produced with 'mesocosm' or 'large volume rearing' systems are more similar in behaviour and morphology to their wild counterparts. With respect to freshwater fish, molluscs and crustaceans, there is not enough experience with comparable rearing systems for drawing such conclusions.

Conclusions
The Group recommends that for larval rearing of marine fish species, methods such as the 'mesocosm' or 'large volume rearing' should be used. The specific requirements for these rearing systems include:

- An initial stocking density below 20 eggs or larvae/l.
- Larval rearing tank volume of minimum 20 m^3.
- Feeding of larvae on the natural plankton developing in the tank that is supplemented by externally produced phytoplankton and zooplankton.

With respect to the larval rearing of freshwater fish, molluscs and crustaceans, the Group cannot make specific recommendations at the moment, because of lack of scientifically based investigations.

4.4.4 Microbial control in hatchery

Introduction, scope of this chapter
As in any aquaculture operation, microbial control in hatcheries is essential and standard disinfection methods are applied for the facility and the equipment used. The list of substances in Annex VII covers the general requirements. Specific aspects are discussed below.

Disinfection of eggs
In some cases, fish eggs are disinfected. Disinfection of eggs is mandatory in salmonid species. In marine species, it is not frequently practised. The OIE manual recommends the use of

iodophors for egg disinfection (OIE 2012). Hydrogen peroxide is also used. For organic production, the Group recommends that no other substances than iodophors and hydrogen peroxide should be used for eggs disinfection.

Disinfection of zooplankton

Hydrogen peroxide is often used for disinfection of the live prey before administering the prey to the larvae. The range of substances authorized in Annex VII is sufficient, and there is no need for amendments.

Control of bacteria in hatcheries

The aquatic environment is more supportive to pathogenic bacteria, independently of their host, than the terrestrial environment and, consequently, pathogens can reach high densities around the animals, which then ingest them either with the feed or when they are drinking. As a consequence, culturing several species of aquatic animals (especially larval stages) in many cases suffers from highly unpredictable survival rates because of bacterial diseases (Verschuere et al., 2000). Independently from the species reared, no antibiotics are used in larval rearing, as larvae are in general very vulnerable and cannot tolerate the treatment. Hence, techniques to control pathogenic bacteria are paramount to the further development of the aquaculture sector.
In the review of Defoirdt et al. (2007) a critical evaluation is presented of alternative measures that have recently been developed to control disease caused by *V. harveyi* and closely related bacteria. Techniques discussed include phage therapy, the use of SCFAs and polyhydroxyalkanoates, quorum-sensing disruption, probiotics and 'green water' (see section 4.4.3 for explanation). Some of the techniques have only been studied recently and have only been tested in the laboratory (e.g. disruption of cell-to-cell communication), whereas others have a longer history, including farm trials (e.g. the application of probiotics). Each of the techniques has its advantages but also its limitations. In fact, none of them will probably be successful in all cases. Therefore, it is of importance to develop further all of these alternatives to construct a toolbox containing different sustainable measures. A good management strategy might then use different techniques in rotation to prevent resistance development. Alternatively, it might be valuable to determine which techniques are, and which are not, compatible with each other, to apply them together to maximize the chance of protecting the animals successfully.

Conclusions

Provided that the recommendations in chapter 4.5 are followed, the Group considers that the range of substances in Annex VII covers the general requirements for microbial control in hatcheries. However, alternative methods should be considered as soon as they become available for practical use.

4.4.5 Weaning procedure

Feed mixes

Feed mixes for weaning are different from those for on-growing. The Group thinks that their production in organic quality is technically possible. At the moment, the Group is not aware of any organic weaning diets which would comply with organic production rules. However, some manufacturers would be interested to produce such feeds, if there is sufficient demand.
The Group would welcome that feed companies develop organic diets for the early stages, in order to cover the specific requirements of the juveniles. During the weaning and the pre-growing phase, when dry feeds are used, the rules for organic aquaculture should be applied, both in terms of management and type of feed used.

4.5 Evaluation of substances for cleaning and disinfection

4.5.1 General comments on cleaning and disinfection

Use of substances in the presence of animals – experiences in Denmark
Currently, section 2.2 of Annex VII only includes limestone and dolomite as allowed for use in presence of aquatic animals. However, the possibility of using only these two substances is an urgent challenge for sustainable performance of the organic farming. Negotiations with the Danish authorities resulted in an amending specific authorization (DK, 2010) which allows the use of the following substances in Danish organic aquaculture:

- rock salt/ sea salt
- hydrogen peroxide
- sodium percarbonate
- mixture of hydrogen peroxide and peracetic acid
- calcium hydroxide (slaked lime)

Rock and sea salt, hydrogen peroxide, sodium percarbonate and mixtures of hydrogen peroxide and peracetic acid can be used in the presence of animals; slaked lime can be used prior to inlet to the ponds/tanks. At the time of authorization of these substances in 2010, research at DTU Aqua already had indicated positive results by using the substances as sanitizers in trout farming to keep sufficiently hygienic conditions/suppressed disease incidence. Research already carried out in recent years, and still ongoing, on these few substances has been shown to be efficient against pathogens, environmentally friendly and with no health risks for animals or humans (Pedersen et al., 2006; 2012; 2013). The above mentioned substances have proven to be useful in organic farming, as replacement for formaldehyde, chloramin-T and blue vitriol (copper sulphate). Indeed, organic farming is dependent on sanitizers for proper management and securing fish welfare, as farming in open systems increases the risk of infection with parasites, bacteria, viruses and fungi. Such substances were introduced in Danish aquaculture practice to improve environmental/labour conditions and to reduce possible negative environmental impact.

