

Marine Fish Culture in Britain

I. Plaice Rearing in Closed Circulation at Lowestoft, 1957-1960

By

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Introduction

Intensive and competitive trawling with modern gear can keep fish populations well below the natural level supported by the sea. A reduction in fishing activity, such as occurred in the North Sea during the two world wars, enables fish stocks to recuperate. BORLEY (1923) calculated the catch of plaice per day's absence in the year following the 1914-18 war to be more than double the catch before. Similarly, after the second world war, MARGETTS and HOLT (1948) found the ratio of weight densities 1946:1938 for the North Sea stock to lie between 2.8 and 3.6, with a two-fold increase in numbers and little change in growth rates up to ten years old. The actual increase in plaice abundance due to wartime lay-off was probably in excess of these figures, since trawlers were already fishing hard in 1945.

We may conclude, therefore, that when intensive fishing prevails, as it does today in our near-water trawl fisheries, the food biomass available to fish is not fully exploited. Since the latter part of the nineteenth century, fishery biologists have been trying to resolve the overfishing problem, and have proposed such palliative measures as limits on the size of marketed fish, close seasons, catch quotas, mesh-size regulation, the transplantation of fish from poor to rich feeding grounds and artificial propagation. All these proposals come under the heading of large scale fish husbandry, but this paper and subsequent publications in the series are confined to one aspect, that of artificial propagation, which may have direct application today in depleted fisheries, and which will assume an increased importance in the controlled marine farming techniques of the future.

A Short History of Marine Fish Hatcheries

G. O. SARS (1866) was the first naturalist to perform a successful artificial fertilization on the eggs of a marine fish — the Lofoten cod. At that time, particularly in America, techniques for the artificial propagation of freshwater

fish were well advanced, with hatchery output contributing to the maintenance of freshwater fisheries. To Fisheries Commissioner Spencer F. BAIRD of the U.S.A. (1880), there seemed good reason to suppose that a substantial marine fish hatchery scheme would similarly benefit the depleted fisheries of the Atlantic seaboard. Hatcheries were accordingly erected at Gloucester, Mass., (1878), Woods Hole (1885), and later at Boothbay Harbor (1905). The impact of this early pioneering work was first felt at the London Fisheries Exhibition of 1883, where the American display had a profound effect on those European biologists interested in fish culture. Marine hatcheries were soon operating at Flødevig, Norway, under Capt. G. M. DANNEVIG; at Dunbar, Scotland; at Concarneau, France, and also, at the instigation of Prof. W. A. HERDMAN, on the Isle of Man (Port Erin) and at Piel, Lancashire.

The original American idea was to offset egg wastage on spawning grounds during fishing operations by conducting mass artificial fertilizations of cod and winter flounder from commercial vessels. This scheme was later modified to afford some protection to a small proportion of these fertilized eggs by incubating them in tanks ashore. The newly hatched larvae were then liberated into the sea.

In Norway, DANNEVIG (1910) came to rely on the uninhibited spawning of cod in hatchery ponds for his egg stock, and modified the Clark box, an American incubator, to impart continuous gentle movement to the egg during development. Other European hatcheries adopted DANNEVIG's methods. At the height of marine hatchery effort, hundreds of millions of eyed eggs and newly hatched larvae of the cod, plaice, haddock, winter flounder, and pollack were annually liberated into American and European coastal waters.

From time to time, reports appeared in America and Norway on the 'beneficial' effect of these practices to local fisheries, but pioneer marine fish culturists were never able to prove their point decisively when faced with the criticism that increases in stock may equally well be attributed to natural fluctuations. It is true to say that early expectations were never, in fact, justified by the results of hatchery effort. When the American hatcheries closed down after the second world war, methods were not substantially different from those employed in the early 1900s. The same story applies to the British sea fish hatcheries.

This decline can be fairly attributed to poorly developed techniques. It is not enough to protect a small fraction of local egg production from natural hazards and then to subject them to new hazards in the laboratory. What is needed is a system for rearing large numbers of fish through the tender egg and larval stages, to a point when the death rate has dropped to a steady low level, and the survivors are better equipped to withstand the rigours of life in the open sea. This principle was well understood by HERDMAN (1901), who wrote . . . "But it cannot be too emphatically stated and widely made known that sea fish hatcheries ought not to be merely for the purpose of hatching young fish and then setting them free in the sea. Hatching and *rearing* of fish is the end to have in view and scientific men who have charge of fish hatcheries will not be content till they have succeeded in rearing into young fish, at a reasonable cost, a large proportion of the fry which they can now hatch from the eggs by the million." The principle still stands — the function of a potentially effective fish hatchery should be to nurse the larval fish right through its delicate early stages before liberation into the sea.

Early Development of the Plaice

The plaice (*Pleuronectes platessa* L.) is sexually mature usually in its fourth year of life under natural conditions. At first spawning a female may produce some 50 thousand pelagic eggs, and egg production increases with age. The fertilized egg is buoyant, about 2 mm in diameter and takes 3 weeks to hatch at 6°C, the incubation time being closely related to temperature (APSTEIN, 1909). The emergent larva has symmetrical eyes and enough yolk to last a further eight to ten days at normal sea temperatures. Subsequent larval feeding stages remain pelagic until the onset of metamorphosis, some ten weeks after hatching. The duration of the pelagic phase depends on food abundance as well as on temperature. At metamorphosis, the left eye migrates to the right side, the body becomes flattened and the right side, now dorsal surface, develops heavy pigment in preparation for the demersal habit. Larval development can be classified as follows:—

Stage 1. Yolk still present.

Stage 2. Yolk resorbed but notochord still straight.

Stage 3. Eyes still symmetrically placed. Notochord bent.

Stage 4. Eye migrating but not yet at edge of head.

Stage 5. Eye on or over edge of head.

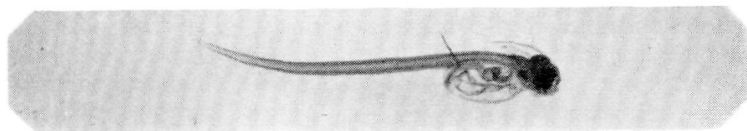
These five stages are shown in Figure 1.

PETERSEN (1894), the eminent Danish biologist, made several unsuccessful attempts to rear plaice from artificially fertilized eggs, and became convinced that . . . "At the pelagic stage, after the yolk has been absorbed, it cannot be kept alive in aquaria . . ." But already small numbers of herring (MEYER, 1880) and cod (ROGNERUD, 1887) had been reared in crude conditions beyond metamorphosis. DANNEVIG (1897) eventually scored a small-scale success with the plaice; similar successes were reported by FABRE-DOMERGUE and BIETRIX (1905) for the sole, and ANTHONY (1910) for the turbot. Since these early efforts, Norwegian workers in particular have contributed to our knowledge of marine fish rearing techniques (DANNEVIG and SIVERTSEN, 1933; ROLLEFSEN, 1939; DANNEVIG and DANNEVIG, 1950).

Large numbers of metamorphosed larvae would be required to raise significantly the later yield of marketable fish, even in a small fishery. The bay fisheries of the Irish Sea might well benefit by annual increments of the order of 1–2 million young plaice. On the other hand, at least 500–1000 million would be needed to double the brood strength of the North Sea plaice fishery in any one year. The technology of mass production awaits a basic rearing technique which can guarantee consistent target production on a small scale. The first steps in this direction were taken at Lowestoft in 1957.

