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## THE BREEDING AND CULTIVATION OF MARINE FISH SPECIES FOR MARICULTURE.

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# Colin E. NASH

The Oceanic Institute, Makapuu Point, Waimanalo, Hawaii 96795, U.S.A.

## ABSTRACT.

The culture of the marine fish has not made as much progress as other species in aquaculture, for example the marine and freshwater prawns. The large number of red seabream released each year by the Japanese into the Seto Inland Sea constitutes one of the major marine fish projects, but the economic benefit for this type of ocean ranching is uncertain, and the cost of the fry is high.

Dover sole and turbot still offer good potential for marine fish culture in European and Mediterranean waters, but many of the farming techniques have been developed for the less valuable but more easily produced plaice.

The grey mullet has increasing farm potential but its commercial markets are limited as it has low value. It is more important as a subsistence brackishwater pond fish in Southeast Asia.

In order to make marine fish culture or farming more successful during the next twenty years, there is great need for a concentration of effort on a few selected species. There is also need for good facilities built on experiences derived from many workers in the field. Taking the lead from the salmonids, it might be very opportune to concentrate on the migratory species of marine fish, which provide far greater opportunities for total management through ocean ranching. The migratory species are more likely to provide the gross tonnages of fish to increase market levels much more than the individual high priced fish cultured in small farms, either on land or in coastal areas.

#### RESUME.

L'aquaculture des poissons marins n'a pas progressé aussi rapidement que celle d'autres espèces, comme, par exemple, les crevettes. Les grandes quantités de daurades royales relâchées chaque année par les Japonais dans la Mer Intérieure, représentent l'un des plus importants projets en cours, mais le coût des juvéniles est élevé, et l'intérêt financier de ce type de repeuplement est incertain.

La sole et le turbot sont des espèces prometteuses pour les côtes atlantiques de l'Europe et la Méditerranée, mais la plupart des méthodes d'élevage ont été mises au point sur la plie, plus facile à produire, mais de moindre valeur.

Les perspectives d'élevage du mulet gris progressent, mais sa valeur commerciale est faible, et les marchés sont limités. Il est surtout important comme aliment de subsistance en Asie du Sud-Est.

Pour obtenir plus de succès dans les vingt années qui viennent, une concentration des efforts sur quelques espèces est indispensable. Il faut aussi de bonnes installations, conçues à partir de l'expérience de nombreux spécialistes. En prenant l'exemple des salmonidés, il pourrait être très intéressant de concentrer les recherches sur des espèces migratrices, qui offrent d'intéressantes possibilités d'élevage en mer libre. Les espèces migratrices sont mieux susceptibles de fournir de gros tonnages, pouvant accroître les marchés, que des espèces chères, élevées dans de petites fermes, à terre ou dans des lagunes côtières.

#### INTRODUCTION.

Of all the marine fish species, excluding anadromous salmonids, the marine flatfish of the order *Pleuronectiformes*, have been cultured the most successfully throughout the last twenty years of contemporary aquaculture. The marine flatfish as a group are a desirable market commodity and valuable to the fishing industry. The total annual landings of the flatfish are approximately 1.7% of the total world catch, or 1.18 million metric tons (FAO, 1974).

The use of brine shrimp nauplii, Artemia salina, by ROLLEFSON (1940) and the basic research on the plaice, *Pleuronectes platessa*, by SHELBOURNE (1964, 1967) were probably more responsible for renewed investigations into marine fish culture and aquaculture than any other single event since the hiatus of propagation and culture for ocean ranching in the last two decades of the nineteenth century. Their results, followed by those of others (FLUCHTER, 1965), led to the mass propagation from breeding adults of large numbers of plaice and Dover sole, *Solea solea*, and their culture in a number of locations (SHELBOURNE and NASH, 1966; NASH and SHELBOURNE, 1967).

Recent work with the turbot, Scophthalmus maximus (JONES, 1972 ; PURDOM et al., 1972), the Black Sea turbot, Scophthalmus maeoticus (SPECTOROVA et al., 1974), the brill, Scophthalmus rhombus (JONES, 1972), lemon sole, Microstomus kitt, (HOWELL, 1972), has enabled these fish to be included among those which can now be reared in reasonable numbers from breeding adults. The most attractive flatfish to the fishing industry is the Atlantic halibut, Hippoglossus hippoglossus, and the Pacific halibut, Hippoglossus stenolepis, but to date only some preliminary capture work has been achieved by the Canadians and the British. The size of adult halibuts is intimidating, making them difficult to catch and keep alive, and to form a broodstock in captivity without adequate facilities. The most attractive flatfish species to the culture industry which can be reared are therefore the turbots and the soles.

