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The role of microalgae in aquaculture: situation and trends

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Abstract:

Algae are utilized diversely in aquaculture, but theirmain applications are related to nutrition. They areused in toto, as a sole component or as a foodadditive to supply basic nutrients, color the flesh ofsalmonids or for other biological activities. The needfor nutritional sources safer than traditional animalproducts has renewed interest in plants in general andalgae in particular. This report deals principallywith the nutritional role of microalgae inaquaculture. The larvae of molluscs, echinoderms andcrustaceans as well as the live prey of some fishlarvae feed on microalgae. Though attempts have beenmade to substitute inert particles for thesemicro-organisms which are difficult to produce, concentrate and store, only shrimp and live prey forfish will accept inert food, and only shrimp accept itfully. Several studies have confirmed that a live, multi-specific, low-bacteria microalgal biomassremains essential for shellfish hatcheries. Majoradvances are expected from new production systemdesigns and operations, from batch-run open tanks tomore sophisticated continuously run and closed loopreactors. Studies are underway to simplify hatcheryoperations by replacing biomass produced on-site withrun-times by that produced and preserved elsewhere. Although still promising, they have not given rise, sofar, to any application for molluscs. Otherapplications of microalgae in aquaculture, from greenwater to making salmon flesh pinker, are examined. Whether produced on or off-site, there remains thequestion of cost effectiveness of microalgalproduction systems. This can only be achieved by substantial upscaling and improved quality control.

Keywords: aquaculture - fish - hatchery - microalgae - molluscs - phytoplankton - post-larvae - shrimp

Introduction

Algae are at the base of the entire aquatic food chain, and support the production of renewable resources by some 100×10^6 t per year, from fishing. Therefore, it is not surprising that the microalgae which compose the phytoplankton play a vital role in the rearing of aquatic animals like mollusks, shrimp, and fish, and have a strategic interest for aquaculture. Moreover, there are numerous applications for molecules from these phototrophic micro-organisms in human and animal food, health, cosmetology. Some of their properties also concern the environment, supporting life in space and renewable energy production (Muller-Feuga, 1977). Macroalgae for human consumption, with a 1997 production of 7.2×10^6 t, will not be discussed here. We will mainly focus on microalgae used as food for aquatic animals, mentioning a few non-food uses. Several authors, Benemann, 1992 in particular, have already made this analysis. Our contribution aims to update and complement it, particularly in quantifying requirements.

All the fisheries and aquaculture production statistics mentioned hereafter come from the United Nations Food and Agriculture Organization (Shatz, 1999), except for shrimp, since the data from Rosenberry (1998) are more recent.

In 1997, world aquaculture produced 35 x 10⁶ t of plants and animals, mainly as human foodstuffs. This followed outstanding growth rates (an average of 10 % per year from 1984 to 1997). At a time when the harvesting and fishing of wild populations has reached critical thresholds, aquaculture's contribution to human nutrition is constantly increasing. For example, the proportion of world fish production derived from aquaculture doubled in less than a decade, from 8 % in 1984 to 16 % in 1993. Fish provides an average of 17 % of animal proteins consumed world-wide, and in some countries, this value can reach 50%.

Figure 1 shows the top ten aquaculture producing countries in 1997. The first, by far, is China with 24×10^6 t, composed principally of fresh water carp (44%) and edible algae (16%), with an average increase of 10% per year from 1984 to 1997. Other producing countries follow with India (1.8 x 10^6 t), Japan (1.3 x 10^6 t) and South Korea (0.6 x 10^6 t). Excepting the latter two, the industrialized countries each produced under 0.5 x 10^6 t per year.

In contrast to air-breathing animals, those in the aquatic medium used by humans for food are rarely herbivorous at the adult stage. The food chain is longer, and only filtering mollusks and a few other animals are true plankton feeders throughout their lifetime. Other farmed animals are carnivorous from their post-larval stage, or omnivorous at best. However, microalgae are required for larvae nutrition during a brief period, either for direct consumption in the case of mollusks and peneid shrimp, or indirectly as food for the live prey fed to small-larvae fish. In these cases, the post-larvae are hatched, bred and raised by specialized establishments called hatcheries. These systems are particularly complex to operate, since they involve artificial production of microalgae and, in the case of small-larvae fish, the production of small live prey such as rotifers. Animals whose rearing does not present these constraints are rare. This is the case of fish like salmonids, whose eggs have sufficient reserves to hatch big larvae capable of feeding directly on dry particles.