Use of substances in organic production
In this chapter, the Group evaluates the inclusion of substances in section 2 of Annex VII of the Reg. 889/2008.
The Group underlines that even for those substances which are *not* listed in Annex VII, the use is still possible with a veterinary prescription (see Art. 25t(2) of Reg. 889/2008).

Biocides legislation
The use of disinfectants is subject to Reg. 528/2012. This regulation distinguishes between various ‘product-types’ of biocides (see Annex V). Disinfectants used in aquaculture fall into product-type 3 ‘veterinary hygiene’. This category includes products for use in all animals (also terrestrial), and there is no specific sub-category for aquaculture.
Most of the substances discussed in this report have been in use for many years. Such substances are subject to the EU’s biocide re-evaluation programme, which is still on-going. For the moment, only few substances have been re-evaluated as biocides. In particular, the public register of ‘draft assessment reports’ on the European Commission’s website ‘CIRCABC’ does not yet contain any entries for product-type 3. For substances which were classified as ‘existing active substances’ in Reg. 1451/2007, national authorizations remain valid until the re-evaluation at EU level is completed.

Veterinary medicinal products legislation

Veterinary medicinal products are in the scope of Dir. 2001/82/EC. According to a guidance document[1], products for the control of external parasites of fish, used by adding the products to the water where fish swim, would normally be considered as veterinary medicinal products. However, the document admits in the absence of claims and in specific cases, they could also be considered as biocides. Finally, the document states that this advice is not legally binding, as only the Court of Justice can give an authoritative interpretation of existing Community law.

Conclusions on regulatory aspects

(1) The requests on disinfectants should not be postponed. Several member states have stated that the organic aquaculture sector urgently needs adaptations in the list of authorized disinfectants. In order to meet this need, the Group has decided to consider also substances for which no re-evaluation as biocides is available yet. This advice should be reconfirmed when the biocides re-evaluation is completed.

(2) It is not always simple to determine whether a given substance falls mainly under biocides legislation, veterinary medicinal products legislation or possibly some other legislation at EU and/or member state level. For these cases, the Group suggests to use a wording which clarifies that these substances must be used in compliance with general legislation, without anticipating association with any specific legislation (see proposals in section 4.6.2).

4.5.2 Tosylchloramide sodium (Chloramine T)

Introduction, scope of this chapter

The Group was asked whether tosylchloramide sodium can be used in the presence of aquaculture animals (inclusion in section 2.2 of Annex VII).

Tosylchloramide sodium is also known by several other names, such as ‘chloramine T’, ‘N-chloro tosylamide’ or ‘N-chloro 4-methylbenzenesulfonamide, sodium salt’. It has the CAS number 127-65-1 (see glossary), and the molecular formula $C_7H_7ClNO_2S \cdot Na$ ($3H_2O$). Despite the similarly sounding name ‘chloramine T’, this substance should not be confused with ‘chloramines’, which are derivatives of ammonia by substitution of one, two or three hydrogen atoms with chlorine atoms.

Authorization in general aquaculture and in organic production

Tosylchloramide sodium was identified as an ‘existing active substance’ (Reg. 1451/2007). At the moment, no draft assessment report is available and it has not been approved for PT 3. Thus, national authorizations remain valid until the re-evaluation at EU level is completed. For some other uses of tosylchloramide sodium (PT 1, 6, 9, 10 and 11), approval was rejected. No MRL is set for tosylchloramide sodium in fin fish (Reg. 37/2010). In organic production, tosylchloramide sodium is currently not authorized.

Technological or physiological functionality for the intended use

Tosylchloramide sodium is a strong oxidizing agent. Its action is based on an irreversible destruction of microbial cell material. It is active against a wide range of micro-organisms,

[1] Doc-Biocides-2002/01 (Version 08.01.2008). Guidance document agreed between the Commission services and the competent authorities of the Member States for the Biocidal Products Directive 98/8/EC and for the Medicinal Products for Human Use Directive 2001/83/EC and the Veterinary Medicinal Products Directive 2001/82/EC. BORDERLINE BETWEEN DIRECTIVE 98/8/EC CONCERNING THE PLACING ON THE MARKET OF BIOCIDAL PRODUCTS, DIRECTIVE 2001/83/EC CONCERNING MEDICINAL PRODUCTS FOR HUMAN USE AND DIRECTIVE 2001/82/EC CONCERNING VETERINARY MEDICINAL PRODUCTS.

including tuberculosis, foot-and-mouth disease and avian influenza. It is used for disinfecting surfaces and tools in hospitals, laboratories, medical, dental and veterinary facilities.
In aquaculture, it can be used for disinfection of installations and tools, as well as for preventive or therapeutic treatments of bacterial gill disease (EMEA 2005). The dossier mentions disinfections of various equipments, egg disinfection at the hatchery, and control of White Spot Disease (*Ichthyophthirius multifiliis*) in on-growing fish.

Necessity for intended use, known alternatives
The dossier does not give sufficient evidence of the necessity for using tosylchloramide sodium. The dossier stresses the use against white spot disease. However, there are alternatives for treating this disease (e.g. salt water, hydrogen peroxide).

Origin of raw materials, methods of manufacture
Tosylchloramide sodium is a synthetic substance. It is manufactured from p-toluenesulfonamide and sodium hypochlorite.

