1987 Halibut Rearing at Seafish Ardtoe

Technical Report No.321
October 1987

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SEA FISH INDUSTRY AUTHORITY Marine Farming Unit - Ardtoe

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SUMMARY

The successful rearing of larval halibut is a very recent phenomenon with successes in both the UK and Norway. Halibut culture in time is expected to become an important commercial activity in North Europe, Faroe and Iceland.

During the 1987 spawning season, the halibut broodstock held at the Seafish Marine Farming Unit at Ardtoe produced almost 700,000 eggs from which over 20,000 larvae were hatched under very carefully controlled and monitored conditions. Two batches of larvae from a total of eight commenced feeding, and this is only the fourth time that this has been achieved and the first time it has been achieved outside Norway. Initially the larvae were fed on copepods and later at about day 39 they were fed on artemia nauplii. The use of an algal culture in the larval rearing tanks were most important to control hygiene.

The feeding of halibut larvae at Ardtoe marked a new departure in that it was associated with fairly high water temperatures (over 15° C). This offers some prospect of a larval rearing technique suitable for commercial-scale production. The largest of the larvae started to show metamorphosis about day 55.

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The production of 18 preweaned fish approaching metamorphosis marks a major advance towards the commercial cultivation of halibut. However, the poor survivals during the incubation and early larval holding stages are to be expected in this pioneering work but the circumstances are well documented. There is a need for considerable further development work to carry this work to a successful conclusion.

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1. Introduction

The successful rearing of larval halibut is a very recent phenomenon. Although captive halibut reached maturity at WFA Ardtoe in the 1970's, the eggs obtained were infertile and the stock of fish eventually succumbed to disease, probably as a result of being held in an unshaded floating cage. Within the past six years, in addition to the Ardtoe unit, several Norwegian research units have established halibut broodstocks and used these fish to produce viable eggs. The first successful larval rearing trials were conducted at Austevoll, near Bergen, using floating plastic enclosures set up in artificial lagoons. larvae were held under conditions close to those thought to exist in their normal rearing areas, i.e. total darkness, low temperature and a supply of natural zooplankton. The first two metamorphosed fish were produced in 1985, followed by a production of over two hundred fish in 1986. The Austevoll unit is continuing with this approach, as is a recently formed commercial production company, Lagoon Management and Construction (LMC). At the same time, another unit, at Sunndalsora near Trondheim, has been investigating the possibility of rearing under more controlled conditions, i.e. indoors, and produced its first two metamorphosed fish in 1986. One difference between these units was the technique for setting up and producing eggs from the spawning stocks: at Austevoll (and LMC) the spawning stock consisted of large fish, some over 100 kg, which were stripped on a daily basis or left to spawn naturally, at Sunndalsora the broodstock were much smaller, around 40 kg and stripping was timed to coincide with ovulation.

The Sea Fish Unit at Ardtoe started to set up a new halibut broodstock in 1981 but was unable to obtain any fish larger than 10 kg until 1985 (when a single fish of 25 kg was received. The unit has accumulated a stock of about 70- fish (TR 317) which are young and, in the short term, relatively unproductive in terms of potential weight of eggs. Against this, the fish are very well established and should have a long productive life ahead of them, as opposed to the larger fish at Austevoll which frequently do not survive three years in captivity. The stripping procedure used at Ardtoe was closer to the Austevoll pattern: normally once per day in the late morning.

The first halibut rearing trial at Ardtoe was in 1985, using two batches of larvae from Austevoll. None of these fish were seen to feed although their maximum survival time of over sixty days indicated the suitability of the system for holding larvae. In 1986, although two of the Ardtoe broodstock produced viable eggs, there were problems with contamination during incubation and none of the resulting larvae survived more than a few days. However, the experience enabled procedures and equipment to be defined for the next series of trials. In the interim, staff at Ardtoe, with the assistance of Miss M. Brancker, Fish Health Consultant to the H.I.D.B., made contact with the Norwegian workers (TR 292), and some modifications to the programme were made in the light of this exchange of information.

2. Procedures

Most of the equipment used in 1987 was identical to that specified in earlier reports, modifications being largely applied to procedures.

i) Egg stripping

In 1987, the Ardtoe procedure was modified to eliminate any contact between the eggs and the water of the broodstock tank: water for receiving eggs was prepared by filtration through 5u and 0.3 μ cartridges followed by U/V treatment and salinity adjustment to 35 or 36 ppt. This water was collected in clean plastic pails and cooled overnight to about 6 $^{\rm OC}$.

The eggs were stripped directly into the water, at least one container per female, and, if the eggs showed a good appearance (i.e. clear and floating), they were fertilised by adding 1 ml of milt diluted into 500 ml of prepared water; normally two males were used. As in previous years, the milt was examined within 10 minutes to assess motility Records were kept of the time of stripping, temperature and salinity of the broodstock tank water, temperature and salinity of the receiving water, the sperm motility and the identities of the female and male fish stripped.

ii) Egg treatment

The pails containing the fertilised eggs were immediately transferred back to a controlled temperature room at about 6°C and left for about 10 hrs., i.e. until the first cell division. A sample of about 150 eggs was then examined for clarity, shape and the appearance of the divided cells (e.g. whether the division was asymmetrical). A count was made of the number of developing eggs in the first 50 examined, this being recorded in

the form of a percentage development. A line of 12 to 15 eggs was measured using a microscope eyepiece graticule and a vernier stage scale to give an approximate mean diameter and another group of about 100 eggs was weighed and the mean weight recorded.

The floating eggs were then transferred to an incubator. At the beginning of the season this was done by netting using a plastic tea strainer and weighing both the floating and dropped out eggs in air. Later in the season, floating eggs were transferred in water to the incubator and only the dropped out eggs were weighted. Where possible, eggs transferred to the incubator were roughly assessed volumetrically and the numbers calculated from the egg diameter.

iii) Incubation

Although eight recirculation systems were prepared and these systems were designed for egg incubation, it soon became apparent that the number of egg batches produced would greatly exceed the accommodation available. Ten plastic bins of 22 lit. capacity were installed to provide accommodation for small batches of eggs. These bins were held on shelving in the largest of the controlled temperature rooms. Although both the bins and the recirculation units had lids, the rooms were kept in total darkness except for brief periods of visual checking each day with a rechargeable electric torch (white).