World production of the main species groups which consume microalgae, at least at the larval stage, reached around 7×10^6 t in 1997, *i.e.* 18% of world aquaculture production. They include (Fig. 2) filtering mollusks, peneid shrimps, and small larvae fish like sea breams, turbot and other flat fish.

The present trend is to avoid using microalgae because they are difficult to produce, and therefore raise investment and wage costs. Although it has been established in numerous circumstances that they are vital for the artificial reproduction of mollusks, their use can be limited for the reproduction of peneid shrimp and of some species of fish. We shall examine these stock's microalgae requirements, and attempt to define their orders of magnitude and their trends. Our approach consisted of a preliminary assessment of the microalgae requirements for one million (10⁶) post-larvae. In the second step, we estimated the number of post-larvae required to achieve full production in the main categories. Both potential and detailed requirements are provided in order to show the upper and lower limits (Table 2). Of course, this approach is inherently inaccurate owing to the wide range of sizes, nutritional values, and habits of use of microalgae all over the world. But it has the advantage of providing quantitative indications of requirements and consequently gives some idea of the predominant masses which require more attention.

Filtering mollusks

The filtering mollusks such as oysters, scallops, clams and mussels $(7.4 \times 10^6 \text{ t in } 1997)$ are herbivorous and consume microalgae throughout their lives. However, the filtration is not selective and these animals are also suspension feeders, taking in living or dead, plant or animal particles which compose plankton. Those filtering mollusks are mainly oysters $(3.1 \times 10^6 \text{ t})$, clams $(1.9 \times 10^6 \text{ t})$, pectinids $(1.3 \times 10^6 \text{ t})$, and mussels $(1.1 \times 10^6 \text{ t})$. Figure 2 shows that the mollusk production is by far the highest for microalgae-consuming species. After a sharp increase in the early 90's, probably due to the availability of new statistics, the progress has slowed. These productions rely on wild phytoplankton present in the natural water masses circulating around the livestock in the open medium.

How much of the phytoplankton biomass is consumed in mollusk farming? If we assume that, because of their shell, the organic part of filtering mollusks production represents 1/5 of the total amount mentioned in the statistics, and that the yield of the phytoplankton to mollusk transformation is 1/10, the total consumption of phytoplankton in 1997 would be about 1.5 x 10⁶ t dry weight (DW), assuming that the livestock is constant all year long and equal to the annual production. This figure is five orders of magnitude lower than overall annual ocean primary production, which can be set at 10¹¹ t DW (Pauly & Christensen, 1995; Longhurst et al., 1995).

As this biomass is produced naturally, the farmer can simply expose his livestock to circulating water masses to take advantage of the natural resource. It is another story when larvae, then post-larvae, are produced in a hatchery, *i.e.* in artificial conditions which eliminate the most penalizing natural hazards. In this case, fodder microalgae must be produced artificially to meet the food requirements of larvae, post-larvae and even broodstock.

Since mollusk larvae rearing techniques were developed in the 60s, microalgae have remained the only food used, although new solutions like yeast, bacteria, microparticles, slurry, paste, dried and frozen microalgae have been explored (Robert & Trintignac, 1997). None of them is sufficiently advanced to date to provide an alternative to live microalgae. The new preparations often present deficiencies, or become a substrate for adverse bacterial development, especially in the early stages.

For most species, phytoplankton requirements differ, depending on whether they are for broodstock, larval or post-larval rearing. The larval stages require high bacteriological and biochemical quality, but in small amounts, for a short time. Post-larvae accept lower quality, but remain sensitive to the biochemical composition and

require amounts nearly a hundred times greater, depending on the length of the nursery stage. The preparation of a broodstock for breeding requires both quality and quantity, but the number of animals is small. Thus, although mass production of live microalgae in the hatchery has been mastered, it is subject to large quantitative and qualitative constraints, summarized in Table 1 for the Pacific oyster *Crassostrea gigas*.