Environmental issues, use of resources, recycling
No evaluation as a biocide is available yet. According to the dossier, tosylchloramide sodium fully degrades within a few hours to a few days, depending on the properties of the receiving water. The environmental fate of the resulting chlorine is not further explained. Tosylchloramide sodium does not accumulate in sediments or trophic chains.
According to Danish environmental legislation[2], the maximum limit for discharge of tosylchloramide sodium is 5.8 µg/l. This precludes the use of tosylchloramide in practical aquaculture.

Animal welfare issues
If used correctly, the Group has no concerns over its impact on animal welfare. On the contrary, the prevention or cure of gill disease or other diseases is beneficial for the welfare of fish.

Human health issues
No evaluation as a biocide is available yet. If used correctly, the Group has no concerns over human health effects.

Impact on food quality
No issue

Traditional use and precedents in organic production
Tosylchloramide sodium has no traditional use in EU organic production.

Authorised use in organic farming outside the EU / 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. The NOP rules for livestock production do not allow the use of tosylchloramide sodium.

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[2] BEK nr 1022 af 25/08/2010 (Danish national consolidation act no. 1022 of 25/08/2010 ‘Consolidation act on environmental quality requirements for wetlands and requests to discharge of pollutants to water courses, lakes and the sea’).

Reflections of the Group / Balancing of arguments in the light of organic farming principles
Tosylchloramide sodium is a synthetic substance. There are authorized alternatives in Annex VII, and veterinary treatments would be an additional option.

Conclusions
The Group concluded that the use of tosylchloramide sodium for disinfection is not in line with the objectives, criteria and principles of organic farming as laid down in Council Regulation (EC) No 834/2007. It should therefore not be included in Annex VII.

4.5.3 Hydrogen peroxide / sodium percarbonate

Introduction, scope of this chapter
Hydrogen peroxide is already authorized for use in the *absence* of aquaculture animals (section 2.1 of Annex VII). The Group was asked to evaluate whether it could also be used in the *presence* of aquaculture animals (section 2.2 of Annex VII). In addition, the dossier also requests the authorization of the 'powder form' (sodium percarbonate).
Hydrogen peroxide has the chemical formula H_2O_2. It is the simplest peroxide and a strong oxidizer. Sodium percarbonate is an adduct of sodium carbonate (Na_2CO_3) and hydrogen peroxide (H_2O_2), with the formula $2Na_2CO_3 \cdot 3H_2O_2$. Due to its content of hydrogen peroxide, it is also an oxidizing agent.
This chapter discusses mainly hydrogen peroxide. Sodium percarbonate is mentioned in some places.

Authorization in general aquaculture and in organic production
Hydrogen peroxide is authorized for disinfection under general legislation as 'existing substance'. It is also authorized as a veterinary treatment against sea lice in salmonids.
According to Danish national Consolidation Act no. 1671 (22. Dec. 2010), on Organic Food and Organic Aquaculture, Art. 7, § 14 the use of hydrogen peroxide is authorised for use in water in presence of aquaculture animals in Danish organic production (see chapter 4.5.1).

Technological or physiological functionality for the intended use
Hydrogen peroxide has a wide range of uses as a bleaching agent and disinfectant in industry, medicine, dentistry and agriculture. Hydrogen peroxide is very effective against all kinds of pathogenic microorganisms (bacteria, fungi, parasites). In aquaculture, it can be used as a broad-spectrum disinfectant in all live stages of different species of fish and shellfish, including eggs. Hydrogen peroxide has various applications: in the *absence* of animals, it is used as a general disinfectant, as a water sanitizer, i.e. to lower bacterial loads and as part of biofilter maintenance in recirculated systems, and to reduce geosmine during depuration. In the *presence* of animals, it is used at the hatcheries and for on-growing for water treatment against the motile forms of one of the most common parasites of fish: *Ichtyobodo necator* (provoking direct mortality or major branchial injuries leading to bacterial infections) and *Ichthyophthirius multifiliis* (belonging the protozoa ciliates and provoking the White Spot Disease), on eggs and broodstock against *Saprolegnia*, to treat Amoebic Gill Disease in Salmonids and to disinfect zooplankton before feeding to marine larvae (see review by Yanong, 2014). Hydrogen peroxide is easy to use as a dip, flush or bath treatment and concentration can easily be monitored by use of test strips.
Sodium percarbonate is used to disinfect ponds in the absence of animals, and also together with water filtration to treat parasites (Heinecke and Buchmann 2009). It is also used for terminal disinfection and cleaning of systems. It increases pH and liberates hydrogen peroxide (Heinecke & Buchmann, 2009; Møller et al., 2010; Pedersen et al., 2006; Pedersen & Pedersen, 2012; Saez

& Bowser, 2001; Schmidt et al., 2006). Sodium percarbonate may also be used as emergency oxygen (‘oxygen powder’), as it liberates O_2 during degradation.

Necessity for intended use, known alternatives
Hydrogen peroxide has been used in aquaculture as a substitute for other, less preferable, substances such as formaldehyde or tosamylchloramine sodium.

Origin of raw materials, methods of manufacture
Hydrogen peroxide is naturally produced in trace quantities by organisms (Schmidt et al., 2006), most notably by a respiratory burst as part of the immune response. The substance used for disinfection is manufactured synthetically, most frequently by the anthraquinone process.
Sodium percarbonate is produced industrially by reaction of sodium carbonate and hydrogen peroxide, followed by crystallization. The Group has no evidence for a natural occurrence of sodium percarbonate.