In addition to the incubation of eggs in the recirculation systems and the 22 lit. bins, eggs were incubated on two occasions in the standard turbot incubation units i.e. 80 lit. conical bottomed bins with a constant supply of recirculated ambient sea water at 15° C and having a central standpipe screened by a 500u mesh which was constantly swept by a rising current from

a perforated airline ring at its base.

Normally all incubation was carried out in filtered and U/V treated sea water adjusted to 36 ppt salinity but when eggs showed a loss of buoynacy this salinity was increased in an effort to hold them at the surface. No antibiotics were used unless conditions indicated a build up of bacteria. When antibiotics were used, they were normally added in the form of either: a) a 1 ml addition of Tribrissen suspension per day to the 22 lit. bins, or b) a single addition of 50 ppm of Oxytetracycline (in the form of a powder dissolved in 500 ml distilled water) to the recirculation units.

Water exchange during incubation varied with the type of container used and the appearance of the eggs. In the 22 lit. bins there was normally a 10% daily water exchange, unless the water gave off a slight smell when the exchange could be increased up to 75% daily, as thought necessary. Water removal from the 22 lit. bins was via 7 mm i.d. siphon tube and water was carefully transferred into the bins using a 1 lit. plastic beaker. In the recirculation systems, it was only possible to detect the water flow (via the recirculation pump) by the observation of eggs as they were moved by the current. It was necessary to limit this current to a minimum in order to avoid the trapping of eggs on The regulation of the recirculation flow was the filter screen. complex, involving several valve adjustments and the possible change of a filter cartridge. Late in the season, modifications to the recirulation systems gave an improvement in the water quality and subsequent hatching rates. One difficulty in the manipulation of recirculation flow rates was the impossibility of recording data, adjustments were done on an ad lib basis.

The 22 lit. bins were siphoned clean daily but the recirculation systems were restricted to a rapid daily siphoning of the diffuser bowl with occasional siphoning of the bottom of the tank when debris was seen. Daily records were kept of water salinity and temperature in the incubation facilities (additional records of percentage dissolved oxygen and pH were kept after 8th April). All dropped out eggs were weighed and recorded, as were all water replenishments and additions of abtibiotics.

Although such measurements were not routine, tests of dissolved ammonia, nitrite and sulphite were occasionally undertaken and dip slides for bacteria, fungi and yeasts were also used as thought necessary.

iv) Hatching

All egg batches incubated in the 22 lit. bins were counted immediately after hatching and transferred to recirculation systems.

Batches incubated in the recirculation systems were generally left in these units until the larvae were transferred to rearing facilities. Larvae were transferred by 'beakering' very carefully, making sure that both temperature and salinity were equilibrated (within 0.5°C or 1 ppt salinity respectively). When newly-hatched larvae were moved, transfer was carried out as soon as possible after hatching, in the case of batches showing an extended hatching phase it was necessary to set a 'cut-off' point to limit stress on the first-hatched larvae. Some attempt was made to relate all incubation and larval development to time and temperature in the form of 'degree-days' (D°), this value being derived, in the case of incubation, from the daily temperature record and the

number of days since hatching was judged to have been completed.

No records were made of the length or weight of larvae.

v) Larval Rearing

The original programme was for larval rearing trials within the recirculation systems, although it was accepted that some larvae would also be provided for a rearing trial by the DAFS unit at Loch Ewe. The larval rearing systems were maintained with a constant recirculation flow and salinity was increased when larvae showed a tendency to lose buoyancy. Daily cleaning of the diffuser bowl by siphon was maintained throughout the larval rearing phase.

The regime of illumination was not consistent: some batches of larvae were held in total darkness (except for the daily examination with the torch), but others had early phases of illumination (day 5 to day 10) from a red background light which was measured as 17 lux using an OM 200 Meter manufactured by Robin Electronics Ltd. From day 10, a white background light (about 100 lux) was used, both lights being continuous. One early batch received white light illumination via a 3 mm hole in the centre of the opaque lid. Larvae at the first feeding stage were held in open hatchery tanks with a surface illumination of white light at 850 lux.

The presentation of food to larvae was not consistent and followed the experience of the preceding batches, the unit having no detailed information on this aspect. Copepod cultures were set up early in the season, both from locally-caught Trigriopus sp, and from calanoid copepods originating in the tanks at GSP Hunterston or collected in local plankton hauls. Artemia nauplii were also available, as were the hatchery-

produced cultures of the rotifer <u>Brachionus plicatilis</u>. Late in the season, a sample of commercially produced planktonic aquarium food was received from Dryden Aquaculture of Edinburgh, and was presented to larvae in a recirculation system.

3. Results - Egg treatment

Only six out of the twenty eight incubation batches set up (many of which were the combined production of several females over several days) failed to produce any larvae. Of the sixteen batches set up in 22 lit. bins, only three batches failed to produce larvae and one of these failed within three days of setting up. Two of the incubation batches set up in the recirculation systems also suffered total mortality, as did the only batch set up in the turbot incubation system (see Table 1).

It was not normally possible to count the newly hatched larvae in the recirculation systems, such counts being routine when the 22 lit. bins were used. Therefore, the bins provided the best basis for comparison of incubation treatments and techniques:

i) Egg weighing

In earlier seasons it had been the normal practice to weigh turbot and halibut eggs in air before transferring them to the incubation facilities. This had the advantage of giving a reliable 'start point' for calculations of hatching efficiency, but it was thought that it might be the cause of excess stress, particularly to halibut eggs which are much larger. For this reason, many of the halibut egg batches were not weighed in air; some were estimated from calculations of their area on the water surface or their volume in a measuring cylinder. The estimates of egg numbers from these calculations often differed greatly

from the weights of dropped out eggs removed during incubation: the drop out varying from 40% to greatly in excess of the calculated number. The second best production in a 22 lit. bin followed installation of an 80g batch of eggs which had been weighed in air. The best survival of these facilities followed estimation using a measuring cylinder. Similarly, the two best hatches in recirculation systems followed estimates of egg numbers by volume. Unfortunately there were other factors involved which left the situation confused; it was generally concluded that it would be prudent to avoid weighing eggs in air in the 1988 season.

ii) Timing of stripping

The stripping procedure is covered in another report, but one aspect was thought to be significant in the incubation performance: on two occasions, stripping was carefully timed to coincide with the calculated time of ovulation. This was inconvenient in that both involved midnight operations, but the appearance, percentage rate of development and subsequent larval production more than justified the extra effort (the two egg batches were the only ones produced in 1987 which showed development of over 90% of the eggs and a total of over 14,000 hatched larvae was counted).

iii) Spawning stock performance

Only 12 of the female broodstock produced eggs in 1987 and four of these produced eggs which failed to hatch (see Table 2). Of the remainder, two fish were responsible for almost all of the larvae produced, it was fortunate that one of these spawned in the first half of the season and the other in the second half. It is probable that the remaining six running females produced

less that 2,000 larvae between them (this was difficult to interpret since so many of the egg batches were combined) (see Table 3).