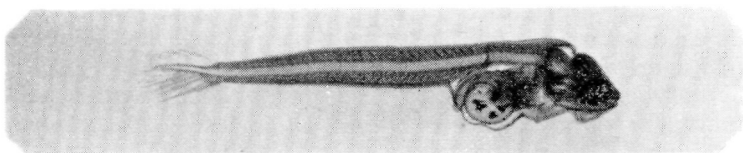
The Development of an Empirical Plaice Rearing Technique

For several seasons prior to 1957, plaice eggs caught at sea in plankton nets had been kept in aerated jars for basic morphological and physiological studies. It soon became evident that overcrowded conditions in small tanks were unsuitable for survival much beyond the hatching stage. By 1957 a closed circulation had been designed with some degree of physical and chemical



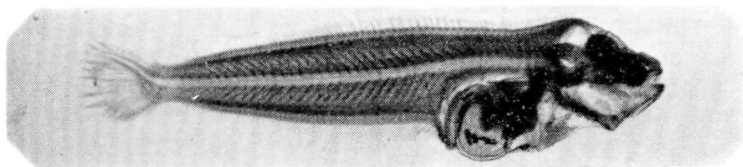
Stage 1

6.0 mm



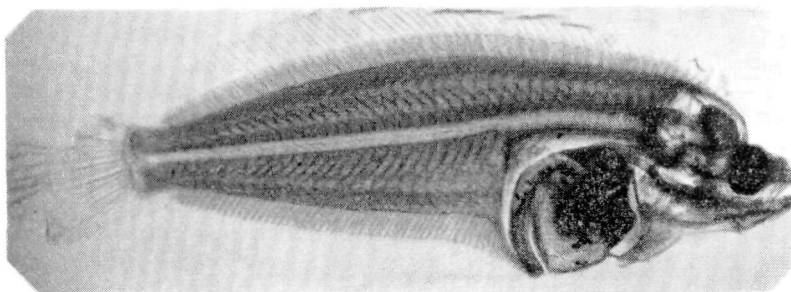
Stage 2

7.5 mm



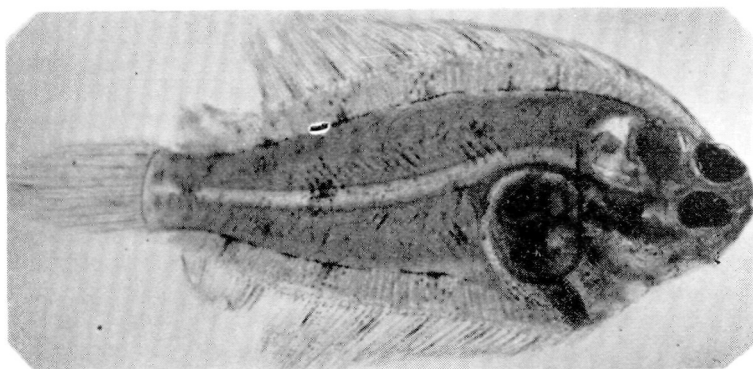
Stage 3

9.3 mm



Stage 4

12.3 mm



Stage 5

12.4 mm

Figure 1. Development stages of larval plaice.

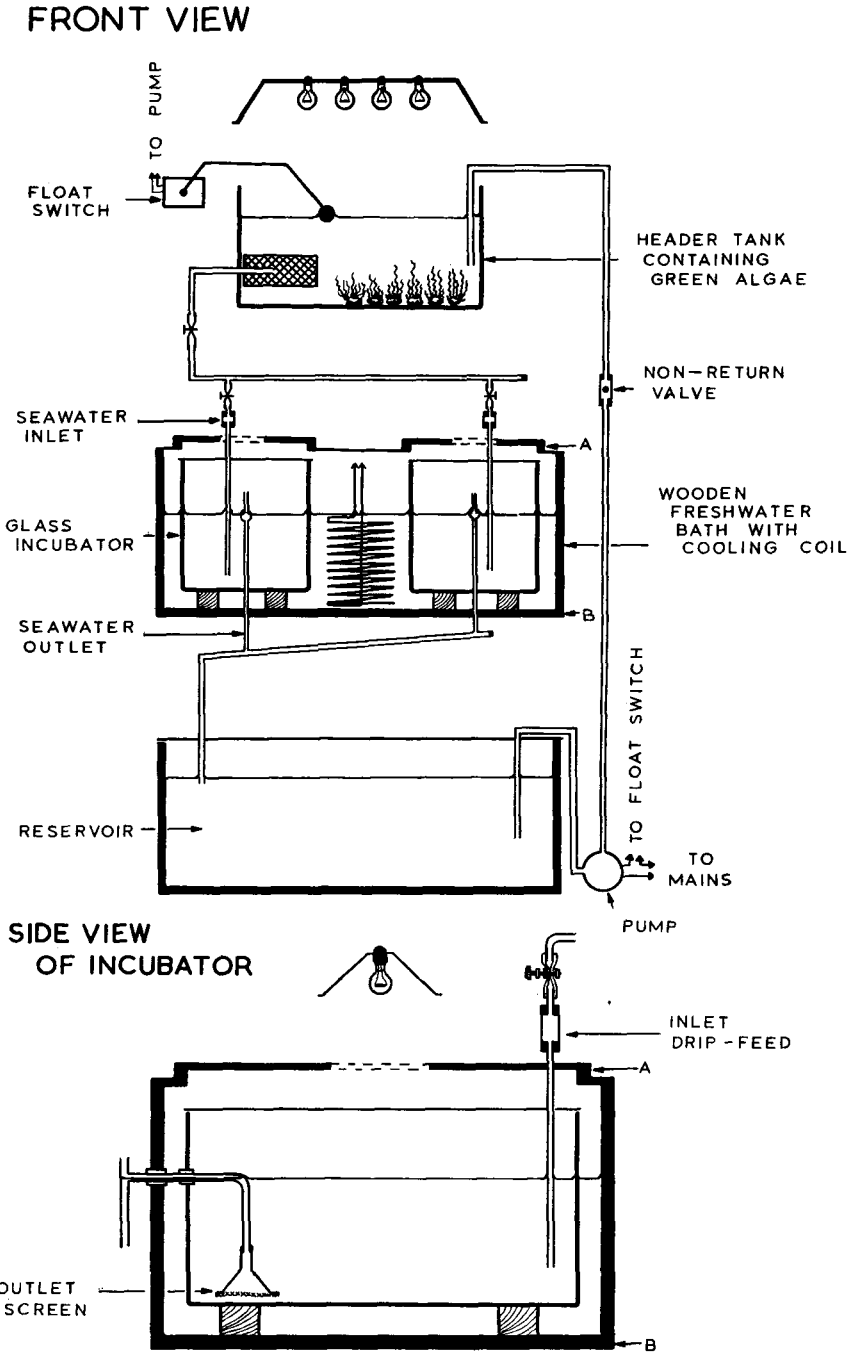


Figure 2. Closed sea water circulation for rearing plaice: Lowestoft, 1957.

control of sea water (Fig. 2). It incorporated a fundamental principle of tropical aquarium technique — the use of photosynthesizing plants to remove CO₂ and other metabolites from the water, at the same time stabilizing the pH and adding oxygen.