Environmental issues, use of resources, recycling
Hydrogen peroxide spontaneously dissociates in water producing dissolved oxygen and water. When discharged into the river system, it disappears in a few hours to a few days (half-life of about 5 days in continental aquatic environment). The more the water contains oxidizable matter the faster the dissolution (consumption by organic matter in the water column, sediment, macrophytes). It does not accumulate in the sediments or in the food chain.
Hydrogen peroxide has little or no chronic toxicity in the aquatic environment, due to its chemical properties. It is also produced naturally by aquatic ecosystems in low concentrations.
Regarding acute toxicity (exposure to hydrogen peroxide less than a few hours), the data is abundant for fish (cf. Schmidt et al., 2006). There is no risk of acute toxicity on the aquatic environment under normal conditions of handling.

Animal welfare issues
The Group has no concerns. On the contrary, it is desirable as water sanitizer.

Human health issues
As long as hydrogen peroxide and sodium percarbonate are used correctly, the Group has no concerns. Sodium percarbonate is easier for handling than hydrogen peroxide.

Impact on food quality
No concerns.

Traditional use and precedents in organic production
Hydrogen peroxide is already authorized for use in the *absence* of aquaculture animals. In at least one member state (Denmark) its use in the presence of animals is authorized (see chapter 4.5.1).

Authorised use in organic farming outside the EU / 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. The NOP rules for livestock production allow the use of hydrogen peroxide ‘as disinfectants, sanitizers and medical treatments as applicable’, i.e. in the absence and in the presence of animals. Sodium percarbonate is not

allowed for livestock production, but it may be used as a pesticide, ‘if the requirements of 205.206(e)[3] are met’.

Reflections of the Group / Balancing of arguments in the light of organic farming principles
Sodium percarbonate acts by releasing hydrogen peroxide as active substance. Nevertheless, hydrogen peroxide and sodium percarbonate are two separate, clearly defined substances. Sodium percarbonate would need to be listed explicitly in Annex VII, in order to be authorized.
Water treatment/sanitation and disinfection is part of good fish husbandry and health management in the aquaculture production. Hydrogen peroxide/sodium percarbonate is efficient against external fungi and parasites that cause heavy damages both at the hatchery and at the on-growing stages. Hydrogen peroxide/sodium percarbonate is easily degradable and considered as safe both regarding human health and environmental impact.

Conclusions
The Group concluded that the use of hydrogen peroxide and sodium percarbonate in the absence as well as in the presence of animals is in line with the objectives, criteria and principles of organic farming as laid down in Council Regulation (EC) No 834/2007.
The Group recommends to include both substances in Annex VII, in the ‘basic list of substances for management of aquatic environments’. The concept of the ‘basic list of substances for management of aquatic environments’ is explained in detail in chapter 4.6.

4.5.4 Peracetic acid and peroctanoic acid

Introduction, scope of this chapter
Peracetic acid and peroctanoic acid are already authorized for use in the *absence* of aquaculture animals (section 2.1 of Annex VII). The Group was asked to evaluate whether they could also be used in the *presence* of aquaculture animals (section 2.2 of Annex VII). The simultaneous listing of these two acids can apparently be traced back to the existence of a commercial product containing these two active substances. The Group chose to carry out a general evaluation of these substances, not restricted to a particular commercial product.
Peracetic acid (also known as peroxyacetic acid, or PAA; CAS number: 79-21-0), is an organic compound with the formula CH_3COOOH. This organic peroxide is a highly corrosive, colourless liquid. It is frequently used as a disinfectant, also without peroctanoic acid.
Peroctanoic acid (also known as peroxyoctanoic acid; CAS number: 33734-57-5) has the formula $CH_3(CH_2)_6COOOH$. It is similar to peracetic acid, but has a longer hydrocarbon chain. This substance is not commonly used for disinfection. The only use that the Group is aware of is in the commercial product mentioned above, where it is a minor ingredient. The Group had very limited information on peroctanoic acid at disposal.
Authorization in general aquaculture and in organic production
Both for peracetic and peroctanoic acid, applications as PT 3 biocides are pending.
According to Danish national Consolidation Act no. 1671 (22. Dec. 2010), on Organic Food and Organic Aquaculture, Art. 7, § 14 the use of peracetic acid is authorised for use in water in presence of aquaculture animals in Danish organic production (see chapter 4.5.1).

Technological or physiological functionality for the intended use
When peracetic acid dissolves in water, it dissociates to hydrogen peroxide (which is also authorized; see chapter 4.5.3) and acetic acid. Peracetic acid is a very powerful oxidant; the

[3] reference to US law (‚National Organic Program‘)

oxidation potential outranges that of chlorine and chlorine dioxide. Peracetic acid is used in aquaculture as a sanitizer, similar to applications with hydrogen peroxide (see chapter 4.5.3 above) (Kitis, 2004; Meinelt et al., 2009; Pedersen et al., 2009; Pedersen et. al., 2013).
Based on structural analogy, the Group assumes that a similar reaction occurs when peroctanoic acid dissolves in water, again releasing hydrogen peroxide.

Necessity for intended use, known alternatives
Peracetic acid and peroctanoic acid have been used in aquaculture as a substitute for other, less preferable, substances such as formaldehyde or tosylchloramide sodium.

Origin of raw materials, methods of manufacture
Peracetic acid is a synthetic substance. It is produced by autoxidation of acetaldehyde. The Group could not ascertain the origin of peroctanoic acid, but assumes that this is also a synthetic substance.

Environmental issues, use of resources, recycling
Peracetic acid is easily degradable as it reacts with organic matter (chemical oxidation) in the water, as well as on surfaces. Degradation products of peracetic acid are via acetic acid to acetate and CO_2 and H_2O as end products. Half-life at realistic dosage is typically below 1 hour in aquaculture systems. Moreover, peracetic acid trade products require only small application doses, thus limiting the environmental impact (Pedersen et al., 2009; Pedersen et al., 2013).
For peroctanoic acid, the Group assumes similar degradation, also finally leading to CO_2 and H_2O as end products.