Typically, a commercial hatchery operates about 8 to 10 months a year. Once they exceed 3 mm, the animals are generally transferred to an open medium or grown in outdoor nurseries. Under such conditions, algae consumption is even higher, from 40 to 100 m³ of 10⁶ cells/ml in extensive culture per 10⁶ juveniles (6 to 12 mm). As shown in Table 1, one million 0.2 to 3.0 mm post-larvae require about 14 kg of microalgae (DW). The species of microalgae commonly utilized are *Isochrysis galbana affinis Tahiti*, *Skeletonema costatum, Pavlova lutheri, Chaetoceros calcitrans*, whose mean dry cellular weight is about 20 pg.

France produced 147,150 t of oysters in 1997, which theoretically required about 5×10^9 post-larvae. The collecting of wild spat on artificial substrates remains the main source of supply in this country and hatcheries cover 10% of requirements. In fact, the overall production of hatcheries is some 500 10^6 post-larvae. The European Atlantic coast's production ranges from 600 to 800 10^6 post-larvae (R. Robert; pers. comm.). Under these conditions, the microalgae production as calculated from the above ratio is between 8 and 11 t DW per year for the post-larvae production in this region.

On the western coast of the USA, 80% of post-larvae production comes from commercial hatcheries. The main one is operated by the Coast Seafoods Company, which produces 20 x 10⁹ eyed larvae per year, sustaining production of 40,000 t of market size oysters, which is just under half of USA oyster production (98,148 t in 1997). The

requirement of microalgae for this production is about 20 t DW per year, according to the ratio given above.

With 3.7 x 10⁶ t, China alone produces 68% of the world's filtering mollusks, 4 to 5 times more than the European and American continents combined. Thus, any inaccuracy regarding Chinese yields will have an amplified effect on our estimate. For instance, farming of *Argopecten irradians*, the bay scallop, has rapidly expanded in China since its introduction in 1982 with 200,000-300,000 t produced in 1997 (Tang & Fang, 1999). As all of the spats come from hatcheries, and assuming that our ratio is suitable for this species of scallop, this production would require over 300 t DW of microalgae, which greatly surpasses western production. Therefore, it seems risky to attempt an estimation of world requirements without complete information on Chinese production. However, we can say that world requirements would have exceeded 10,000 t DW in 1997, if hatcheries had been the sole source of juveniles. This is a high upper limit considering that wild spats are still collected world-wide.

Other uses of microalgae consist in refining the oysters prior to sale. In France, an intensive technique based on producing the diatom *S. costatum* in subterranean salt water doubles the flesh content and triples the glycogen content in 30 days at temperatures ranging from 8 to 12°C, resulting in a substantial increase in the market price. Another technique called the "greening" of oysters, which consists in their acquiring a blue-green color on the gills and labial palps, raising the product's pre-market value by 40%. The agent responsible for this is a pigment produced by the diatom *Haslea ostrearia* which grows naturally in ponds on the western coast of France. This refining process puts the oyster in contact with naturally or artificially grown algae (Barille et al., 1994). Then, in

an attempt to improve the final product quality, these new processes promote microalgae consumption.

The main threat to world shellfish culture consists in epizootic diseases which could decimate the livestock and harshly affect business. This occurred in Europe with a virus disease in the Portuguese oyster *Ostrea angulata* (Grizel & Heral, 1991), with *Bonamia* in the flat oyster *Ostrea edulis* (Grizel & Tige, 1982) and, though to a lesser extent, with the brown ring disease in the clam *Tapes philippinarum* (Paillard et al., 1994). Genetic research for disease-resistant strains is important to shellfish farmers. In France, public research has focused on this objective since the beginning of the decade. But genetic breakthroughs can only be transferred to industry if traditional wild spat collecting practices are abandoned for hatchery supply. Recourse to hatchery products will be generalized once the products of genetic selection are on the market, as often seen in animal husbandry. *Bonamia*-resistant flat oysters are expected for the early years of the coming millennium, while studies have begun for the selection of an immunity-reinforced strain of *C.gigas*.