One factor which made planning of incubator accommodation difficult was the expected spawning of a very large (35 kg) female held in a recently constructed tank. Facilities were held in readiness for eggs from this source but the animal failed to spawn. This resulted in the loss of some data since egg batches had been combined (to economise on space) which, had they been incubated separately, would have given more information on the potential of some of the spawning stock and would possibly have increased the number of larvae produced. More effective use of the incubation facilities could have been made, with minimal loss of larvae, if transfer to incubation had been limited to batches showing over 60% development (see Table 4).

There was an indication of a reduction in egg quality at the end of the season; the last six batches, produced on or after 12th April, showed poor development and failed to produce any hatched larvae.

Of the 21 kg of eggs stripped, 10.6 kg were transferred to incubation facilities but only 22,000 larvae were hatched, corresponding to about 400 g of eggs.

iv) Incubation Facilities

On two occasions, eggs were incubated in the turbot incubation units. These units were not modified for the trials and, although a few larvae hatched on each occasion, the hatching was extended over several days and none of the larvae survived the turbulence and abrasion in the units.

a) Effect of temperature

Incubation was carried out within the temperature range 3.8

 $^{-9.3}{}^{\circ}$ C. The best four batches in terms of numbers of hatched larvae (which produced 84% of the counted total) were incubated within the range $4.7{}^{\circ}$ C $^{-}$ 8.9 ${}^{\circ}$ C. The greatest diurnal temperature variation recorded in these batches was $1.1{}^{\circ}$ C. The duration of incubation varied between 90 D $^{\circ}$ and $109D^{\circ}$ in these batches. The mean temperatures during the incubation of the best two batches were, in the recirculation systems: $6.3{}^{\circ}$ C and $5.4{}^{\circ}$ C, and in the 22 lit. tanks: $8.2{}^{\circ}$ C and $5.7{}^{\circ}$ C.

b) Effect of salinity

The best four batches (as detailed above) were incubated in water within the salinity range 34.5 ppt to 37.5 ppt. The maximum duirunal variation within these batches was 2.5 ppt.

c) Effect of dissolved oxygen concentration and pH

Records of dissolved oxygen and pH only commenced on 8th April, and appeared to be a good indication of general water quality. On many occasions the rise and fall of pH readings followed the same trends in dissolved oxygen readings at a time when the instruments gave consistent readings from day to day in stable tanks. It is not thought that the instruments used were accurate enough for great credence to be placed on the actual records but they were valuable as indicators of trends. Following the recording of a pH reading of 6.96 in one of the recirculation systems, the filter medium was removed from the column and an aeration tube was installed. These improvements were subsequently carried out on all the recirculation units after which there were no pH readings below 7.5 and very few below 7.7 in these systems.

d) Effect of antibiotics

The Tribrissen antibiotic was received as a suspension and

was not used in the recirculation systems since it was feared it might not disperse adequately. All antibiotic treatment in those systems was restricted to Oxytetracycline, added after dissolving in distilled water. This treatment proved effective in reducing egg dropout but there was some suspicion that it might have had an adverse effect on the embryos, for none of the larvae hatched from eggs treated in this manner survived longer than 17 days. The 22 lit. incubation tanks were treated as needed by adding 1 ml of Tribrissen suspension daily. Some of this could be seen later as a deposit on the bottom of the tank. However, this was not thought to be a disadvantage since that zone contained the dropped out eggs. There was no indication that Tribrissen had any adverse effect on either eggs or larvae.

e) Effect of water exchange

In the recirculation systems, there was only occasional replacement of water lost through leakage but in the 22 lit. bins, deteriorating water quality (indicated by smell as well as low dissolved oxygen and pH readings) necessitated regular exchange of up to 75% of the tank volume. The water was removed by siphon, at which time the dropped out eggs were collected, and sterilised, high salinity, water was then added using a 1 lit. beaker. This procedure caused great turbulence in the tank but there was no indication that it was responsible for subsequent egg drop out. The use of recirculating water in the larger incubation systems presented some difficulties, since the eggs showed a tendency to lose buoyancy and become trapped on the filter screens. This happened in all the egg batches incubated in these facilities, even when the salinity was raised to over 37 ppt.

4. Results - Larval rearing

- a) The hatched larvae were concentrated into eight larval rearing batches, only two of which held fish up to the commencement of feeding. Although a variety of feeds were presented to larvae in the recirculation systems and the fish were observed in apparent feeding activity, only one observation of gut contents was made and it is thought that the systems were not suitable for larval feeding.
- b) The daily records indicate a loss of buoyancy in larvae of all batches between day 5 and day 10. It was not shown that increasing salinity would bring these larvae to the surface. Of the two batches which survived to commence feeding, one (Table 5) was in water which did not exceed a salinity of 35 ppt during the first ten days, the other 'successful' batch (Table 6) having larvae on the tank bottom on day 5, at 35.5 ppt, but these larvae had apparently risen to the surface on day 7, at a salinity of 36 ppt. Some of the 'unsuccessful' batches were held for lengthy periods at salinities of over 37 ppt without affecting their tendency to drop to the bottom. The situation is obviously complex and may be a reflection of general larval health rather than a specific defect in the holding facilities.
- c) There was no discernable ill effect related to the illumination of larval tanks by either red or white light. In fact, larvae newly transferred to rearing tanks (i.e. at 200 0°), showed a tendency to congregate in the brightest part of the tank.
- d) Larval survival from hatching to the commencement of feeding was generally very poor, even in the batches which

produced feeding larvae, the survival to transfer at $70 - 345 \, D^{O}$ was not thought to have exceeded 10% of the number originally in the tank (the first batch was uncounted).