Two 60 cm × 30 cm × 30 cm moulded glass incubators with plastic covers were partially immersed in a wooden freshwater bath cooled by a coil linked to a modified domestic refrigerator. Sea water ran into each incubator at a slow controlled rate from a glass header tank, through a 6 mm bore polythene tube delivering below the surface. The outlet pipe was a right-angled length of glass tubing, the horizontal limb of which carried a rubber seal where it passed through a 1 cm hole drilled in the end wall of the incubator, at a point 22 cm above the bottom. The vertical limb, below water level, was tightly fitted into an inverted polythene filter funnel with its mouth screened by 23 mesh per cm bolting silk to prevent the flushing out of eggs and larvae. Sea water discharged into a covered reservoir under the water bath, from which it was intermittently transferred back to the header through an electric centrifugal pump fitted with a plastic volute and operated by a mercury float switch. The header contained washed fronds of the green alga *Enteromorpha intestinalis* L. growing on pebbles and collected from a local estuary. The alga received strong illumination from a battery of tungsten filament lamps which were independently switched; a rise in pH above 8.1 could be countered by decreasing the illumination. The total capacity of the circuit was about 200 l sea water. The glass incubators were dimly illuminated with artificial light through wide slits in the water bath covers, and a slow circulation of cooling water was maintained in the baths by strong aeration.

The shortcomings of this first closed sea water circuit were many. The irrigation rate was necessarily slow during larval feeding, to prevent undue loss of larval food through the bolting silk of the outlet screen, which, being of relatively small surface area, was always in danger of blockage. At a high ambient temperature and with a slow flow, temperature gradients developed in the incubators; these were a matter of concern, as the effect of sudden temperature change on larval survival was not thoroughly understood. *Enteromorpha* is not completely adapted to live continuously submerged in high salinity water and required renewal from time to time. A slow increase in salinity took place with evaporation, sometimes to a level well above that encountered in the sea. Pump failures were common.

Nevertheless, results were encouraging and demonstrated that a closed circulation could be used to rear plaice larvae from the egg stage, through and beyond metamorphosis.

A bigger system was built in 1959 (see Fig. 3). It had two main components; a sunken reservoir 4.6 m × 3 m × 1.2 m deep, of 15 cm reinforced concrete containing 11,000 l sea water, and an adjacent 6.7 m × 4 m × 2.4 m high brick-built fish hatchery. *Enteromorpha* was once again used for CO₂ and pH control, being submerged on trays consisting of polythene film on a wooden frame with no metal fastenings. The trays were held in position by nylon cords attached to strong points outside the reservoir; their position below the surface could be adjusted. Sixteen 80 watt fluorescent tubes, arranged in four banks of 4 above the trays, provided strong illumination for *Enteromorpha* at night. Each bank of lights was supported from a stout timber gallows spanning the tank, by a nylon rope and pulley system permitting vertical adjustment. The

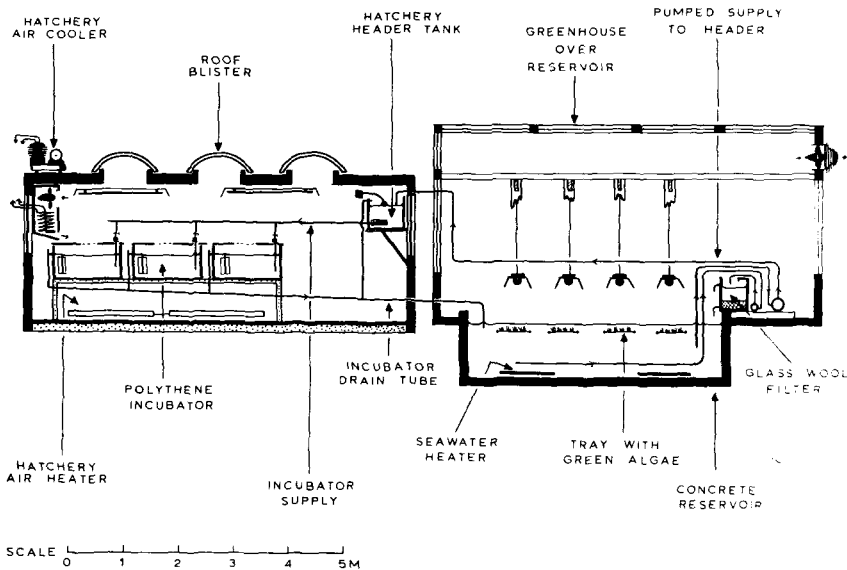


Figure 3. Closed sea water circulation for rearing plaice: Lowestoft, 1960.
 (Note: In 1959, the hatchery housed 16 glass incubators which were replaced in 1960 by 9 large polythene tanks.)

reservoir was covered by a cheap and lightly built greenhouse equipped with a controlled heating system for use in frosty weather. Low wattage immersion heating panels, completely embedded in epoxy resin and operated by floating thermostats, were used to offset excessive cooling of reservoir water during the winter months. There were no safeguards against rapidly rising temperatures during the spring and early summer. Shortly after the start of the 1959 season, a filtration system was installed. Reservoir water was pumped at a rate of 900 l per hour through glass wool pads in four polythene containers. Undesirable phytoplankton blooms were checked in this manner and a low degree of turbidity maintained for long periods. Replacement increments of fresh offshore sea water were added to the reservoir from time to time, and also distilled water in a vain attempt to offset evaporation and increasing salinity during hot weather.

A small electric pump under the control of a mercury float switch transferred sea water from the main reservoir to a header tank in the hatchery, to be distributed by gravity feed to sixteen glass incubators arranged in two rows of eight on metal stands. These incubators were of the same size and design as that shown in Figure 2 for 1957–58. Water ran back into the reservoir along a common drain tube. The inside walls of the hatchery were painted white; overhead illumination was provided by three glass ‘blisters’ in the roof, augmented by four 80 watt fluorescent tubes. Water baths were not used in the 1959 experiment. Side lighting was cut out by erecting an opaque cloth screen around the cluster of incubators. Thermostat heaters prevented excessive air cooling in cold weather; rising temperatures in warm weather could not easily be counteracted.

As far as possible, metal surfaces in contact with sea water were kept to a

minimum; only stainless steel (pump impeller) or chromium plate (reservoir thermostats) was used. Copper, brass and aluminium were strictly avoided. All sea water conduits were constructed from polythene, with glass and rubber inserts.

Although 60 cm × 30 cm × 30 cm glass tanks are useful for experimental studies, they are fragile and difficult to work. In an effort to increase larval production substantially during 1960, they were replaced by nine 152 cm × 61 cm × 61 cm black polythene tanks disposed in three rows of three on metal supports (Fig. 3). Each tank held about 450 l sea water when operational, boosting the circuit capacity from 11,000 to 15,750 l. The inverted filter funnel system for screening the outlet was replaced by the design reproduced in Figure 4. The vertical limb of the submerged outlet pipe was perforated along part of its length, the lower end corked, and the perforations screened with a 61 meshes per cm nylon bag held at a distance from the pipe with polythene discs. Tanks received dim illumination through two slits in a plastic-faced hardboard cover.

One of the main shortcomings of the 1959 set-up had been the lack of control over sudden increases in water temperature during sunny weather. Two air coolers were constructed from domestic refrigerator units, and installed in the hatchery for the 1960 season. They improved temperature control, but were not completely adequate for the task.