Shrimp

Shrimp farming production reached 737,200 t in 1998, an increase of 12% from 1997 (Rosenberry, 1998). This mainly takes place in subtropical regions of America (28%, 457 hatcheries) and south-east Asia (72%, 3,718 hatcheries). Thailand is the main producer with 210,000 t, followed by Ecuador with 130,000 t in 1998. Production systems in the two groups of countries use microalgae differently. They are necessary from the second stage of larval development (zoea) and in combination with zooplankton from the third stage (myses). So, although of short duration, those larval stages require microalgae

culture facilities which vary with the size of the hatchery and the level of control of medium parameters.

We can distinguish "green water" hatcheries from "clear water" ones. The former are small and medium-sized hatcheries associated with the on-growing farms of southeast Asia, where operations rely more on experience than on mastering techniques.

Naturally occurring microalgae blooms are encouraged in large ponds with low water exchange where the larvae are then introduced. Sometimes fertilizers and bacteria are added to induce more favorable conditions. This production system, with poor control of microalgae, provides the better part of shrimp production. On the other hand, large-sized hatcheries require highly paid technicians, multimillion dollar investments, and highly controlled medium conditions. Those hatcheries are mainly located on the American continent. The observed trend is toward specialized production, particularly with the supply of post-larvae in the hands of big, centralized hatcheries. They open a pathway to new techniques, especially the genetic selection of strains with stronger immunity.

It takes about 1 m³ of 3.10⁶ cell/mL microalgae culture to produce 10⁶ post-larvae, that is to say, at the rate of 20 pg per alga, about 65 g DW (G. Cuzon; pers. comm.). This is only valid for clear water hatcheries. But, in green water hatcheries, since microalgae contribute to stabilizing and improving the quality of the rearing medium while providing food for the zooplankton, they are produced in far greater quantities than the strict needs of larvae feeding. In the latter case, the figure given above is a lower limit and should be multiplied about tenfold.

The larvae feed consists in a combination of microalgae and early stages of the phyllopod crustacean *Artemia* sp., as well as dry food proposed on the market or manufactured locally. The main microalgae genera used are *Skeletonema*, *Chaetoceros*,

Tetraselmis, Chlorella and Isochrysis. Although widely used, dry formulated feeds do not work on a 100% replacement basis. Even when they are used, microalgae culture systems are kept in operation for emergencies. However, the trend is towards reducing or even avoiding recourse to microalgae.

Small larvae fish

The use of microalgae in fish hatcheries is required for both production of live prey, and maintaining the quality of the larvae rearing medium. It could also be used in the formulation of dry fish food for on-growing.

The use of small, live, plankton feeder preys, namely the rotifer *Brachionus* plicatilis, is still a prerequisite for success in hatcheries of marine small-larvae finfish like sea breams (130,964 t in 1997) and flat fish (38,203 t in 1997). These preys can be raised on yeast-based artificial feeds, but this is much less efficient than with phytoplankton. Microalgae present an interest on three levels: (i) quick recovery of rotifer populations after collapse (7 to 13 days, compared to 20 to 35 days with yeast); (ii) improved nutritional quality of live prey; and (iii) lower bacterial contamination, especially from *Vibrio*. For numerous fresh and sea water animal species, the introduction of phytoplankton in rearing ponds leads to much better results in terms of the survival, growth and transformation index than when effected in clear water. Moreover, for sea bream, this condition has became an economic necessity.

The reasons behind the positive role of microalgae in the larvae rearing ponds of fish, as well as shrimp, have not been completely elucidated. There is no doubt that water quality is improved and stabilized by oxygen production, pH stabilization, etc., but this does not explain everything. The action of some excreted biochemical compounds is generally mentioned, as well as the induction of behavioral processes like initial prey

catching. Other positive functions such as regulating the bacterial population, probiotic effects and stimulating immunity, have also been suggested, but they are not sufficiently understood. So far, only their action as a raw material has been considered, giving rise to what are called "green water" and "pseudo-green water" techniques (Dhert et al., 1998).