Animal welfare issues
The Group has no concerns. On the contrary, the substances are desirable as water sanitizers.

Human health issues
As long as they are used correctly, the Group has no concerns.

Impact on food quality
No issue.

Traditional use and precedents in organic production
Peracetic acid and peroctanoic acid are already authorized for use in the *absence* of aquaculture animals. In Denmark, peracetic acid is also authorised for use in *presence* of aquaculture animals (see chapter 4.5.1).

Authorised use in organic farming outside the EU / 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. The NOP rules for livestock production allow the use of peracetic acid for cleaning and disinfection in the absence of animals, while peroctanoic acid is not allowed.

Reflections of the Group / Balancing of arguments in the light of organic farming principles

Water treatment/sanitation and disinfection is part of good fish husbandry and health management in the aquaculture production. For peroctanoic acid, very limited information was available, so that the Group had to derive some informations from analogy with peracetic acid. However, the Group thinks that this can be justified by the similarity of these two substances, Peracetic acid and peroctanoic acid are efficient against bacteria and parasites that cause heavy damages both at the hatchery and at the on-growing stages. Both are easily degradable and will fully decay before discharge. They are considered as safe both regarding human health and environmental impact.

Conclusions
The Group concluded that the use of peracetic acid and peroctanoic acid in the absence as well as in the presence of animals is in line with the objectives, criteria and principles of organic farming as laid down in Council Regulation (EC) No 834/2007.
The Group recommends inclusion of both substances in Annex VII, in the 'basic list of substances for management of aquatic environments' (see chapter 4.6).

4.5.5 Hypochlorous acid produced from mixtures of potassium peroxomonosulphate and sodium chloride

Introduction, scope of this chapter
The Group was requested to evaluate the acceptability of a solid commercial product which releases hypochlorous acid in water. The request was accompanied by a comprehensive documentation from the manufacturer, but not with a member state dossier. The Group chose to carry out a general evaluation of the active substances involved, not restricted to the particular commercial product.
The product contains a mixture of common sea salt/rock salt (NaCl) with a strong oxidizing agent in solid form. The oxidizing agent is a triple salt consisting of the active constituent potassium peroxomonosulfate ($KHSO_5$), along with potassium hydrogen sulfate ($KHSO_4$) and potassium sulfate (K_2SO_4). When this mixture is added to water, chlorine (Cl_2) is formed.
Chlorine by nature reacts with water, and forms hypochlorous acid (HOCl) or the hypochlorite ion (OCl^-) and the hydrogen ion (H^+). Which form predominates depends on the pH of the water: under acidic conditions, hypochlorous acid predominates, while hypochlorite predominates under alkaline conditions. To stabilize the system and to achieve predominance of hypochlorous acid, an organic acid is added to the commercial product.
It is noteworthy that the hypochlorite ion is also formed when sodium hypochlorite or calcium hypochlorite (both included in section 2.1 of Annex VII) are dissolved in water.

Authorization in general aquaculture and in organic production
The production of hypochlorous acid from mixtures of potassium peroxomonosulphate and sodium chloride is considered as '*in-situ* generated biocidal active substance' (EC 2014) and falls under the scope of the new biocides legislation (Reg. 528/2012). For such substances, the deadline for making a declaration of intention to notify a precursor has just expired (EC 2013), and no decisions have been published yet.

Technological or physiological functionality for the intended use
Hypochlorous acid is a broad spectrum disinfectant, which is effective against a wide range of bacteria, viruses, fungi and mycoplasmas, including a range of plant, animal and human pathogens. It may be used in greenhouses, poultry, swine, equine and bovine production and in

aquaculture. In aquaculture, it is intended for disinfection of equipment, and it is commonly used in footbaths.
According to the dossier of the manufacturer, the biocidal activity of hypochlorous acid is greater than that of hypochlorite.

Necessity for intended use, known alternatives
Hypochlorous acid might be an important sanitizer, along with similar substances listed in Annex VII.

Origin of raw materials, methods of manufacture
The oxidizing agent potassium peroxomonosulfate is synthetically manufactured.

Environmental issues, use of resources, recycling
According to the dossier of the manufacturer, the environmental profile of hypochlorous acid is considered to be identical to hypochlorite, which is already listed in Annex VII.

Animal welfare issues
No issues identified.

Human health issues
According to the dossier of the manufacturer, hypochlorous acid produced in this way is expected to be safer than sodium hypochlorite. In this sense, the Group has no concerns, if such products are used correctly.

Impact on food quality
If used in the absence of animals, the Group has no concerns.

Traditional use and precedents in organic production
Sodium hypochlorite and calcium hypochlorite have been authorized in organic production for a number of years, and are included in section 1, as well as section 2.1 of Annex VII. In aqueous solution, they form the hypochlorite ion, which is transformed into hypochlorous acid under acidic conditions. Thus, the same active ingredient has been authorized before.

Authorised use in organic farming outside the EU / 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. The NOP rules for livestock production do not allow the use of potassium peroxomonosulphate.

Reflections of the Group / Balancing of arguments in the light of organic farming principles
The substances used to generate hypochlorous acid are synthetic, but the same is true for sodium and calcium hypochlorite. Because the active ingredient hypochlorous acid is the same as for sodium hypochlorite and calcium hypochlorite, the use of such mixtures seems similarly acceptable to the Group. The safety of handling is an argument in favour of hypochlorous acid produced from mixtures of potassium peroxomonosulphate and sodium chloride. The availability of an additional disinfectant may create new options for disinfection in aquaculture. In the Group’s opinion, sufficient disinfection is clearly preferable to the use of veterinary drugs.