- e) Of the six batches which failed to survive to first feeding, four hadbeen treated with Oxytetracycline during incubation. Both of the two batches which did produce feeding larvae had received either no antibiotic or Tribrissen during incubation. While the use of Oxytetracycline cannot be proven to be the cause of poor larval performance, its use has not been associated with success in these trials; at the same time, the use of Tribrissen has only been followed by a poor survival. The generally poor survival of all batches is an indication that there are severe problems involved in the health of newly-hatched halibut.
 - f) Larval Feeding Trial March 1987 (Table7)
- i) The tank was drained on 3rd April and the bodies of the last two fish were found on the bottom. The addition of Nannochloris algal culture (5 lit/day throughout) may have been responsible for the total absence of decay on the tank bottom, in spite of the lack of cleaning during the larval rearing phase.
- ii) Feeding activity was noted at about 300 D^0 and on many occasions thereafter. It is likely that feeding was terminated by a temperature drop on 19-20/3, there being no heater fitted to this tank.
- iii) Although no feed was noted as having been added before day 26, the tank had been stocked with copepods and given daily additions of algal culture for many weeks. The tank water remained green throughout the feeding trial, fish being impossible to see when they were more than 25cm below the surface.

- g) Larval Feeding Trial May 1987 (Table 8)
- i) The first observed feeding was on day 23 and day 28 (250 $\rm d^{0}$) fish were seen to feed immediately after transfer to the tank.
- ii) Plankton (collected by a 1/2 metre plankton net close to the unit) was introduced on day 23 and was later found to have been contaminated by medusae (largely <u>Sarsia</u> sp.) and between day 28 and day 41, 456 medusae were removed from the tank. There were no observations of larvae being attacked by medusae.
- iii) The water remained cloudly throughout the trial and fish tended to move down and out of sight after a few days in the tank. On day 39 and day 40, only 6 fish were seen but then the observed count increased daily up to d51 when 23 fish were seen. The few fish which remained at the surface for several days after transfer tended to have a sicky appearance; some showed signs of tail rot and latterly several fish were obviously suffering from oedema. None of these fish were seen to improve in health or feed and it is thought that all of them died soon after taking up a position at the surface.
- iv) There were no regular counts of the copepods since they were unevenly spread throughout the water. There were conspicuous concentrations of copepods in the brightest area of the tank (850 lux at the water surface) until day 38 when feeding activity suddenly reduced them to a very low density.
- v) Since it was not thought that sufficient copepods could be collected in the plankton net (even if medusae were not present) Artemia nauplii were presented on day 39. Fish were seen to have nauplii in their guts on the following day, but possibly more significantly, the guts also contained unhatched

cysts i.e. the fish were feeding on inert particles. This phase of feeding activity commenced when temperatures were above 16° C.

- vi) Feeding on Artemia nauplii continued and on day 55, the largest of the larvae were showing signs of metamorphosis. It was feared that the fish could then take up positions at or near the tank bottom and therefore the water was clarified by starting a water flow (about 10%/day) and stopping the addition of algal culture at day 56.
- vii) Daily measurements of pH and dissolved oxygen were taken throughout the rearing phase; the pH meter failed on day 24 and day 68.
- viii) As with the March trial, the tank bottom was found to be clean and free from odour when the tank was finally emptied.

5. Conclusions based on 1987 Halibut Hatchery experience

- 1. The treatment of egg-receiving water by filtration and U/V was worthwhile and will be adopted as standard practice.
- 2. It is not thought that any ill effects resulted from the temperature variations or light intensity variations involved in the early phase of egg development.
- 3. Some loss of egg buoyancy was noted when eggs were collected in U/V-filtered water from the ambient supply. In future, all eggs will be collected in U/V-filtered sea water adjusted to a salinity of 36 ppt.
- 4. The few egg batches stripped at the calculated point of ovulation were of conspicuously better quality than those obtained in the routine daily strippings. It would be worthwhile to organise all strippings of proven female stock on the basis of optimal timing.
- 5. Although it was not possible to definitely ascribe poor incubation performance to any particular fault in the egg handling technique, the best results, late in the season, followed egg collection in which the eggs were not removed from the water i.e. egg numbers were estimated from volumes of eggs and measured egg diameters. This technique is inherently inaccurate and gives a poor indication of subsequent incubation success but it may be necessary in order to limit stress. Trials to assess the resistance of halibut eggs to handling stress could be undertaken by an outside agency as and when a surplus is available.
- 6. The number of egg batches stripped during March was more than the unit could comfortably handle. Increases in accommodation and staff will be organised before the next halibut production season and, at the same time, accommodation will be

used more economically by not incubating batches showing a poor rate of development.

- 7. Although the only attempts to incubate halibut eggs in ambient sea water were inconclusive, this was thought to be related to the design of the system and the poor quality of the eggs used, rather than any inherent problem in the use of sea water. A new design of ambient sea water incubator will be tested next season.
- 8. The regular measurement of pH and dissolved oxygen to assess water quality in egg incubators was thought to be worthwhile and will be incorporated as routine for all egg holding, incubation and larval rearing facilities in the 1988 season. The instruments must be calibrated at least once per week, more often if abnormal readings are encountered.
- 9. The practice of exchanging incubator bin water by beakering seemed to be acceptable in that one batch showed a high survival when this technique was used. Practical difficulties involved were associated with the need to continually make up large quantities of high salinity water and maintain it at the incubator temperature. An improved system will be in operation in 1988.
- 10. The use of Oxytetracycline during incubation has been associated with the production of poor quality larvae. This may be no more than a coincidence but, as a precaution, egg treatment in 1988 will be restricted to Tribrissen, Flumequil or Oxolynic Acid.
- 11. The controlled temperature rooms functioned well for most of the season, loss of refrigerant in the new units occurred on two occasions before and after the main production phase and

was traced to leaks through cracked copper connections. The systems will be thoroughly checked before the 1988 season.