In the case of the sea bream *Sparus aurata*, the microalgae requirement for the rearing and enrichment of rotifers is 6 x 10⁹ cells for a 60-day old juvenile, which represents about 0.06 g DW per juvenile (N. Papandroulakis; pers. comm.). This result was obtained using the pseudo green-water technique, which consists in introducing algae produced elsewhere into the rearing medium. This technique is particularly efficient, and the use of the previous ratio gives an evaluation which rather minimizes the requirements. However, if we generalize this ratio to world production of small larvae fish, the microalgae requirement can be set at a minimum of 51 t DW per year in 1997.

Because of essential long chain polyunsaturated fatty acids (PUFA) requirements, fish farming is dependent on marine lipids. Formulated dry feeds for intensive fish rearing are composed of 30 to 60 % meal and 10 to 20 % marine fish oil, generally from clupeids. The most commonly accepted predictions for the year 2020 are for 220 x 10^6 t of aquatic products, 100×10^6 t of which will come from aquaculture. With this prospect, the 20 to 30×10^6 t of fish now available for reduction into meal and oil will not meet more than 5 to 7% of the demand for formulated dry feed for fish farming. If we also consider the specific requirements of terrestrial animals and man, the shortage of essential PUFA could amount to 10 to 15×10^6 t in 2020, if nothing is done (P. Divanach; pers. comm.). Though inconceivable today due to high cost- prices, the use of microalgae as a commercial source of PUFA (Apt & Behrens, 1999), and even of energy, high quality proteins, vitamins and sterols, seeing their high content, remains a potential solution. The

combination of price increases for fish oil, due to a growing shortage, and improved cost effectiveness of other sources (including genetically modified organisms) will make substitution possible in future. Considerable research is focused on this problem world wide.

Astaxanthin and canthaxanthin are the only pigments that can fix in the flesh of salmonids, whose pinkening represents a US\$ 100 million, rapidly expanding market (Verdelho & Baylina, 1995), almost entirely held by the Swiss firm Hoffmann-La Roche. This feed additive is produced by chemical synthesis and available at a price of US\$ 3000 /kg. Consumer tastes are such that demand for natural products is increasing. Today, the biological supply sources for astaxanthin are the yeast *Phaffia rhodozyma* (Sanderson & Jolly, 1994), despite its low content (0.4 %), marketed by the Dutch company Gist Brocades, and the fresh water chlorophycea *Haematococcus pluvialis* (Borowitzka et al., 1991), containing up to 5 %. Some companies, such as Algatec-Sweden, Norbio-Norway, Biotechna-UK, Aquasearch, Cyanotech, Maricultura, Danisco Biotechnology and Oceancolor -USA expect to enter the competition. Assuming that the penetration rate on this market of astaxantin from *H. pluvialis* is 10%, the overall production of this algae would reach 20 t DW per year.

Discussion

The world microalgae requirements for hatcheries examined above are summarized in Table 2. Whereas the potential requirements, calculated as if hatcheries were the sole source of post-larvae, exceed 10,000 t a year, adding up all productions detailed above gives a minimum of 531 t a year. The wide gap between these extreme limits should be narrowed in future principally thanks to better visibility of Chinese mollusk hatcheries production. The situation in that country still seems similar to that described by Newkirk

(1991) who stated that the gain in production of the traditional and new species was a well kept secret.

In fact, the major part of world microalgae requirements comes from mollusks for which no substitution is yet possible, and for which China is the main and growing producer. Though microalgae production for aquaculture involves several species, making for a complicated system, there is no need for cropping since the algae are used as row cultures in rearing tanks or ponds, simplifying the post-culture processes. This production is probably on the same order as that of *Spirulina* which ranges from 2,000 to 4,500 t DW per year, depending on the source.