All these species are temperate water fish of the Atlantic and the North Sea, but some flatfish range into the colder waters around Greenland, or into the warmer waters of the Mediterranean and the Black Sea. The countries with an opportunity to culture marine flatfish are therefore many and are distributed across a broad range of latitudes.

Although the marine flatfish have dominated the marine fish culture scene, other equally important results have been obtained. Complete production cycles have been developed for other tropical and sub-tropical species. For example, excellent techniques have been established for the live-bearing topminnow, *Poecilia vittata* and other bait fish species (BALDWIN, 1975), the threadfin or moi, *Polydactylus sexfilis* (MAY, 1976), the red seabream, *Sparus major* (HANAMURA, 1976) and the rabbit fishes of the genus *Siganidae* (LAM, 1974). All these marine fish have local important commercial value.

Other than these examples, breeding and culture successes predominantly involve captive temperate water fish and, for the most part, are still confined to rearing a small percentage of larvae through metamorphosis to juvenile stages and subsequent grow-out.

None of the species has been carried through pilot scale hatchery production or farming. For example, three White Sea species, the navaga, *Eleginus navaga*, polar cod, *Boreogadus saida*, and Arctic flounder, *Liopsetta glacialis*, (ARANOVICH *et al.*, 1975) have been reared in the laboratory ; and the sea bass, *Dicentrarchus labrax* (BARNABE, 1974) can be produced in good numbers on the large laboratory scale. Similarly, successes have been achieved with several other important captive marine species, such as the pompano, *Trachinotus carolinus* (FINUCANE, 1970), the anchovy, *Engraulis mordax* (LASKER *et al.*, 1970; HUNTER, 1975) and the gilthead, *Sparus aurata* (RENE, 1974). Many other marine species have been reared from eggs taken from plankton tows (BLAXTER, 1968; HASSLER and RAINVILLE, 1975) but these are excluded from this review.

Mass propagation has been achieved with two species of mullets which breed at sea, namely the grey mullet, *Mugil cephalus* (LIAO, 1969; KUO *et al.*, 1973) and the white mullet (HOUDE *et al.*, 1976). These are two species which have a broad range of distribution and are important subsistence fish for developing countries. For all these examples, with the exception of the grey mullet, no culture practices have been taken beyond the laboratory or experimental stages. A hatchery has been proposed for grey mullet propagation in Hawaii, and is in the process of being constructed.

In summary, marine fish propagation has progressed slowly over the last twenty years. The greatest successes are still attributed to the culture of the marine flatfish, particularly the plaice and Dover sole.

# ADULTS IN CAPTIVITY.

Nearly all the marine fish species considered desirable for culture because of their high market value have been held in captivity for long periods with little effort. The golden age of culture and attempted ocean ranching at the turn of the century confirmed that many adult fish could be caught and artificially fertilized at sea, or moved great distances to the coastal hatcheries that were being constructed at the time (IFC, 1908; GROSS, 1947).

Since then, most of the marine laboratories of the world have worked on the biology of these species and have continued to hold adults in captivity for research or public aquaria. Apart from the Pacific and Atlantic halibuts, all the commercial flatfish species have been contained before and through spawning activity, and it is true to say that the practices for obtaining viable eggs from these fish are almost one hundred years old.

The intensive effort and expense utilized in the early days of marine fish cultivation are both needless and prohibitive. Modern methods and management can sustain the same populations of adult broodstock for many years with few annual replacements. The requirements are for good husbandry and correct feeding in the right environmental conditions, and the production of viable eggs is assured.

Artificial stripping and spawning in captivity are not practiced on a wide scale although simple to perform and do little harm to the scale-less fish. More care has to be taken with scaled fish. Spawning stock are brought in from the sea and are usually permitted to spawn naturally in onshore tanks. The fish are first introduced into these tanks well in advance of the spawning season. Experience has shown that it is better to have the spawning stock on site and accustomed to their environmental conditions at least six months prior to spawning. Thereafter, a third of the stock are replaced each year.

The use of controlled methods with hormone injections for the breeding of captive fish has been reviewed by SHEHADEH (1970). Injection with salmon gonadotropin, followed by natural spawning, has been essential for the successful breeding of the grey mullet (SHEHADEH and ELLIS, 1970). LIAO (1969) had used the homogenates of mullet pituitaries to achieve similar results. Induced spawning out of season with this species has also been effected with environmental manipulation using a photoperiod of 18D/6L at 20° C for a period of 120 days (KUO *et al.*, 1974).