- 12. The recirculation systems functioned fairly well but needed modification halfway through the 1987 season to improve water quality. A tendency of eggs and larvae to lose buoyancy meant that many of each were lost after becoming trapped on the surface of the 'banjo' filters, design modifications are planned to reduce this problem.
- 13. The original design of the recirculation systems involved a biofilter phase, but this was abandonded when it was realised that the low temperatures involved would limit the effectiveness of the nitrifying bacteria. During the 1987 trials, temperatures were increased to a point at which biofiltration could become practical. Conditioning such biofilters presents difficulties in the early, low temperature, stages and it may be necessary to prime the system with a connercially available bacterial conditioner.
- 14. Loss of buoyancy in both eggs and larvae may prove to be one of the more difficult problems to solve; it should be possible to keep eggs on the surface by increasing the salinity, but this high salinity would have to be reduced before the larvae could be moved, with a risk of subsequent larval dropout, both eggs and larvae could be held in suspension by an upwelling current, but this implies an increased flow which would increase screening problems.
- 15. The recirculation units would probably function better as incubators if they were modified to use a flow of U/V-filtered ambient sea water during that phase. This would involve a redesign of the diffuser/cup arrangement to recirculate eggs. One disadvantage of such a system could be a loss of control over

temperature and salinity. The Norwegian units claim better incubation performance than we have been able to achieve so far. This may be related to the quality of the eggs, but it is possible that their incubation systems have inherent advantages over our designs, this must be investigated.

- 16. The performance of the incubation and early rearing facilities indicates that the light regimes adopted were not injurious to either eggs or larvae. The same regime will be used in 1988, i.e. white light torches used sparingly during the incubation phase, continuous red light five days after hatching and continuous white light for larvae after day 10.
- 17. The presentation of food to larvae should start at about 220 ${\tt D}^{\tt O}$ and should be in the form of a culture of copepods containing all stages of copepod larvae.
- 18. Cultures of copepods in larval rearing tanks should be set up several weeks before they are needed to a) ensure they are well established in the tanks and b) give the opportunity to remove any medusae which might have been introduced with plankton.
- 19. The use of Nannochloris algal culture in the larval rearing tank was thought to have been very important in that its supposed antibacterial activity was thought to have enhanced tank hygiene. Certainly, the tank bottom was found to be free of ciliates after 40 days of rearing without significant water exchange.
- 20. The rearing of halibut larvae at a temperature of 17°C resulted from the use of a tank in the main (uncooled) hatchery building. The fact that larvae survived showed that the temperature regime was not obviously harmful. It is possible that halibut larvae would give good results at even higher

temperatures and at least one trial in 1988 should involve larval rearing at 19°C .

- 21. There was no sign that halibut larvae would commence feeding on rotifers. In 1988, halibut feeding trials will concentrate on the use of copepods and Artemia. It is possible that halibut will accept Artemia as a first feed if the temperature is appropriate. A trial will be conducted in 1988 to examine this possibility.
- 22. The presentation of Artemia at day 39 was possibly significant in that the larvae were seen on the following day to have eaten large numbers of Artemia cysts. It is likely that, if they were prepared to take one form of inert particle at this age, they would have taken any other particle of the same size. There is a clear possibility of the use of a micro-encapsulated diet for the early feeding of halibut.
- 23. The tolerance of elevated temperatures by halibut larvae during these trials was unexpected. The use of high temperature rearing could have a benefit in accelerating the rate of larval development and enabling the hatchery to use its limited facilities more efficiently. It is not known whether the larvae are tolerant of such high temperatures at an early stage, or if the use of temperatures of 15°C in the recirculation systems would give rise to water quality problems, but these items must be investigated.
- 24. The best incubation performance in terms of percentage hatch was achieved within the temperature range 7.6 8.9°C. At this temperature, the eggs hatched in 11 days without the use of antibiotics. If this regime can be used routinely, it would greatly increase the capacity of the incubation facilities.

6. Recommendations

- The egg production in 1987 was not, as anticipated, in the form of a few large batches but came as a continuous run of small batches. The hatchery unit was not set up for this and it presented some problems, particularly at times when it was thought that the very big female would spawn, i.e. incubator accommodation had to be reserved for possible large batches. Next year there will, we hope, be a greater chance of large batches but the majority of the eggs are likely to be in small batches, as they were this year. The implication of this is that we will need much more incubation space in 1988. Two strategies 1) the turbot incubation units will be modified are planned: for halibut eggs, turbot production will be suspended during the many more 22 lit. bins will be purchased halibut season, 2) and fitted into the existing controlled temperature rooms.
- b) Even when eggs were held at fairly low density in the recirculation units, there were still problems with the quality of the water. The recirculation units will be modified to use filtered ambient sea water during the incubation phase, recirculation will only be used after the larvae have hatched. The use of Oxytetracycline will be stopped and it will be replaced by Oxolynic acid and/or Flumequil.
- c) Incubation has been shown to be successful at temperatures over 7°C and 1988 eggs will be routinely incubated at higher temperatures than were used in 1987.
- d) There have been shown to be four phases in the rearing of halibut: incubation; early larval holding; larval feeding up to metamorphosis; and weaning. Most of the emphasis in 1987 was placed on the first and third phases, but many of the potential

- h) Only two of the broodstock females showed good egg production in 1987, any health problems affecting these fish could put the 1988 production at risk. All stress on the broodstock must be kept to a minimum and efforts to increase and improve the broodstock should be continued.
- i) The size of halibut eggs and larvae may indicate the need for greatly increased volumes of incubation and larval rearing accommodation when compared to turbot production. It is possible that a production hatchery for halibut could need two or three times the space presently needed for the equivalent turbot production. If there is a significant improvement in the number of good eggs produced, the present hatchery area may be inadequate to handle all the larvae in 1988. Contacts with outside agencies should be maintained with a view to making surplus eggs and larvae available to them, it being understood that such availability may be at very short notice.

Acknowledgments

This project has enjoyed the enthusiastic support of all of the staff at Seafish Ardtoe, notably Peter Smith and Jon Sherwood who were involved in broodstock, Kay MacIver, who maintained the algal and rotifer cultures, Sandy Edmond, Jimmy Johnston and Jimmy Wood who undertook the many structural and mechanical modifications involved, and Judy MacGillivray who typed the report.

feeders were lost in the immediate post-hatching period. The reasons for this mortality are not clear and must be investigated.

It is possible that larvae have been lost as a result of poor water quality caused by attempts to feed larvae in the recirculation systems. None of these attempts have shown any sign of success and they will not be repeated. At the same time, the tendency of apparently healthy larvae to lose buoyancy presents particular dangers in the present facilities; either the facilities will have to be greatly modified or new facilities will have to be designed for the accommodation of young larvae.