Conclusions
The Group concluded that the use of hypochlorous acid produced from mixtures of potassium peroxomonosulphate and sodium chloride in the absence of animals is in line with the objectives, criteria and principles of organic farming as laid down in Council Regulation (EC) No 834/2007. It should be included in Annex VII, along with sodium hypochlorite and calcium hypochlorite.

4.5.6 Sodium chloride (salt)

Introduction, scope of this chapter
Sodium chloride (NaCl) is commonly known as salt. It is already authorized for use in the *absence* of aquaculture animals (Annex VII). The Group was asked to evaluate whether it could also be used in the *presence* of aquaculture animals. The use of marine water can have the same effect as sodium chloride solutions on freshwater fish. On marine species, the use of freshwater can have the same effect.

Authorization in general aquaculture and in organic production
Some of the potential applications of sodium chloride in fish production clearly do not fall under the scope of biocide legislation.

Technological or physiological functionality for the intended use
Sodium chloride has many potential applications in fish production. It serves to reduce osmoregulatory stress during transport and handling, and to avoid methemoglobia (brown blood disease). In addition, it controls some parasites (protozoans on the gills and skin of fish) (Francis-Floyd 1993). Details of the use are given by Francis-Floyd (1993). Freshwater fish (together with their freshwater parasites) are exposed to solutions of sodium chloride or sea water. Dosage and treatment time must be adapted to the fish species and to the practical conditions. Similarly, marine fish (together with their marine parasites) can be exposed to freshwater. In both cases, the antiparasitic action is caused by the osmotic pressure.

Necessity for intended use, known alternatives
With respect to its use for reducing stress during transport and handling, there are no alternatives. With respect to its effect on parasites, alternatives exist, but sodium chloride is preferable, because it is natural.
Freshwater is currently one of the treatments of choice for Amoebic Gill Disease, which is caused by *Neoparamoeba perurans,* a parasome. It has no toxicity issues and dosage is easier to manage than with hydrogen peroxide.

Origin of raw materials, methods of manufacture
Sodium chloride is a natural substance of mineral origin. It is obtained either from sea water, or from mineral deposits.

Environmental issues, use of resources, recycling
The Group is not aware of sodium chloride accumulation in free flowing water systems as an environmental concern. Note that in terrestrial cropping systems, salinisation is a major concern and needs to be avoided (see EGTOP report on greenhouse production).

Animal welfare issues
Stress reduction during transport is beneficial. If properly used, the Group has no concerns.

Human health issues
The Group has no concerns.

Impact on food quality
The Group has no concerns.

Traditional use and precedents in organic production
The use of sodium chloride is already authorized in the *absence* of aquaculture animals (section 2.1 of Annex VII). In addition, sodium chloride is authorized as a fertilizer (Annex I), as feed material (Annex V) in food processing (Art. 27.1.e) and for seaweed dehydration (Art 29a.1).

Authorised use in organic farming outside the EU / 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. The NOP rules for livestock production allow the use as feed, for health care and on management tools and production aides. However, it may not contain synthetic anti-caking agents or other prohibited substances.

Reflections of the Group / Balancing of arguments in the light of organic farming principles
From the mode of action, the Group considers such treatments as management practices rather than as disinfection. Not only sodium chloride should be authorized, but also treatment with marine water and freshwater (as explained above).

Conclusions
The Group concluded that the use of marine water and freshwater is in line with the objectives, criteria and principles of organic farming as laid down in Council Regulation (EC) No 834/2007.
The Group recommends:

1. to include sodium chloride in the basic list of substance in Annex VII, and
2. to amend Art. 25s(6) as follows: "For biological control of ectoparasites preference shall be given to the use of cleaner fish<u>, and to the use of freshwater, marine water and sodium chloride solutions".</u>

4.5.7 Slaked lime

Introduction, scope of this chapter
Calcium *hydroxide* ($Ca(OH)_2$) is traditionally called ‘slaked lime’. It is obtained when calcium oxide is mixed with water. Calcium *oxide* (CaO) is also known as ‘quicklime’ or ‘burnt lime’. The terminology in Annex VII is inconsistent: In section 1, both lime and ‘quicklime’ are mentioned (but the same substance is meant). In section 2.1, it is incorrectly referred to as ‘lime’ (a term which connotes various calcium-containing inorganic materials, such as carbonates, oxides and hydroxides of calcium).
The Group was asked (1) to clarify whether slaked lime can be used in the absence of aquaculture animals (as a consequence of the listing of quicklime in section 2.1), and (2) whether slaked lime can be used in the presence of aquaculture animals (listing in section 2.2). No dossier was submitted to support these requests.

Authorization in general aquaculture and in organic production

Both for calcium hydroxide and calcium oxide, dossiers for biocide PT 3 are currently being examined. The Group could not verify whether all applications of quicklime and slaked lime will fall under biocides legislation.

Technological or physiological functionality for the intended use
Following harvest, ponds are normally cleaned (as far as possible) and dried out for one or two weeks, to kill unwanted organisms including parasites, diseases and their vectors. Particularly in ponds, the bottom is often also limed. The method is described by OIE (2012), Boyd (2012) and Tonguthai (2000). Quicklime attacks any organism by desiccation/dehydration. In addition, both quicklime and slaked lime will raise the pH to 11 – 12, which also kills unwanted organisms. Quicklime is more effective than slaked lime.
In aquaculture, both recreational and commercial ponds are often fertilized to improve fish production. In ponds built on acidic soils and filled with fresh water of low mineral content, much of the phosphorus added in fertilizers becomes tightly bound in pond sediment where it is not available to support phytoplankton growth. Proper liming can improve phosphorus availability and greatly enhance pond productivity.