The greenhouse over the reservoir was treated externally with white paint to reflect sunlight, and summer water temperatures decreased accordingly. A further four glass wool filters were installed to give a total of eight and a filtration capacity of 1800 l/hr. Apart from these modifications, the reservoir arrangements were essentially the same in 1960 as in 1959. Perhaps the greatest

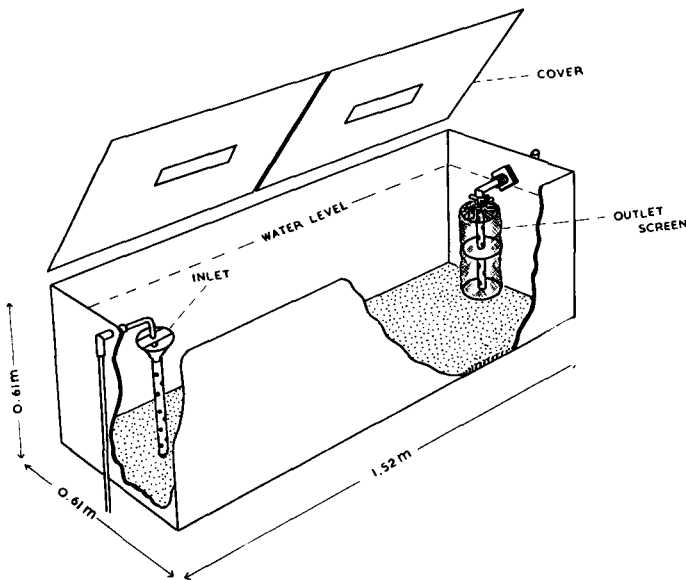


Figure 4. Details of polythene incubator used for rearing plaice in closed circulation at Lowestoft, 1960.

fault in the 1959–60 circulation was the use of a greenhouse to cover the reservoir. It was cheap, easily erected, and admitted daylight for algal photosynthesis, but the practical difficulties of temperature control outweighed these considerations. Reservoirs and hatcheries should be insulated against seasonal and diurnal air temperature changes.

The Main Points of Experimental Procedure

(1) Water and egg collection

The North Sea plaice spawning season usually starts south-west of the Dover Straits in late December, and extends progressively into the southern North Sea and Helgoland Bight between January and April. It became normal practice to collect offshore sea water during late January, allowing sufficient time for thorough filtration and temperature adjustment before egg collection started in early February.

A short sea survey with plankton net was usually enough to locate an abundant egg population. Collections were made with a 2 m stramin net towed just below the surface at a speed not exceeding 1.5 knots. Expected catches can be of the order 8 to 10 thousand plaice eggs per hour in various stages of development. Cod and dab eggs were also caught. Catches were quickly but gently emptied into a large volume of sea water in a glass tank, and the dead plankton and debris allowed to settle. Floating eggs were then skimmed from the surface with a polythene coffee strainer and transferred to asbestos-cement deck tanks (capacity 450 l) two-thirds filled with sea water, at a density of 10 thousand eggs per tank. As far as possible, egg collections were timed to coincide with the short and infrequent spells of calm weather. Back in harbour, eggs were once again skimmed and transferred to smaller glass containers for transport to laboratory incubators. By keeping an eye on weather reports, and by gentle handling and good timing, it was possible to complete collections with little egg mortality.

(2) Egg stocking rate

Arbitrary stocking rates of 300–500 sorted mid-stage eggs (gastrulae) per glass incubator were used in 1957–59, and 4–5 thousand mixed eggs — in all stages of development — per large polythene incubator in 1960. Since then, egg densities have been doubled without depressing survival rates. Gastrulae were chosen as experimental material following ROLLEFSEN (1932) who found the early embryos of cod to be more prone to mechanical damage than gastrulae.

(3) Egg condition during incubation

The shells of plaice eggs in the sea remain perfectly clear throughout the incubation phase. In contrast, tank eggs are characterized by increasing shell cloudiness during development, due to the growth of sessile bacteria. Opacity has varied from year to year in our experiments, which have so far lacked bacteriological control. Since introducing a good filtration technique, however, early bacterial troubles have decreased. The effect of bacteria on the percentage hatch of Pacific sardine, turbot and Norwegian cod eggs is already known (OPPENHEIMER, 1955); the ultimate effect on larval survival is not. In experimental aquaria, particularly with closed circulations, the bacterial hazard is

greatly magnified, and requires further study. Contaminated eggs tend to adhere to one another and gentle stirring was necessary from time to time, to break up clumps.

(4) Rates of water flow

The speed with which metabolites are removed from, and oxygen transported to, a tank will depend largely on the irrigation rate. Permissible flow in the circulations already described was much slower than optimum, for reasons of design. Up to and including 1959, 23 meshes per cm bolting silk was used to screen the outlet pipe. Although retaining eggs and larvae, this mesh was not fine enough to hold back small food organisms, and a fast flow quickly flushed away daily rations. Accumulating debris on the screen caused backing up of water in the tank, sometimes with ultimate overflowing. A finer mesh becomes blocked more quickly at the same high rate of flow, and any increase in screening area is not easily achieved without contributing to surface hazards in rearing tanks.

For these reasons, the rate of water flow was kept to a minimum: between 2 and 4 l/hr through a small glass incubator and 40 l/hr through the larger polythene tanks. A practical irrigation rate under the limitations of present tank design is about one tenth of the operational volume per hour.

(5) Tank hygiene

Dead eggs, larvae and food debris were, as a general rule, removed every other day by pipette. Strong illumination during cleaning operations produces heightened larval activity, often accompanied by a characteristic spasm in which the larval axis becomes momentarily contorted into an S-shape. Cleaning is now carried out by the light of an electric torch, and larvae can avoid the beam if disturbed by it.

A common early larval abnormality in unhygienic surroundings is the condition known as 'water belly', oedema of the abdomen. A recent correlation has been found between the incidence of 'water belly' and the presence of putrefying food in rearing tanks. Although scavenging is not the general rule, some pelagic plaice larvae do adopt this feeding habit. It is rewarding practice to remove decaying material as frequently as time permits, particularly in closed circulations.

Coastal nursery grounds are usually found on sand. Tank bottoms were therefore covered to a depth of 4 mm with washed sea sand during the late pelagic stage of larval development, in an effort to simulate natural conditions. This makes tank cleaning more difficult. Sand is not essential to structural metamorphosis, yet it may have some obscure influence on the course of later development.

(6) Temperature, pH and salinity

Normally developing plaice eggs have been found in water of 10°C in the Dover Straits at the start of the spawning season, and at 4°–5°C further north in March. They may be assumed to tolerate temperatures within this range. Temperature toleration is one thing; optimum temperature for egg and larval development, in a particular set of circumstances, is another. Whereas 10°C may be perfectly suitable for egg development in natural conditions, that same temperature may stimulate bacterial growth in the rich organic environment

of a closed circulation to the point where shell contamination becomes a problem, or where the speed of chemical change under bacterial influence outstrips the adaptability of an embryo or larva.

The temperature regime in offshore waters is much more stable than that of shore hatcheries, unless efficient control systems are used. The effect of sudden temperature variation is still not understood, and has high priority in future research programmes. Different temperature regimes were adopted during the period 1957–60 in our plaice rearing experiments; in retrospect, a practical schedule is (a) to incubate eggs at less than 7°C to restrict bacterial growth on shells, (b) rising to 8°C during early larval feeding, (c) followed by a gradual increase to between 10° and 12°C at metamorphosis. These proposals are empirical and apply to closed circulations only. They will undoubtedly be modified in the light of further knowledge.