- e) Larval rearing in 1988 will closely follow the successful trial in 1987. This implies the 'greening up' of several hatchery tanks with algal culture and added zooplankton some weeks before they are likely to be used.
- f) It seems likely that the best first-feed for larval halibut will be wild caught Copepods. These may not be available in February so it will be necessary to maintain one hatchery tank for Copepod culture throughout the winter.
- g) Although we cannot predict a very large scale of success next year, we must be prepared to take advantage of any halibut we do manage to rear. This implies the need to have weaning facilities available from early May. At the present time it is not thought that halibut will prove cannibalistic but it is not known how well widely different ages of fish will coexist in the same tank. If it is available, the 'natural spawning' tank (a circular indoor tank, 3.7m in diameter and 2m deep) could be the best facility to hold the weaned fish. Alternatively, (or additionally) it might be necessary to construct a sea cage facility for halibut ongrowing.

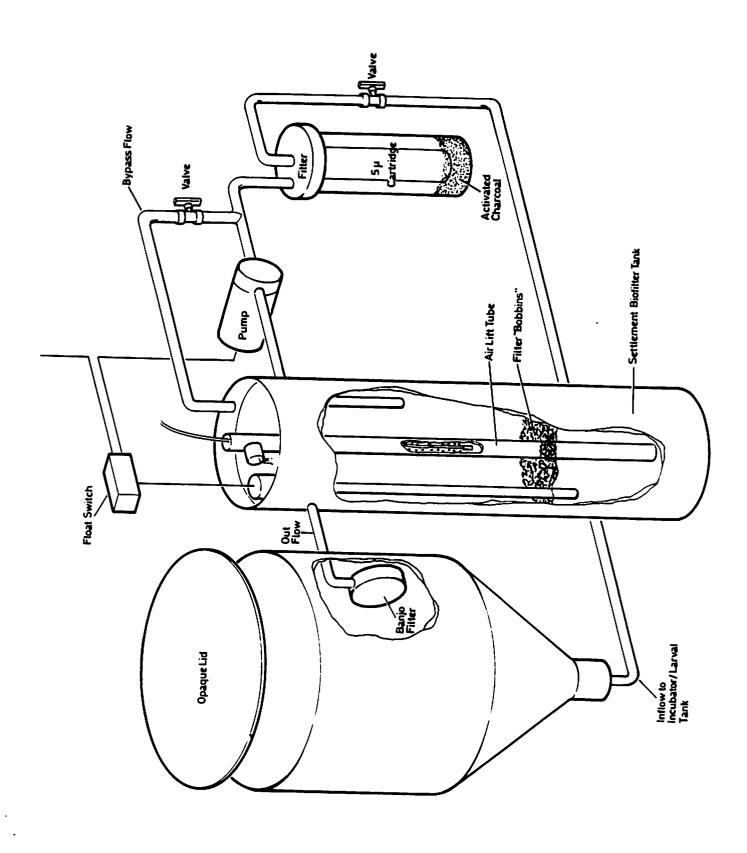


Table 1 Halibut stripping batches

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3/35	31.3	390	190	n.c.	875	A2
3/36	31.3	580	405	n.c.	875	A2
4/1	1.4	280	160	n.c.	875	A2
4/2	6.4	462	257	374	22	v
4/3	6.4	162	150	259	22	VI
4/4	6.4	340	277	323	22	VII
4/5	6.4	50	0	0		
4/6	7.4	575	530	747	22	VIII
4/7	7.4	130	115	2391	22	IV
4/8	9.4	90	10	n.c.	22	II
4/9	9.4	408	326	25	22	II
4/10	9.4	257	147	150	22	I
4/11	9.4	2179	2105	7600	650/540	D7/8
4/12	11.4	295	0	0		
4/13	12.4	912	887	7660	480	C6
4/14	12.4	94	0	0		
4/15	14.4	258	0	0		
4/16	15.4	263	0	0		
4/17	22.4	1211	560	0	80	T16
4/18	24.4	639	396	0	22	IX
4/19	27.4	841	256	0	22	V

Total	batches	stripped	70 (1 batch divided)
Ħ	11	incubated	47
11	11	failing to hatch	11

i.e. A maximum of 36 batches could have produced larvae. Unfortunately, many batches were amalgamated and therefore data was lost. Most batches incubated in recirculation systems were not counted immediately after hatching.

Total weight of eggs stripped 21,081 gm " " incubated 10,631 gm

Assuming the eggs were approximately 60 per gm., the number incubated was about 640,000.

Counted larvae produced

22,154

Table 2 Spawner Performance

Female	Number of strippings	Duration of production(d)	Batches hatched	Batches ?	Batches D/O
4/5	13	28	8	3	2
4/2	6	23	0	2	4
OR/1	6	26	1	4	1
OR/4	2	25	0	0	2
4/8	7	22	0	5	2
OR/3	4	21	0	0	4
OR/2	9	29	2	3	4
4/1	12	28	5	6	1
4/3	5	31	0	0	5
4/6	3	25	0	2	1
4/4	7	21	1	2	4
4/7	3	7	0	0	3
Total	77		17	28	32

These results are given in the order in which the females spawned.

In the context of these results a 'stripping' was defined as an occasion when apparently healthy eggs were produced. There were many occasions when the appearance of the eggs was poor, such eggs were discarded immediately.

The first two females in the table producing all the eggs in the first half of the spawning season.

Batches marked '?' could be judged as having hatched or dropped out since they were combined with other batches during incubation and some larvae hatched from the combined batches.

Although twelve females produced eggs, four of these produced no larvae and three of the remainder were not proven to have produced hatched larvae.

The largest female in the broodstock failed to spawn.

Table 3 Hatched larvae related to spawners

Spawning Female	Larval count
4/1	18,157
4/2	?
4/3	0
4/4	348
4/5	2,625+
4/6	?
4/7	0
4/8	?
OR/1	?
OR/2	524+
OR/3	0
OR/4	0

21,654+

All the counts marked '?' totalled less than 2,000 during the season.