Induced spawning has been attempted with many marine fish. It has been used to induce final ovulation and oviposition. Oviposition was attained with turbot (FLUCHTER, 1972) threadfin (MAY, 1976), pompano (FINUCANE, 1970), milkfish, *Chanos chanos*, (NASH and KUO, 1976), sea bass (BARNABE, 1974), gilthead (RENE, 1974), and eels, *Anguilla anguilla* and *Anguilla japonica* (SHEHADEH, 1970). Of those that were induced through oviposition, the quality and/or quantity of eggs was poor and few could be fertilized. MAY (1976) reported subsequently that the threadfin spawned naturally without the aid of hormone treatment.

Correct nutrition for all broodstock is important and breeding fish require certain nutrients which can only be supplied in the diet. In the absence of a complete knowledge of the nutritional requirements of spawning stock, fish are fed with fresh natural foods at intervals between their standard ration of prepared food at about 3 % of their body weight per day for carnivores (SHELBOURNE, 1964 ; HOUDE, 1973), and providing a small supplement to the naturally available feed in ponds for omnivores or herbivores (SHEHADEH *et al.*, 1973).

Natural spawning of flatfish has occurred in many irregular shaped tanks and pools both indoors and outdoors. Modern methods, which require good control of the environmental conditions, have been developed for protected indoor tanks about 4' deep and of floor area 120-150 square feet. Exposure to the natural diurnal photoperiod is preferred by way of adjustable skylights which reduce the light intensity to between 100-350 m.c.

The tanks are supplied with pumped tidal seawater at  $6-10^{\circ}$  C at such a rate that the volume of water in each tank is replaced four times each day. The fish are held in the tanks at a density of one fish per four square feet of tank bottom. The level of dissolved oxygen in the water is maintained at over 80 % saturation. The salinity is entirely dependent on the locality of the area but should be as high as possible, preferably between  $30-40^{\circ}/...$ 

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Spawning fish are stocked in the ratio of one female to two or three males. This ratio is necessary to reduce the chances of including a high proportion of immature and infertile male fish in the population.

The fish spawn freely in late winter and early spring and the fertilized eggs, which for all the most desirable species are pelagic, are skimmed from the water surface with a net and transferred to the hatchery for incubation.

Artificial stripping and spawning reduces the amount of tank space needed for the holding of broodstock but the practice has not been relied on to meet the demands of a prototype commercial flatfish hatchery. It is however worthwhile trying to use this form of fertile eggs production (as is achieved for the salmonids) providing that the expense of obtaining new broodstock each year is accepted.

## FECUNDITY.

Compared with the fecundity of the freshwater and anadromous fish, the fecundity of the marine fish is extremely high. At the present low level of marine fish propagation technology, this high fecundity is a disadvantage. Marine fish eggs are invariably small (less than 2 mm in diameter), and the emerging larvae are equally small with little feed reserves in the yolk-sac. The larvae are therefore delicate to handle and require feeding usually within two to seven days.

The following table illustrates some of the reasons for the success of salmon and trout production in hatcheries, why the plaice and Dover sole are among the first marine fish species to be cultured, and why turbot, mullet and rabbit fish, etc., have proved more difficult.

Species	Diameter (mm)	Fecundity (n x 1000)
Salmon	6.0	1 - 4
Trout	4.0	0.5 - 1
Plaice.	2.2	250 - 500
Lemon sole	1.4	150 - 650
Dover sole	2.2	150 - 350
Turbot	1.0	1000 - 4000
Milkfish	1.1	2000 - 5000
Halibut	0,9	2000 - 9000
Mullet	0.9	750 - 1000
Moi	0.8	200 - 500
Rabbitfish	0,60	200 - 500

 TABLE 1 : Approximate diameters (millimeters) of eggs and fecundity of some marine species, and salmon and trout.

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EGG INCUBATION.

The viable eggs of most marine species are pelagic. Non viable eggs may float or stay suspended for several hours before becoming opaque and sinking out. Bouyancy of the eggs is maintained by the oil globule. Eggs with high viability are thought to have a single oil droplet (NASH and KUO, 1975). This is very evident for the grey mullet (LIAO *et al.*, 1971). However, LAM (1974) records multi-oil droplets in viable eggs, and others have observed the same with other species.