The use of *Enteromorpha intestinalis* to control pH has already been described. It was chosen, in the first place, to avoid the changes in salt balance occurring with direct chemical control. During the plaice spawning season, the pH of the sea usually stands between 8.1 and 8.3. In static rearing tanks the index may quickly drop below 8.0 under the influence of metabolic CO₂ from eggs, larvae, and bacteria. Since *Enteromorpha* was first introduced into rearing trials, pH levels above 8.0 have been easily maintained, even with manual operation. It would be a comparatively simple matter to achieve automatic stabilization by linking a pH meter control unit to a variable light system.

North Sea plaice spawn in high salinity water (35‰). Their eggs and larvae are presumably adapted to develop at this high salt concentration, so from the outset, offshore water has been used in our closed circulations. Increases in salinity due to evaporation have been a source of concern; adjustments were made from time to time with distilled or tap water, causing slight fluctuations that are unlikely to occur in the stable environment of the sea. It remains to be seen whether or not salinity stabilization is desirable. If so, it could be achieved by coupling a sensitive conductivity cell to a solenoid valve controlling the input of distilled water into the system.

(7) Light

Tank illumination is necessary as plaice larvae are visual feeders. Their tolerance of light intensity is thought to be quite wide, but sudden changes, such as occur during tank inspections, are undesirable. Strong sunlight is thought to affect the survival of cod larvae (DANNEVIG and SIVERTSEN, 1933).

Between 1957 and 1960 rearing tanks were blacked out except for a centrally situated slit in the top cover through which light was admitted at a low intensity of the order 100 lux at the water surface, as recorded with a photometer corrected by filter to the wavelength response of the human eye. Recent work suggests that higher light intensities of 400–500 lux may assist 'first-feeding' larvae to capture food, and that decreased light later on may help survival by depressing activity in the restricted space of a rearing tank. Illumination was continuous in 1957–58; in 1959–60 tanks were illuminated only during the day.

The value of a black tank wall lies in the way it contrasts an illuminated nauplius, making food capture easier for early larval feeders. Translucent polythene tanks do not give this contrast and are unsuitable for plaice rearing experiments.

(8) Tank design

A pelagic plaice larva is adapted to an active life in the open sea, out of contact with surfaces until metamorphosis. Surface contact is an unavoidable tank hazard which may unduly strain the adaptive resources of a larva, and it is therefore important to keep a tank interior as simple as possible, with no unnecessary inclusions. Two closely apposed surfaces can act as a lethal trap for roaming larvae. Crevices are a particular menace — larvae swim into them and seem unable to back out.

The very delicate skin of an early feeder is an important barrier to the loss of water and the entry of chlorides under the osmotic gradient. Disturbance of skin secretions by surface contact, or other structural damage, may impair the efficiency of this barrier, and present a salt control problem beyond the capabilities of a larva. Tank outlet screens have been a continual source of trouble and mortality since this rearing project started. Our tank design has given some measure of success, but could bear considerable improvement.

(9) Aeration

Eggs were not directly aerated during incubation, nor subjected to the intermittent agitation recommended by DANNEVIG (1910). Direct aeration has the effect of driving floating eggs into regions of least water movement, and unless very gentle indeed, can cause heavy mortalities among delicate, newly hatched larvae. Oxygen release by *Enteromorpha* was sufficient to meet larval demands during the pelagic phase; after metamorphosis, direct aeration was of definite value in our closed systems, particularly in deep tanks.

Larval Food Production and the Feeding Technique

The efforts of early pioneers to rear marine fish were usually hampered by inadequate and unsuitable larval food supplies. ROLLEFSEN (1939) first demonstrated the value of *Artemia salina* L. (brine shrimp) nauplii for rearing sea fish. The brine shrimp is not indigenous to Britain, but its eggs can be imported in bulk from America, at a reasonably low cost. They hatch within 42 hours at temperatures between 20° and 23°C, in normal sea water, and can be readily cultured to the metanaupliar and adult stages in the laboratory, for feeding to larger fish. *Artemia*, therefore, forms a reliable staple diet for large-scale plaice rearing projects. There is one drawback — the nauplius is rather large for 'first-feeding' plaice larvae. The nauplii of *Balanus balanoides* L. are much smaller and it has become general practice to offer these during the first week or so of feeding, until larval survivors are large enough to live and grow on *Artemia* alone. Barnacle nauplii are easily obtained in spring by stripping adults from intertidal surfaces and covering them with sea water in glass tanks. The nauplii are immediately released and, being strongly phototactic, can be concentrated by unidirectional light. The use of *Balanus* as a first food is a purely empirical measure — its practical value has not yet been proved beyond doubt.

In 1957–59 the feeding technique was as follows:— About 5 days after the first egg hatched in each tank, a small ration of washed barnacle nauplii was offered. Daily increments were maintained for 3 days before adding the first *Artemia* nauplii, which were hatched in beakers in a water bath and separated

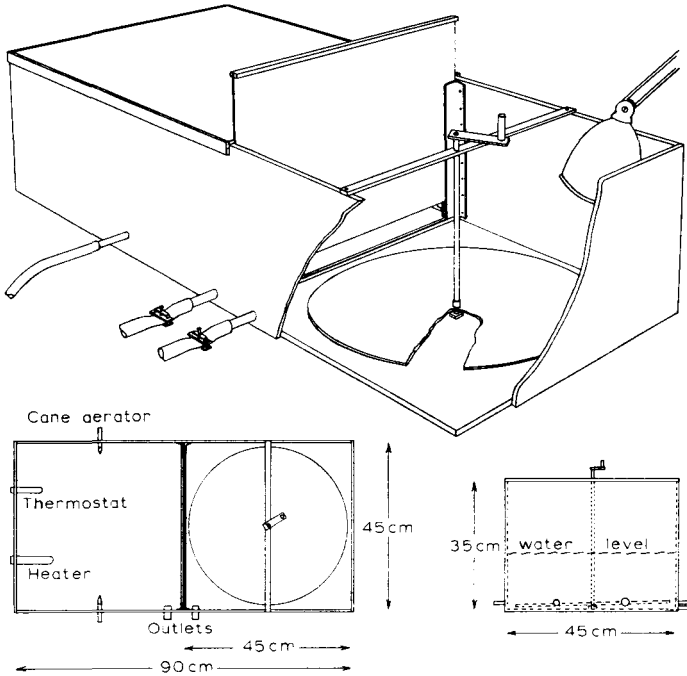


Figure 5. *Artemia salina* hatching box and separator as used at Lowestoft, 1959.

from egg shells and unhatched eggs by exploiting their phototactic behaviour. High *Artemia* concentrations were gradually built up and *Balanus* increments tapered off.

Rations of *Artemia* nauplii were subsequently offered every morning in quantities sufficient to maintain a residual food population after 24 hours of larval feeding. Following metamorphosis, the diet was varied with the oligochaetes *Enchytraeus albidus* and *E. bucholzii*, which are readily cultured on damp peat enriched with pre-cooked meal.

Dead and partly digested *Artemia* nauplii form a pink carpet of autolysed tissue on the bottoms of tanks, which should be regularly removed. Light enters rearing tanks through the cover slit, attracting nauplii to the centre and surface; plaice larvae feed on the fringes of this tightly bunched food population. The system has been reasonably effective, but further work is required to determine the ideal disposition of predator and prey.

Any planned increase in fish production must be accompanied by a corresponding increase in larval food production. An *Artemia* hatching box was designed in 1959, which could be operated at a low hatchery air temperature, using limited quantities of offshore sea water (Fig. 5). It consisted of a resin-bonded plywood box coated internally with dark epoxy resin, and divided into two halves by a black plastic partition which could slide up and down between flexed strips of 1 mm thick polythene, fixed to opposite walls. One half, the egg incubator, was fitted with two aerators, a thermostatically controlled aquarium heater, an outlet pipe, and a light-proof lid; the other half, the separator, with a large plastic helical wheel for stirring, and an outlet pipe.