Table 4 Hatching rate related to percentage development

Batch	Dev.(%)	Hatch
1/1	23	poor
1/1	50	107 from 80g
2/1	83	225 from 90g
2/2	72	31 from 203g
2/3	87	480 from 84g
2/4	87)	
2/5	49)	uncounted poor
2/6	83	1544 from 80g
2/8	89	238 from 75g
2/9	88	ni1
2/12	86	poor
3/1	80)	
3/2	84	nil
3/5	74)	
3/6	66)	
3/11	87)	
3/13	58)	
3/14	78	poor
3/16	64)	
3/18	68)	
3/19	76)	
3/21	80	nil
3/23	68)	
3/24	42)	
3/25	68)	
3/30	80	poor
3/35	68)	
3/36	62)	
4/1	78	poor
4/2	86	374 from 257g
4/3	80	259 from 150g
4/4	65	323 from 277g
4/6	74	747 from 530g
4/7	90	2391 from 115g
4/8	80)	
4/9	80	25 from 336g
4/10	61	150 from 147g
4/11	92	7600 from 2105g

4/13	90	7160 from 887g
4/17	19	nil
4/18	22	nil
4/19	26	ni1

The batches showing under 70% development used over 30% of the incubation facilities but produced under 3% of the counted larvae.

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Table 5 Early Larval Holding - February 1987

Date	D	D°	T°C	Sal.	Transfer
14/2	4	28.1	7.1	35	
15/2	5	35.5	7.4	35	
16/2	6	42.5	7.0	35	
17/2	7	49.5	7.0	35	
18/2	8	56.6	7.1	35	+ 200
19/2	9	63.5	6.9	34	. 200
20/2	10	70.4	6.9	34.5	
21/2	11	77.3	6.9	34.5	
22/2	12	84.4	7.1	34.5	
23/2	13	91.4	7.0	*	
24/2	14	98.6	7.2	34	
25/2	15	106. 0	7.4	34.5	
26/2	16	113.3	7.3	35	
27/2	17	120.4	7.1	35.5	- 100m
28/2	18	127.9	7.5	35	
1/3	19	137.6	9.7	34.5	
2/3	20	147.5	9.9	34.5	
3/3	21	157.2	9.7	34.5	
4/3	22	167.0	9.8	35	
5/3	23	176.9	9.9	35.5	
6/3	24	186.5	9.6	35	
7/3	25	195.9	9.4	*	
8/3	26	205.3	9.4	36	(-1000m
9/3	27	215.3	10.0	35	(- 60
10/3	28	226.0	9.7	35	- 46
11/3	29	236.0	10.0	*	
12/3	30	246.1	10.1	35	
13/3	31	256.3	10.2	35	- 60m
14/3	32	266.5	10.2	35	
15/3	33	276.9	10.4	34	- 2
16/3	34	287.1	10.2	35	- 2
17/3	35	297.5	10.4	35	- 2
18/3	36	307.3	9.8	35	- 2
19/3	37	317.5	10.2	34.5	- 2
20/3	38	326.9	9.4	34.5	
21/3	39	336.0	9.1	*	- 2
22/3	40	345.4	9.4	35.5	
23/3	41	354.9	9.5	35	

None were seen after d40 and the batch was abandoned on d41. Measurement of D° was approximate since there were no records for the first three days. It is not thought that the estimate is more than 2 D° in error.

On d13 it was noted that most of the larvae were swimming 'head down', normally a sign of poor health, but on d19 their appearance had improved considerably. The red light was turned on on d20 and the first food was presented on d21. White lighting commenced on d22 and the presented food (copepods were maintained by adding Nannochloris culture)thereafter. Rotifers (prefed on 'Seclo') were added on d26. Fish were transferred to a hatchery tank from d27.

Only one fish was seen to have gut contents, on d39, and this was transferred to the hatchery tank.

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Date	D	D°	T°C	Sal.	D/0 ₂	pН	Transfer
23/4	1	6.2	6.2	35	81	8.06	+ 374
24/4	2	13.0	6.8	36	100	7.91	+ 259
25/4	3	21.3	8.3	36	99	7.95	+1259
26/4	4	30.0	8.7	35.5	92	7.98	+2352
27/4	5	37.2	7.2	35.5	88	7.95	
28/4	6	43.9	6.7	36.5	92	7.94	
29/4	7	50.2	6.3	36	93	7.83	
30/4	8	56.9	6.7	36	92	7.94	
1/5	9	63.1	6.2	*	94	7.89	
2/5	10	68.0	4.9	35.5	97	7.87	
3/5	11	72.6	4.6	36	93	7.80	- 5
4/5	12	77.4	4.8	35	98	7.87	
5/5	13	84.3	6.9	36	98	7.88	
6/5	14	92.3	8.0	36.5	94	7.87	
7/5	15	100.6	8.3	37.5	96	7.84	- 1
8/5	16	109.5	8.9	36	74	7.86	
9/5	17	118.9	9.4	36	90	7.86	- 3
10/5	18	129.0	10.1	36	94	7.84	- 5
11/5	19	139.3	10.3	35.5	90	7.84	- 6
12/5	20	149.6	10.3	35.5	90	7.91	- 6
13/5	21	160.9	11.3	34.5	93	7.89	- 6
14/5	22	172.2	11.3	34	93	7.74	- 6
15/5	23	183.7	11.5	33.5	84	7.82	- 10
16/5	24	195.8	12.1	34.5	87	*	- 10
17/5	25	208.7	12.9	33	87	*	- 30
18/5	26	221.8	13.1	33.5	86	*	- 75
19/5	27	235.3	13.5	33.5	83	*	- 100
20/5	28	249.1	13.8	33.5	86	*	- 110
21/5	29	263.6	14.5	33.5	83	*	- 27
22/5	30						- 36
23/5	31						- 10

On d5 it was noted that, although no mortalities were removed, some healthy fish were removed from the bottom when the tank was siphoned. These were returned to the tank in an apparently healthy condition. On d7 it was noted that there were no larvae on the bottom of the tank.

The tank was totally dark until d5, when the cover was removed and the red light switched on. This gave a reading of 17 lux at the water surface. A white light was switched on on d10, this gave a reading of 94 lux at the water surface. All illumination was continuous.

The first sign of optical pigment was on d15 and, in order to minimise stress on transfer, the tank salinity was reduced by adding 10 lit of distilled water per day between d16 and d23. The first fish transferred to the rearing tank (on d15) sank immediately but fish transferred after d19 kept station in the water column.