Necessity for intended use, known alternatives
Slaked lime is used for disinfection of ponds, i.e. drying out to get rid of viruses or other serious pathogens. For practical reasons, slaked lime or quicklime are ideal for this purpose. As the Group has no concerns over the use of these substances (see below), they are preferable to other chemical disinfectants. Liming helps to reduce the period of fallowing.
Slaked lime has also a use for water conditioning. It is used to remove solid iron compounds (‘red ochre’; Fe_2O_3). In waters with a high content of ferric hydroxide, the use of slaked lime is essential, in order to avoid high mortality of the fish due to accumulation of ochre in the gills. It can also be used to remove aluminium compounds from the inlet water, where this is necessary. Such treatments take place prior to inlet to the ponds/tanks. Hence, in the Group’s opinion, such uses can be classified as a use in the *absence* of animals.

Origin of raw materials, methods of manufacture
Calcium oxide is usually made by thermal decomposition of calcium carbonate (materials such as limestone or seashells) (‘calcination’). It does not occur in nature.
Calcium hydroxide is obtained by mixing calcium oxide with water. In nature, calcium hydroxide occurs in the form of the mineral ‘portlandite’.

Environmental issues, use of resources, recycling
Upon contact with water, calcium oxide is transformed to calcium hydroxide. By reaction with carbon dioxide from the air, this is transformed to calcium carbonate, and pH falls to 8.5 or less within a few days (Boyd 2012). The Group has no environmental concerns.

Animal welfare issues
The Group has no concerns.

Human health issues
Quicklime is caustic, and can burn the skin and cause serious eye injury (Boyd 2012). It reacts violently with water, so that particles may shoot-out, which presents a special risk for the eyes. Slaked lime is also caustic, but less hazardous. When proper precautions are taken (wearing of personal protective equipment), both substances can be used safely.

Impact on food quality
No issues identified.

Traditional use and precedents in organic production
Quicklime is already authorized for use in the absence of animals. According to Danish national Consolidation Act no. 1671 (22. Dec. 2010), on Organic Food and Organic Aquaculture, Art. 7, § 14 the use of slaked lime is authorised in Danish organic production for use ‘in presence of aquaculture animals’ (i.e. prior to inlet, see chapter 4.5.1). Shellfish may be treated with a lime solution to control competing fouling organisms (Art. 25p(2) of Reg. 889/2008). The Group assumes that ‘lime solution’ refers to aqueous solutions of slaked lime.

Authorised use in organic farming outside the EU / 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. The NOP rules for livestock production allow the use of ‘hydrated lime’ (=slaked lime) for health care, against external parasites and on management tools and production aides. It may be used as a topical disinfectant and external pest control. It may only be used in organic livestock production, ‘if the requirements of 205.238 are met’.

Reflections of the Group / Balancing of arguments in the light of organic farming principles
Quicklime and slaked lime are two well-defined, separate substances. Slaked lime needs to be listed separately, in order to be used. In comparison to the already authorized quicklime, the properties of slaked lime can be summarized as follows:

- less effective than quicklime;
- environmental impact similar or better than for quicklime;
- safety for the user clearly better than for quicklime.

Slaked lime has various uses in aquaculture (see above). Some of these uses take place in the absence of animals (e.g. disinfection of dried ponds, treatment of water prior to inlet into the rearing units), while others take place in the presence of animals (e.g. treatment of shellfish). The Group supports all these use of slaked lime. In section 4.6.2, the Group suggests a new structure for Annex VII, which avoids the need for distinction between application in the presence or absence of animals.

Conclusions
The Group concluded that the use of slaked lime is in line with the objectives, criteria and principles of organic farming as laid down in Council Regulation (EC) No 834/2007.
In Annex VII, the entry ‘lime’ should be replaced by ‘quicklime (calcium oxide) and slaked lime (calcium hydroxide)’.

4.5.8 Reconfirmation of advice from 2008

Introduction, scope of this chapter
The Group was asked to reconfirm the advice on various substances given in 2008 by an ad-hoc expert group. Due to time constraints, the Group could not make full evaluations, but indicates in which areas clarifications are most needed. The Group underlines that this preliminary advice should not be used for final decisions on these substances.

Humic acids

Humic acids are natural substances. Manufacturers claim that they improve water quality and/or reduce the susceptibility of fish towards pests and diseases. Clarifications are needed in the following areas:

- exact uses
- authorization under general legislation
- necessity and alternatives.

Iodophors

The Group sees a need for disinfection of eggs, and possibly also a limited use for equipment in aquaculture facilities and footbaths. The evaluation as a biocide is pending. Clarifications are needed in the following areas:

- necessity and alternatives for disinfection of equipment in aquaculture facilities

Potassium permanganate

Potassium permanganate is rather toxic. Clarifications are needed in the following areas:

- toxicity
- environmental impact
- authorization under general legislation
- necessity and alternatives

Formalin

Formalin (formaldehyde) is carcinogenic. It is still widely used in some countries. In Annex VII, it is listed only for terrestrial animals, and not for aquaculture. Even if formalin is not in the current aquaculture mandate, it has been previously questioned by the Group (see EGTOP report on poultry). Clarifications are needed in the following areas:

- toxicity
- authorization under general legislation
- necessity and alternatives

4.6 Proposals for regulation of cleaning and disinfection in Annex VII

4.6.1 Rationale for establishment of a 'basic list of substances for management of aquatic environments'

Introduction

Typically, synthetic disinfectants can be used only in the absence of aquaculture animals, while synthetic veterinary substances are used in the presence of aquaculture animals. This distinction is reflected by different general legislation (biocides vs. veterinary medical substances), and it is currently also reflected by two separate sections in Annex VII (section 2.1 vs. 2.2).