In operation, the box was filled with sea water to a desired depth, the central partition pushed down tightly on to a fixed rubber strip athwart the box floor and a calculated quantity of *Artemia* eggs added to the incubator compartment. A concentration of 0.75 g eggs per litre of sea water in the incubator gave high percentage hatchings. The lid was then fixed in position and the air and heat switched on. After 44 hours at 23°C, the air and heat were switched off and the incubator contents allowed to settle. The separator was then illuminated with a 60 watt filament lamp, and the plastic partition cautiously raised, not more than 1 cm at a time. *Artemia* nauplii swam under the partition towards the light, and separation from hatching debris was usually complete within 90 minutes. The partition was firmly closed, and, for controlled feeding experiments, the stirrer operated and 1 ml samples withdrawn from the separator for density counts. Otherwise, the hatch was run off through 61 meshes per cm nylon fabric to concentrate the nauplii, washed in clean sea water, resuspended, and acclimatized to tank temperatures before being fed to the fish. Gentle aeration of concentrated *Artemia* is recommended. Two or three successive hatches can be obtained from the same sea water if in short supply, but best results demand water renewal for each hatch. When daily feeding is the routine two hatching boxes are required, since the production cycle takes two days.

Plaice larvae can be reared beyond metamorphosis on *Artemia* nauplii alone. Metanauplii and adults were used for further growth and fattening. Several 450 l asbestos-cement tanks filled with sea water were enriched with 0.5–1.0 kg dead mussels and placed in a sunny position outside. Temperatures were maintained between 12° and 15°C with controlled aquarium heaters. A thick flagellate population, mainly *Tetraselmis* and *Dunaliella* spp., appeared within three weeks, after which *Artemia* nauplii and unhatched eggs were added, and the temperature raised to 18°C. *Artemia* grew rapidly in these conditions; metanauplii were ready for cropping after two weeks. By this time the flagellate population had been grazed down, so mixed cultures of baker's yeast and *Phaeodactylum tricornutum* were added whenever the clarity of the water indicated food shortage. Adult *Artemia* may be too large to be swallowed by plaice larvae less than 25 mm long. They are, however, attacked, killed and eaten piecemeal.

The oligochaetes *Enchytraeus albidus* and *E. bucholzii* were also fed to metamorphosed fish. They are grown on a damp peat-loam mixture enriched with pre-cooked oatmeal (after BLOUNT, 1937). The worms should be washed and separated before feeding to fish. Whereas individual worms are readily eaten, worm clumps seem less attractive to plaice larvae.

Larval Production 1957–1960

Annual production and survival rates for the period are given in Figure 6. A single fish passed safely through metamorphosis in 1957, from an experimental stock of 1000 eggs. A modest advance was made the following year — 100 survivors from 1500 eggs. Hopes of maintaining a 6–7% survival and expanding production to 1000 young fish were not realized in the bigger 1959 circuit. Eggs were in good condition after transfer to the laboratory, but quickly became contaminated with sessile bacteria at the prevailing temperature of 9°C, and losses were high. Circuit water conditions were poor. New concrete needs thorough leaching to remove toxic aluminates; it is possible that harmful

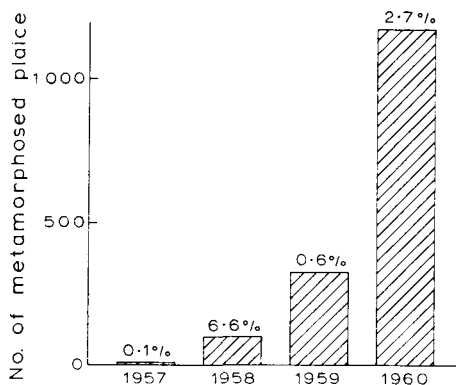


Figure 6. Annual production and percentage survival of metamorphosed plaice from sea-spawned eggs: Lowestoft, 1957–1960.

traces still remained when the system became operational. Filtration units were installed some time after the start of the experiment to counter increasing turbidity following heavy egg mortality, and to free the reservoir from a flourishing *Phaeocystis* outburst. A total of some 50,000 sea eggs were collected during the first half of February, in an attempt to build up a reasonably sized feeding stock in poor incubation conditions. About 1300 strong feeders were present in the circuit at mid-March, distributed among sixteen glass incubators in the hatchery. Just over 300 of these passed safely through metamorphosis during the following ninety days (Fig. 7).

No special effort was made in either 1959 or 1960 to plot the course of the mortality curve from the early egg stage up to first-feeding. This period will be fully dealt with in later publications. When reasonable, but not strictly controlled aquarium conditions prevail, slight though continuous egg loss may be expected during the 2–3 weeks of incubation, to be followed by a significant increase at hatching. In our experience so far, the greatest mortalities subsequently occur between hatching and first-feeding, corresponding to the 'critical period' of HJORT (1914). This is more probably related to the state of the emergent larva after protracted incubation in artificial conditions, than to the availability of food. A further phase of larval loss can also be expected between the onset and completion of metamorphosis. Thereafter the mortality curve generally flattens off; its slope may then be determined by the degree of overcrowding on the tank bottom.

The 1960 experiment was also interrupted by a series of early mishaps. Leaks developed in the large welded polythene incubators at low water temperatures. Repairs were effected with the experiment under way, necessitating frequent transfers of stock from one tank to another. At least 25% eggs and early larvae were lost during these manoeuvres. At the end of March there were some 2,400 strong feeders from a stock of 43,000 mixed eggs (Fig. 7). Just under 50% of all feeders passed through metamorphosis compared with 25% in 1959, giving a final survival of 2.8% (unadjusted for accidental egg losses) at a density of 137 fish per m². Early bacterial troubles were less and incubation temperatures lower than in 1959 (Figs. 8 and 9), although the salinity rose well above normal towards the end of the experiment. The main

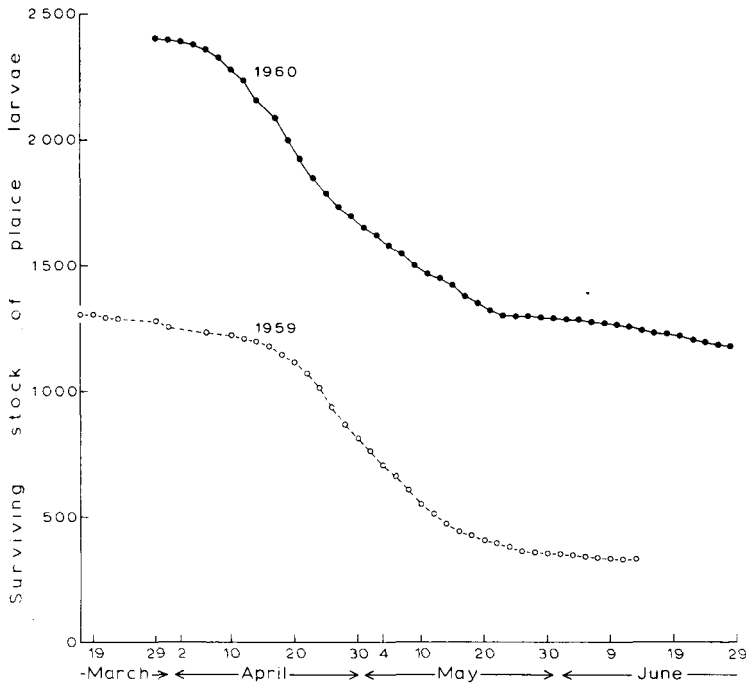


Figure 7. Survival curves of feeding plaice larvae in closed circulation at Lowestoft, 1959 and 1960.

mortality phases were similar in 1960 to those of the previous year — a high larval death rate between hatching and first feeding, followed by a second protracted period after the onset of metamorphosis. The effect of improved technique during 1960 is best shown by comparing the 2-daily death rate during feeding, with that for 1959 (Fig. 10).