None of the fish in this tank were seen to have taken feed although inert diet was added daily from d25 and live diet from d28. Fish transferred to the rearing tank were first seen to be feeding on d25.

Trancist	Яq	-0 \ű	.Ink	$\mathfrak{D}^{\circ}_{-}\mathfrak{T}$	$\sigma_{\mathcal{Q}}$	m	67.3%
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gove on distrand, noted that, elthough no mertalicles were removed, some healthy first war removed from the bettom when the true archemed. These were returned the carried and animal constitution and the configuration of the constitution of the constitution of the the tenth of the tenth.

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Home of the fish in this cank pare maen to have tagen feed although inert distance was acceded daily trem d25 and live dist from d28. Fich sugnators of to the rearring tank care tiess need to be reading on d25.

Table 7 Larval rearing - March 1987

Date	D	D°	T°	Sal.	Feed	Fish transfer
6/3	24	186.5	10.6	32.5		+ 3
7/3	25	195.9	10.7	*		+ 2
8/3	26	205.3	10.0	*	lm rots + alg	+ 2
9/3	27	215.3	10.2	*		+60
10/3	28	226.0	10.2	* '	lm rots + Selco	+46
11/3	29	236.2	10.2	*	Plankton	+ 2
12/3	30	246.1	9.9	33		+ 2
13/3	31	256.2	10.1	*	1.1m rots	+ 2
14/3	32	266.5	10.3	34		+ 2
15/3	33	277.1	10.6	33		+ 2
16/3	34	287.5	10.4	*		+ 2
17/3	35	298.1	10.6	*	Feeding activity	+ 2
18/3	36	308.3	10.2	*	-	+ 2
19/3	37	318.5	10.2	32.5		+ 2
20/3	38	327.7	9.8	*		
21/3	39	336.3	8.6	*		+ 2
22/3	40	345.1	8.8	34	5000 Art naup	
23/3	41	354.3	9.2	33.5	6000 " "	
24/3	42	363.7	9.4	33	7000 " " + Plank	•
25/3	43	373.3	9.6	*	5000 " "	
26/3	44	382.7	9.4	33.5	6000 " "	
27/3	45	392.4	9.7	*		
28/3	46	401.7	9.3	*		
29/3	47	410.8	9.1	33.5		
30/3	48	420.9	10.1	*		
31/3	49	431.4	10.5	*		
1/4	50	442.3	10.9	*	Last 2 fish seen	
2/4	51	453.4	11.1	*		

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Date	D	D°	T°	Sal.	D/0 ₂	pН			7	ransfer
10/5	18	129.0	13.7	33	90	7.88				
11/5	19	139.3	13.5	33	86	7.88				+ 1
12/5	20	149.6	13.4	33	90	7.88				+ 1
13/5	21	160.9	13.6	*	81	7.77				+ 1
14/5	22	172.2	12.8	33	85	7.61				+ 1
15/5	23	183.7	13.7	33	80	7.64 + P1	ankton			+ 5
16/5	24	195.8	13.7	32.5	85	*				+ 5
17/5	25	208.7	14.1	32.5	84	*				+15
18/5	26	221.8	14.1	33.5	95	*				+50
19/5	27	236.6	14.8	33	84	*				+50
20/5	28	251.7	15.1	33.5	82	*			-Med	+60
21/5	29	266.9	15.2	33	85	*			-Med	+27
22/5	30	282.2	15.3	33.5	89	*			-Med	+36
23/5	31	297.9	15.7	33	90	*			-Med	+10
24/5	32	314.1	16.2	33	92	*				
25/5	33	330.7	16.6	33	87	*			-Med	
26/5	34	347.7	17.0	32.5	83	7.79			-Med	
27/5	35	346.6	16.9	33.5	75	7.67			-Med	
28/5	36	381.2	16.6	33.5	83	7.59				
29/5	37	397.3	16.1	32	84	7.64			-Med	+ 1
30/5	38	413.6	16.3	33	89	7.67			-Med	
31/5	39	430.3	16.7	33	89	7.74 +500	00 Art	nau	P	
1/6	40	446.9	16.6	32.5	85	7.80 +0.4	m "	**		
2/6	41	463.8	16.9	33.5	88	7.74 +800	00 "	11		
3/6	42	480.9	17.1	33	87	7.76 + 1m	•	11	-Med	
4/6	43	497.7	16.8	33.5	89	7.67 + 1m	11	11		
5/6	44	515.5	17.8	33	87	7.76 +0.6	m ¹¹	n,	+alg	
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7/6	46	549.9	17.2	33.5	85	7.86 + 1m	11	11	11	
8/6	47	566.9	17.0	33	84	7.83 + 1m	11	"	11	
9/6	48	583.9	17.0	33	87	7.85 + 1m	11	11		
10/6	49	600.4	16.5	34	82	7.83 +1.3	m "	11	71	
11/6	50	607.0	16.6	34	84	7.66 + 2m	"	***	11	
12/6	51	623.7	16.7	34	84	7.73 +1.4	m "	***	**	
13/6	52	640.5	16.8	33	84	7.74 + 1m	11	**	**	
14/6	53	657.2	16.7	33.5	87	7.67 +0.5	m "	**		
15/6	54	674.0	16.8	33.5	87	7.77 +0.5	m "	**	**	
16/6	55	691.0	17.0	34	87	7.72 +1.8	8m "	11	Ħ	
17/6	56	708.0	17.0	34	87	7.72 +1.6		11	**	
18/6	57	724.9	16.9	33.5	87	7.79 +1m		**		
19/6	58	741.8	16.9	34	88	7.78 +0.4	·m 11	11		
20/6	60	758.9	17.1	33.5	87	7.81 +0.3		11		
21/6	61	776.1	17.2	33.5	90	7.85 +0.8		11		
22/6	62	793.4	17.3	33.5	87	7.81 +0/7		**		
23/6	63	810.9	17.5	33.5	96	7.93 +0.5		**		- 4
24/6	64	828.2	17.3	33.5	90	8.11 +0.5		11		
25/6	65	845.5	17.3	*	91	7.84 +0.5		11		- 4
26/6	66	862.7	17.2	33	89	7.93 +0.5		Ħ		
27/6	67	880.3	17.6	33.5	98	7.81 +0.5		**		
28/6	68	897.9	17.6	*	*	* +0.5		11		
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