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Rearing of turbot larvae (Scophthalmus maximus L.) on cultured food organisms; results from the 1979 and 1980 rearing seasons+)

by
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Abstract
In 1979 and 1980 two generations of turbot larvae were fed with mass-cultured food organisms such as rotifers (Brachionus) as well as nauplii, copepodids and adults of copepods (Eurytemora and Acartia). In the 1979 experiments the larvae reached metamorphosis after 30 to 36 days and at a length of 22 mm, rearing temperatures being 13.2° to 17.8°C. In 1980 the temperatures ranged between 17.0° to 21.8°C and the larvae metamorphosed from day 17 to day 26 when they measured 16.8 mm. Survival rates from day 3 to metamorphosis were 50 % for the 1979 and 34 % for the 1980 experiment.

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Introduction

Based on assessments of the biological and economic factors, PURDOM et al. (1972) have demonstrated the potential of the turbot (Scophthalmus maximus L.) as a species for marine aquaculture. The successful weaning on dry diets and postmetamorphosis feeding with artificial feedstuffs for further fattening is described by BROMLEY (1978), PURDOM et al. (1972), and KUHLMANN et al. (1980). The development of turbot production on a commercial scale has been impeded by difficulties in the larval rearing process. Almost all experiments concerning rearing techniques used rotifers for initial feeding, followed by Artemia nauplii and adults until metamorphosis (JONES, 1972; KINGWELL et al., 1977; HOWELL, 1979). In trials with exclusive Artemia feeding, the survival rates were found to be very low (< 10 %) and the time span until metamorphosis was very long (up to 70 days). Therefore a key factor effecting successful propagation was probably the lack of suitable larval nutrition.

Recently, the feasibility of weaning early stage turbot larvae of 6 - 17 mm (2 - 3 weeks old) on to commercial salmon starter diets was demonstrated experimentally, obtaining good growth and survival rates of 10 to 70 % during the weaning phase (BROMLEY, 1978). The partly replacement of Artemia by foodstuffs with variable ingredients contributed to an improvement of the rearing procedure. An alternative approach is seen in the use of different cultured planktonic invertebrates right from the beginning of larval feeding.

In the aquaculture station Kiel-Bülk, experiments during the last years aimed at the establishment of cultures of algae and different invertebrates as a basis for rearing marine fish larvae. After a period of testing the technical and biological foundations for the cultivation and suitability of green algae (Nannochloris), rotifers (Brachionus plicatilis), calanoid and harpacticoid copepods (Eurytemora affinis, Acartia tonsa, and Tigriopus) and mysids (Neomysis integer) (NELLEN et al., 1979; WITT et al. 1980), first experiments were carried out on the rearing of turbot larvae as consumers of the cultured organisms.

This paper deals with the 1979 and 1980 outdoor experiments on the rearing of turbot larvae on cultured food organisms up to O-group fish. Some observations on the weaning and growth of the O-group fish on different artificial and natural diets are added.

Material and methods

During the spawning seasons 1979 and 1980 eggs of turbots from the Western Baltic Sea (Langeland) were fertilized in filtered sea water (15 $^{o}/oo$ S). For incubation the eggs were spread out on a 500 µm nylon gauze that was mounted horizontically in a partly shaded 2 m^3 tank. This had to be done as the eggs of this subspecies of turbot are not buoyant. (KUHLMANN & QUANTZ, 1980). A constant water flow of ambient brackish water was set with salinities of 15.4 ($\pm$0.3) ‰ and temperatures of 15.5 ($\pm$0.3) ^{o}C in 1979 and 16.3 $^{o}/oo$ S ($\pm$0.9) and 16.8^{o}C ($\pm$1.3) in 1980. These temperature-salinity conditions were found to be close to the optimum for the incubation of turbot eggs of this origin (KUHLMANN & QUANTZ, 1980).

After hatching the water flow was stopped and the nylon screen removed. For the 1979 experiment the newly hatched larvae remained in the incubation tank and cultured green algae (Nannochloris) were added. For the 1980 experiment the 2 days old larvae were transferred from the incubation tank into a 3 m^3 rearing tank which had been prepared with an algae population.

Temperature, salinity, pH, NH_4, and NO_2 were measured regularly. Mean values and range during the experiments were as follows:

	Trial I (1979)		Trial II (1980)	
	Mean	Range	Mean	Range
°C	15.1 ± 1.4	13.2 - 17.8	19.2 ± 1.4	17.0 - 21.8
S ‰	16.9 ± 0.7	15.6 - 18.0	17.2 ± 0.3	16.6 - 17.8
pH	8.74± 0.2	8.40- 8.92	8.36± 0.3	8.06- 8.86
NO_2 ($\mu g \cdot l^{-1}$)	7.5 ± 6.4	6.6 - 20.3	2.39± 1.4	0.7 - 5.6
NH_4 ($\mu g \cdot l^{-1}$)	67.3 ±49.5	14.0 -201.6	78.0 ±73.8	2.8 - 215.6

The larvae were measured for total length, standard length, wet weight, and dry weight. At the same time the stomach content was examined. The density of the food organisms was determined daily. Mortality rates were estimated from numbers of dead larvae regularly siphoned from the tank bottom and from the remaining larvae at the end of the rearing phase as well. In 1980 the initial number of larvae could be exactly determined during transfer, as only an estimate could be taken in 1979. The feeding schedule was similar in the experiments of both years: Feeding started 3 days after hatch when the following cultured food organisms were added to the rearing tank:

1) rotifers (*Brachionus plicatilis*) and harpacticoid nauplii (*Tigriopus*) from a 2 m^3 *Brachionus* culture, fed with algae,
2) calanoid nauplii (*Eurytemora affinis* (80 %) and *Acartia tonsa* (20 %)) and
3) adult calanoid copepods (*E. affinis* and *A. tonsa*) and cladocerans (*Podon*) (the latter were available only in 1979) all harvested from 30 m^3 zooplankton polycultures.