The highest 1960 survival occurred in a 60 cm × 30 cm × 30 cm glass incubator, where 109 young plaice survived from 1000 sorted gastrulae, the best result ever achieved for any tank. Thus plaice larvae can be reared to a final density of at least 500 metamorphosed fish per m² of tank bottom.

Survivors were measured, either directly or from photographs, and put into 5 mm groups (Table 1). Although efforts were made during the experiment to maintain a reserve of food in tanks at all times, the final size range for each tank population was characteristically wide, even in the glass incubator which was stocked with eggs of roughly equal age. Less populated tanks also had greater proportions of larger larvae. Some pelagic forms grow faster and metamorphose sooner than others, probably gaining a considerable advantage in the subsequent struggle for survival on the relatively overcrowded bottom of an aquarium tank.

Normal metamorphosing plaice develop heavy pigment on the dorsal (right) side, in preparation for the demersal habit. Pigment abnormalities do occur in the sea, but appear to be much more frequent among tank-reared stock. In 1960, 62.6% of all survivors were normally pigmented. The remainder were pigment deficient in some respect, ranging from slight loss in the gill area to

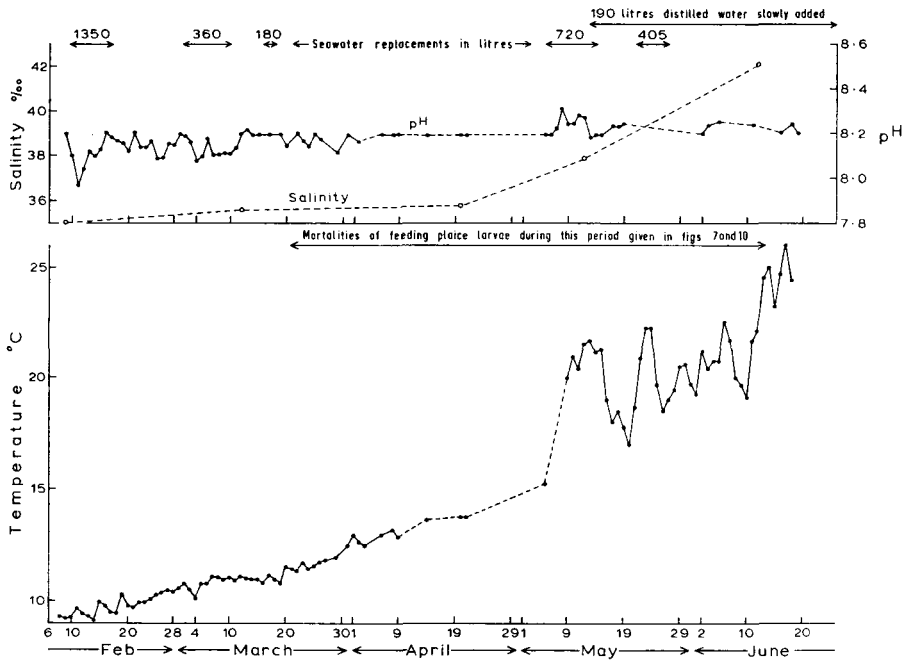


Figure 8. Reservoir temperature, pH, and salinity records: Lowestoft, 1959.

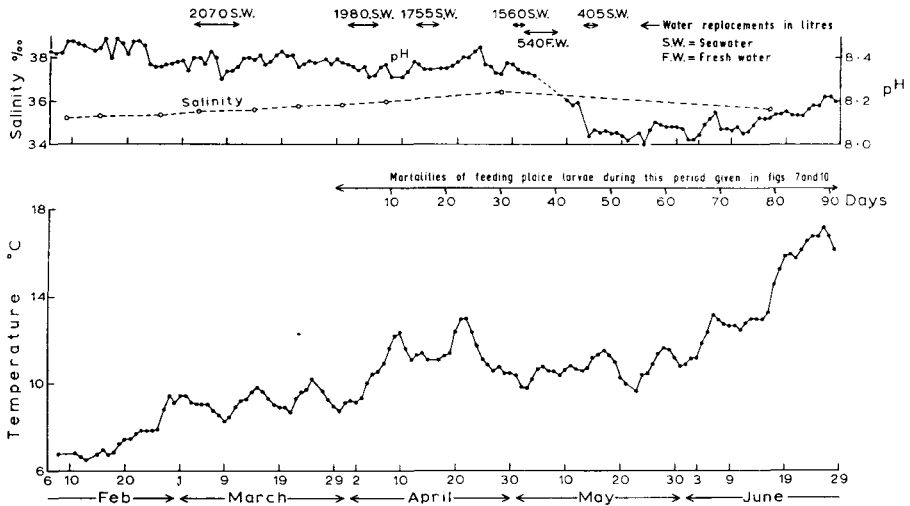


Figure 9. Reservoir temperature, pH, and salinity records: Lowestoft, 1960.

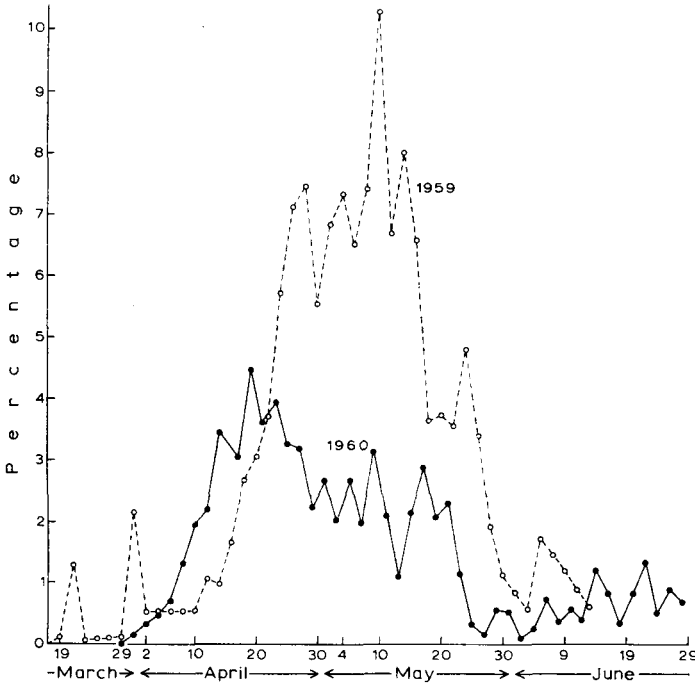


Figure 10. Mortality of feeding plaice larvae, as a percentage of remaining stock per 2-day interval: Lowestoft, 1959 and 1960.