Most of microalgae requirements are supplied today by firms in-house, growing them in specialized units, or within the larvae rearing tanks. This is less due to a desire for independence than to the need for immediate availability of live microalgae. A supply of live and concentrated microalgae products at competitive prices would probably lead to sweeping changes in hatchery production techniques. In fact, algae culture generates high investment and running expenses, which producers want to minimize. Benemann (1992) estimates that this in-house cost price of microalgae ranges from US \$ 250 to 1000 per kg DW, whereas the large facilities specialized in commercial microalgae production, which operate highly controlled production systems like closed photobioreactors (Borowitzka, 1996), market their products at substantially lower prices, between US \$ 50 and 300 per kg DW. This difference makes it possible to bear additional costs brought about by preservation, storage and delivery of special products to hatcheries. Recently developed techniques to produce and preserve microalgae could create a favorable situation for the rise of these new products. Heterotrophically-grown microalgae seem to be an inexpensive production means (Gladue, 1998) as they are produced in high density by

classic fermentation. In Japan, freshwater microalgae of the *Chlorella* genus are already widespread on the rotifer production market and consequently, most fish hatcheries do not include a microalgae production facility. Though difficult to evaluate, the demand of aquarium owners complements that of hatcheries.

But the potential consumer must first be convinced of the efficiency of such products. Numerous studies have been devoted to the subject over the last decade. For example, a European program (Muller-Feuga et al., 1998) set out to examine the conditions for substituting hatchery algae by ones produced elsewhere, concentrated, processed for storage and transportation, for larval rearing of the sea bream *Pagrus aurata*, the oyster *Crassostrea gigas*, and the scallop *Pecten maximus*. Results were encouraging for the sea bream, and mostly negative for mollusks. Standards of preserved microalgae consumption for sea bream have been set, and the need for several species of live microalgae with low bacteria levels has been confirmed for mollusks. The economic stakes are attractive enough to mobilize even stronger international research efforts on commercial species larvae nutrition, mainly focusing on PUFAs and other essential compounds, where microalgae would compete with formulated dry feeds.

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Table 1. Microalgae nutritional requirements and rearing conditions of the oyster *Crassostrea gigas* at different stages (R. Robert, in Muller-Feuga, 1997).

	1 breeder	10 ⁶ larvae	10 ⁶ post-larvae	
			(0.2-3.0 mm)	
Amount of microalgae	0.5 to 2.0	15 to 20	1,000 to 1,500	
in L/day at.6.10 ⁶ Cell/mL				
Multispecific mixture	yes	yes	recommended	
Bacteriological quality	normal	good	good	
Duration of the step	1 to 3 months	0.5 to 1 months	2 to 3 months	

Table 2. Upper and lower estimates of microalgae dry weight biomass production required by the post-larvae of world aquaculture in 1997 (1998 for shrimps), and mid-term trends of this production. Upper estimates are calculated according to aquaculture productions, by multiplying the number of post-larvae required for these productions and the microalgae diet ratio given in the text. Lower estimates are the summing-up of the productions stated in the text (^a due to increased hatchery contribution; ^b due to increased formulated feed use; ^c in proportion with production). Sources: FAO for Mollusks and small larvae fish, Rosenberry (1998) for shrimp.

	Aquaculture	Number of	Overall	Microalgae	Microalgae biomass		Trends
	productions	10 ⁶ post-larvae	10 ⁶ post-	requirements	(t d. wt per year)		
	(t/year)	per t of final	larvae	per 10 ⁶ post-	Upper	Lower	_
		product		larvae	estimates	estimates	
				(kg d. wt)			
Mollusks	7,442,555	0.1	744,256	14.0	10,420	330	Sharp increase ^a
Shrimp clear water	206,416	0.3	68,805	0.06	4	4	Decrease ^b
Shrimp green water	530,784	0.4	224,786	0.65	146	146	Increase c
Small larvae fish	169,167	0.005	845	60.0	51	51	Increase c
Total	8,348,922				10,620	531	

Figures and legends

Figure 1. Aquaculture productions of the top ten producing countries, and their variation from 1984 to 1997 (each bar corresponds to a year). Source: FAO.

Figure 2. Aquaculture productions of the main groups of species consuming microalgae at juvenile stages, and their variation from 1984 to 1997. Source: FAO.