However, some of the substances used in organic aquaculture have an 'atypical' mode of action and do not fit well into this distinction. In some cases (e.g. seawater, sodium chloride), the use falls under different legislation (or perhaps none), depending on the intended purpose of the use. In order to avoid further complications at the level of organic legislation, the Group considers that a different way of listing in Annex VII would be prefereable. The listing concentrates on those requirements which are important from the point of view of organic principles, and leaves flexibility with respect to general legislation.

Careful evaluation of uses

The Group underlines the need for a careful evaluation of all uses of chemical substances in aquaculture. Such evaluations are already carried out in the framework of general legislation. They concern the active substance and/or the commercial product, and they take place at EU and/or member state level. A careful evaluation must take into account the substance, dosage and treatment time, as well as the fish species, the parasite or pathogen, and possibly the environment or management system.

The current distinction between section 2.1 and 2.2 is over-simplified and cannot deal with such complex interactions. There is no guarantee that a substance listed in section 2.2 can be safely used in the presence of aquaculture animals, unless there would be precise indications regarding fish species, dosage, treatment time and environmental factors. Such a level of detail would be impractical to handle in Reg. 889/2008, and it would also duplicate product authorization/registration at EU and member state level.

Proposal for establishment of a 'basic list'

The Group considers that for a few non-controversial substances, the organic regulation should not make restrictions regarding their use. This does *not* mean that these substances can be used in whatever way an operator wishes. It simply means that the organic regulation makes no *additional* requirement on top of those made by general legislation and product authorization.

Such an approach has been suggested previously by the Group for selected food additives ('basic tool box'; see EGTOP report on organic food), for disinfectants ('basic toolbox'; see EGTOP report on greenhouse production) and for plant protection products ('basic list of active substances'; see EGTOP report on plant protection products II). In analogy, the Group suggests to call this section 'basic list of substances for management of aquatic environments'.

The proposal is limited to non-controversial substances (as shown under point 2.1 of section 4.6.2 below) for which no concerns have been raised in the organic sector; for all other substances (e.g. iodophors), the Group fully supports the current practice of specifying uses in Annex VII.

With respect to *non-controversial substances*, the Group points out the following arguments:

- Even for substances where no specifications are given in Annex II, the use must follow the specifications laid down by general legislation/product authorization.

- With respect to organic farming principles, there is no added value in repeating these specifications.
- For the EU authorization process, these specifications represent an extra burden, because use categories must regularly be updated (for examples, see the evaluations of sodium chloride and a few other substances here).
- For organic aquaculture, these specifications unnecessarily delay the adoption of newly approved uses.

4.6.2 Proposed new structure of Annex VII, section 2

As explained above, the Group recommends to create as a first section within Annex VII a 'basic list of substances for management of aquatic environments'. This necessitates a rearrangement of the sections within Annex VII. If all recommendations in chapter 4.5 are followed, section 2 of Annex VII would look as follows (new text is <u>underlined</u>):

2. <u>Substances for use in aquaculture and seaweed production</u> referred to in Article 6e(2), 25s(2) and 29a.

<u>2.1 Basic list of substances for management of aquatic environments</u>
<u>Substances may be used for all purposes authorized under general legislation</u>

- sodium chloride
- quicklime (calcium oxide), slaked lime (calcium hydroxide)
- hydrogen peroxide, <u>sodium percarbonate</u>*
- organic acids (acetic acid, lactic acid, citric acid)
- humic acid
- peroxyacetic acid
- peracetic and peroctanoic acids
- limestone (calcium carbonate), dolomite

2.2 Substances for cleaning and disinfection of equipment and facilities, in the absence of aquaculture animals
<u>Substances may be used for all purposes in the absence of aquaculture animals authorized under general legislation</u>

- ozone
- sodium hypochlorite, calcium hypochlorite,
- <u>mixtures of potassium peroxomonosulphate and sodium chloride producing hypochlorous acid*</u>
- caustic soda
- alcohol
- potassium permanganate
- iodophores

<u>2.3 Substances for limited use in aquatic environments</u>
<u>Substances may be used for very limited purposes indicated here</u>

- iodophores (<u>for disinfection of eggs)</u>
- tea seed cake made of natural camelia seed (use restricted to shrimp production)
- copper sulphate; only until 31 December 2015

* new substances, for which inclusion is proposed in this report (chapter 4.5).

5. LIST OF ABBREVIATIONS / GLOSSARY

Annex VII	Annex VII to Regulation 889/2008
CAS number	CAS numbers are unique numerical identifiers assigned by Chemical Abstracts Service (CAS) to every chemical substance described in the open scientific literature. CAS numbers are given in this report, because many of the substances discussed here have multiple colloquial names.
MIST	minimal invasive surgical technique
MRL	maximum residue level (see Reg. 396/2005)
On-growing	rearing of aquaculture animals from the juvenile stage to harvest size.
PT	product type. European biocides legislation distinguishes 22 types of biocidal products (see Reg. 528/2012, Annex V). Product-type 3 (disinfectants for veterinary hygiene) is of particular relevance in the context of this report.
RAS	Recirculation Aquaculture System
The Group	The Expert Group for Technical Advice on Organic Production (EGTOP)

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