Table 1

1960 Plaice Rearing Experiment at Lowestoft
Size composition of tank survivors

(Larval length measurements from tip of snout to posterior margin of caudal fin, allowance being made for bitten tails)

Length (mm)	Large polythene tanks									Glass tank
	1	2	3	4	5	6	7	8	9	
10.0-14.9	-	7	2	21	1	5	-	-	-	1
15.0-19.9	2	69	21	96	8	69	3	2	4	7
20.0-24.9	4	74	34	108	25	99	11	11	24	34
25.0-29.9	5	32	33	41	33	26	15	6	33	31
30.0-34.9	7	8	8	6	25	2	19	7	21	29
35.0-39.9	3	2	1	-	7	1	8	5	2	7
40.0-44.9	2	1	-	-	1	-	1	-	1	-
45.0-49.9	1	-	1	-	-	1	-	-	-	-
50.0-54.9	2	-	2	-	-	-	-	-	-	-
Metamorphosed survivors per tank	26	193	102	272	100	203	57	31	85	109

complete absence except for traces around the eyes and on the fins. The factors responsible for pigment peculiarities in plaice are not yet known and require special study, as the effective production of a hatchery can only be measured in terms of normally pigmented larvae with reasonable chances of further survival in the sea.

Biting also occurred during the demersal phase. Accurate estimates of its incidence could not be prepared for all tanks; a bitten tail or fin is not easy to detect from photographs. At the end of the experiment, all fish in the glass incubator were preserved in formalin. Of these fish, 25% were bitten.

It is unrewarding practice to attempt a quantitative assessment of factors affecting larval survival in tanks, before a basic rearing technique has been evolved. The work between 1957 and 1960 was mainly directed towards learning what technical mistakes to avoid during the course of a four-month plaice rearing enterprise. A closed circulation sets limitations on larval survival and production — there is an inevitable long-term deterioration in water conditions with the slow accumulation of metabolites from larvae, food, and bacteria. Nevertheless, survivals of 5% can be expected in our circuit, using modifications of the 1960 technique and keeping to a total stocking rate between 50 and 100 thousand eggs. We are now in a good position to study the effect of environmental variables peculiar to aquaria in a more systematic manner, with reasonable expectation of positive results. Such information will assist in technical refinement, and enable us to approach the problem of economic mass-production by the most direct route.

The main aim over the next few years will be to achieve maximum production of normal larvae for the least effort and lowest expenditure. At the end of this phase of the programme we should be well placed to attempt really critical field experiments, designed to measure the effect on subsequent yield of known liberations of metamorphosed plaice into selected coastal fisheries.

Summary

A project ultimately aimed at mass-producing metamorphosed fish from plaice eggs was started at Lowestoft in 1957. The first experiments were conducted on sea-spawned eggs in a 200 l closed circulation of sea water, using photosynthesizing plants (*Enteromorpha*) to control metabolites. This system was superseded in 1959 by an 11,000 l circulation comprising separate reservoir and hatchery, which was further expanded to 15,750 l in 1960.

An empirical plaice rearing technique has been steadily developed which takes into account some of the main physico-chemical and biotic factors peculiar to aquaria. Total annual production of metamorphosed fish since 1957 has been 1, 100, 300, and 1200 in successive years, representing a 3–5% survival from the egg stage. Occasional survivals as high as 10% at a density of 500 fish per m² have occurred in individual tanks. Mortality rates under our hatchery regime were generally highest (a) between hatching and first-feeding, (b) between the onset and completion of metamorphosis. The size range of survivors from any one tank after four months in the aquarium was characteristically wide, extending from 10 mm to a maximum of 50 mm. Only 62% of the final stock in 1960 were normally pigmented on the dorsal side, and in the one tank where an accurate estimate was possible, 25% had bitten fins.

References

- ANTHONY, R., 1910. "The cultivation of the turbot". Proceedings of the 4th International Fishery Congress, Washington, 1908, Pt. 2. Published as Bull. U.S. Bur. Fish., **28**: 861-70. (Translation.)
- APSTEIN, C., 1909. "Die Bestimmung des Alters pelagisch lebender Fischeier". Mitt. dtsh. SeefischVer., **25**: 364-73.
- BAIRD, Spencer F., 1880. "Report of the Commissioner for 1878". U.S. Comm. Fish., Pt. 6: xv-lxiv.
- BLOUNT, R. F., 1937. "Cultivation of *Enchytraeus albidus*". Contrib. in "Culture methods for invertebrate animals". 590 pp. Dover Publications Inc., New York.
- BORLEY, J. O., 1923. "The plaice fishery and the war. Preliminary report on investigations". Fish. Invest. Lond., Ser. 2, **5** (3): 1-56.
- DANNEVIG, A., & DANNEVIG, G., 1950. "Factors affecting the survival of fish larvae". J. Cons. int. Explor. Mer, **16**: 210-15.
- DANNEVIG, A., & SIVERTSEN, E., 1933. "On the influence of various physical factors on cod larvae; experiments at the Flødevig sea-fish hatchery". J. Cons. int. Explor. Mer, **8**: 90-99.
- DANNEVIG, G. M., 1910. "Apparatus and methods employed at the marine fish hatchery at Flødevig, Norway". Proceedings of the 4th International Fishery Congress, Washington, 1908, Pt. 2. Published as Bull. U.S. Bur. Fish., **28**: 801-09.
- DANNEVIG, H. C., 1897. "On the rearing of the larval and post-larval stages of the plaice and other flat-fishes". Rep. Fish. Bd Scot. 1896, Pt. 3: 175-93.
- FABRE-DOMERGUE, P., & BIETRIX, E., 1905. "Développement de la Sole (*Solea vulgaris*)". Travail du Laboratoire de Zoologie Maritime de Concarneau. 243 pp. Vuibert et Nony, Paris.
- HERDMAN, W. A., 1901. "Guide to the Port Erin aquarium". 15th Rep. Lpool. Mar. Biol. Comm.: 43-84.
- HJORT, J., 1914. "Fluctuations in the great fisheries of northern Europe viewed in the light of biological research". Rapp. Cons. Explor. Mer, **20**: 1-288.
- MARGETTS, A. R., & HOLT, S. J., 1948. "The effect of the 1939-1945 war on the English North Sea trawl fisheries". Rapp. Cons. Explor. Mer, **122**: 27-46.
- MEYER, H. A., 1880. "Biological observations made during the artificial rearing of herrings in the western Baltic". Rep. U.S. Comm. Fish. for 1878, Pt. 6: 629-38. (Translation.)
- OPPENHEIMER, Carl H., 1955. "The effect of marine bacteria on the development and hatching of pelagic fish eggs, and the control of such bacteria by anti-biotics". Copeia 1955, No. 1: 43-49.
- PETERSEN, C. G. J., 1894. "On the biology of our flat-fishes and on the decrease of our flat-fish fisheries". Rep. Danish biol. Sta. for 1893: 1-146.
- ROGNERUD, C., 1887. "Hatching cod in Norway". Bull. U.S. Fish Comm., **7**: 113-19. (Translation.)
- ROLLEFSEN, G., 1932. "The susceptibility of cod eggs to external influences". J. Cons. int. Explor. Mer, **7**: 367-73.
- ROLLEFSEN, G., 1939. "Artificial rearing of fry of sea water fish. Preliminary communication". Rapp. Cons. Explor. Mer, **109** (3): 133.
- SARS, G. O., 1866. "Om Vintertorskens, (*Gadus morrhua*), Forplantning og Udvikling". Forhandl. Vid. Selsk. Christiania, 1866. Translation in Rep. U.S. Comm. Fish. for 1873-74 and 1874-75, Appendix A: 213-22.