Pelagic eggs are removed at interval from the spawning ponds or tanks with fine nets, and taken to the incubators containing seawater at temperatures and salinities as close to that of conditions in the spawning location. After 24 hours, during which time the majority of the infertile eggs sink, the viable eggs are transferred to the main incubation tanks in constant temperature conditions. Conditions for incubation are extremely important and are believed to be more critical for control than the close regulation of the larval rearing conditions (NASH and KUO, 1975). The importance of temperature, salinity, oxygen level and light have been illustrated by JOHANSEN and KROGH (1914), SHELBOURNE (1964), and BLAXTER (1968), among many others.

Increased survival was indicated by SHELBOURNE (1964) incubating flatfish eggs in static filtered seawater under controlled conditions and with the addition of the antibiotics sodium penicillin G (50 I.U.) and streptomycin sulphate (0.05 mg/ml). Similar use of these antibiotics was tried initially by other workers, but are at present not widely used.

Egg density in the incubators is important and is dependent on the method of incubation being static or agitated by mechanical stirrers or aeration. In the static technique for flatfish, the egg density is about 2,000 eggs/ft<sup>2</sup> of water surface (SHELBOURNE, 1964). The grey mullet eggs (NASH and KUO, 1974) are incubated in aerated tanks at a density of 250 eggs/liter. Depending on the levels of circulation, egg densities within this range are probably desirable.

Throughout incubation the eggs are sensitive to environmental change and light intensity. Incubation is preferred indoors under subdued light and with regulated temperature and salinity controls. The eggs of many tropical and semi-tropical marine fish hatch within a period of 24 - 72 hours. Temperate water fish usually take several weeks, and SHELBOURNE (1964) operated a schedule increasing the temperature 0.5° C per week for the six weeks of plaice incubation.

From a large broodstock (which is very desirable) the total number of eggs stocked in incubation tanks or a hatchery is estimated to be about 20 % of the eggs collected from the spawning tanks throughout the season. This is because the quality and viability of the eggs varies, and it is better to confine collection of eggs for the hatchery to the middle of the season. The survival of good quality eggs during incubation is high (over 95 %) providing that a good technique is closely followed, and the incubators are cleaned of eggs which die and sink to the bottom. There can be a variation in technique before hatching depending .../...

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on the species. The eggs may remain in the same static incubator for larval rearing, or they may be removed from circulated incubators as they can withstand a certain amount of careful handling by this time. As the larvae will require progressively more space it is better to have incubators as separate units to the larval rearing containers.

## LARVAL REARING.

Rearing the larvae of the marine species continues to prove the major factor restricting the hatchery propagation and farming of the more valuable fish. The problems associated with larval rearing have been summarized by HOUDE (1973) and NASH and KUO (1975).

Successful larval rearing is influenced strongly by many of the same factors that effect incubation of the eggs. For example, salinity (HOLLIDAY and BLAXTER, 1960; DOROSHEV and ARONOVICH, 1974), temperature (LASKER, 1964; RYLAND and NICHOLS, 1967), oxygen (SYLVESTER et al., 1975), light (BLAXTER, 1968), and density (FLUCHTER, 1972 b), have all been identified by these and many other workers. However, probably the key factor preventing successful propagation is the lack of suitable larval nutrition and the related factors of feed density and water quality. A review of the many different feeds provided to marine fish larve through the years has been provided by MAY (1970). Larval nutrition has been diversely studies from mechanically regulated feeding trials on particulate size (BARNABE, 1976) to detailed bioenergetics (EHRLICH, 1974).

Many marine fish are cultured successfully with a series of live feeds. The presence of phytoplankters in the rearing tanks is common. Many phytoplankters absorb metabolites effectively and maintain ecological stability in the tanks. They also provide nutrition for the rotifer *Brachionus plicatilis*, which is now used widely for the first feed of many species reared in large or small numbers.

Rotifers are usually followed by day old nauplii of the brine shrimp Artemia salina. Intermediate feeds with copepods and amphipods have proved to be beneficial, and survival of the larvae through the early critical stages has been increased for very many species.

Artificial feeds have been compounded from both natural and synthesized materials and tried on a variety of species with mixed results. Microencapsulated or flaked feeds have been used by very many workers and reported widely. Microencapsulation prevents the hazards of tank fouling which is prevalent when other prepared feeds are used, but miniaturization and digestibility of the capsules for first feeding larvae are major problems.

Using the best techniques available, plaice and Dover sole have been produced consistently by many workers and anticipated larval survival at metamorphosis (6 weeks) is as high as 40 % or over. Turbot and lemon sole have been reared through metamorphosis with a lower survival (less than 10 %) as have grey mullet, red seabream, and many other species. However, not all have been produced at such levels consistently, and the reasons for the lack of repeatibility are far from being clear.

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