The different food organisms were taken from the cultures by a selective harvesting system using different mesh-sized filters.

Results

I. Growth

Growth in weight for both year groups is shown as a semi-log curve in fig. 1. The data for dry weight given in Tab. Ia and Ib are partly measured and partly calculated from total length-weight-correlation formulae given by JONES (1972), which correspond well with the present data. From the growth curves and from daily growth rates for different periods it can be seen that after an initial stagnation of the weigth until the start of feeding, the larvae were growing quickly with max. growth rates of 23.0 % $\cdot$ d^{-1} in 1979 and 32.0 % $\cdot$ d^{-1} in 1980 (Tab. II).

With approach of metamorphosis the growth rates decreased. First changes indicating metamorphosis were observed from day 30 on in the 1979 trials. In 1980 such changes started already at day 17. During this time daily growth rates were 7.2 % (1979) and 1.9 % (1980), respectively. Later on the both growth curves were almost identical, the dry weights of the 1980 larvae being little less than the weights measured in 1979.

The growth in length (Fig. 2) was also very similar in both years with slightly higher values for the 1980 larvae grown at higher temperatures.

II. Food

Initial feeding was started from the third day on by offering a mixture of rotifers (Brachionus) and nauplii (Eurytemora, Acartia and Tigriopus) to the larvae. In the course of the experiment bigger organisms (copepodids and adults of Eurytemora and Acartia) were added to the rearing unit. The daily average concentrations of food organisms per ml were:

	1979	1980
Brachionus	3.2	2.0
nauplii	2.0	0.5
copepodites }	0.7	2.6
adult copepodes }		1.3
Podon	0.05	-.-

The stomach analysis are listed in Tab. Ia and Ib. In the 1979 experiment the ingested food organisms shifted from smaller (nauplii) to bigger ones (copepodids) from day 14 on at a fish length of 5.7 mm. Though Brachionus was present at high density the larvae did not feed on them during this year. In 1980 a change from nauplii to Brachionus was observed between day 6 and 10 when the fish length measured 5.6 mm. The change to adult copepods began at day 21 (12.4 mm). At this length the larvae of the 1979 batch turned to feed on Podon. Podon could not be offered in 1980.

During daylight, no distinct rythm of food intake was observed. The number of food organisms in the stomachs of the larvae, sampled every 2 hours during the light phase of 3 days in 1979, increased towards the evening. The same findings were reported by LAST (1978) for other species of pleuronectiform larvae.

The stomach contents of the larvae expressed in ug dry weight are shown in the lower part of Fig. 1. The weights of the ingested food were quite similar in the two years until day 26. Then the uptake of Podon increased the stomach content of the larvae from 1979. During metamorphosis a decrease in food uptake can be seen in both cases (metamorphosis 1979: day 30 - 36, 1980: day 17 - 26).

RILEY (1966) described similarly declining ingestion rates during metamorphosis for the plaice (Pleuronectes platessa). The decrease in food intake resulted in a weaker growth, as was mentioned above.

III. Mortality

The viable hatch during the breeding period under the given temperature and salinity conditions was close to 50 % (KUHLMANN & QUANTZ, 1980). The 1979 experiment was started three days after hatching with approximately 500 larvae which had stayed in the same tank since hatching. In the course of the experiment 200 of the fish were taken for investigations. These larvae were taken into consideration for further calculations. Highest mortality (41 %) occurred between day 17 and 22. This was the time during which the swimbladder was filled. At the end of this period a survival rate of about 50 % was estimated. No further mortality was observed afterwards. In the 1980 experiment 650 larvae were transferred from the hatching unit into the rearing tank. This handling may have induced a high mortality in the following days. A further mortality during swimbladder filling was not observed during this year. 42 larvae were taken for measurements. The overall mortality from day 2 to day 40 was calculated to be 66 %.

IV. Weaning and postmetamorphosis growth

After metamorphosis the young turbots could be weaned easily on different non-living food such as pelleted trout feed and pieces of herring meat. Trout feed was accepted after a period of 5 days during which only dry feed was offered. The herring pieces were accepted from first feeding on. No mortality occurred during the weaning phase and in the following weeks. In 1979 comparative feeding experiments were carried out using trout feed (starter feed 1 mm), herring meat, living mysids (Neomysis integer) from cultures, and combinations of herring and mysids and trout feed and mysids (KUHLMANN et al., 1980). Though a dry diet was accepted, it did not result in as good a growth rate as was observed when herring meat was fed (Fig. 4). The overall daily growth rates (calculated after WINBERG, 1960) varied from 4.07 % for trout feed to 4.58 % for herring meat fed turbots. Food conversion expressed as food dry weight per gain in fish wet weight was estimated to be 0.54 for herring meat and 1.43 for trout feed. The addition of living mysids to the diets positively influenced the growth and food conversion rates.

Discussion

The most common method for rearing fish larvae is based on the food organisms Brachionus and Artemia nauplii. Instead of the relatively artificial food organism Artemia, we alternatively offered mass-cultured zooplankton organisms which cooccur with the larvae in their natural environment. With this possible nutritional deficiencies may best be avoided. In Japan, the food combination of rotifers and copepods is already successfully employed in the cultivation of the red sea bream and other marine fish while in Europe, all efforts to rear turbot larvae up to now relied on the use of Brachionus and Artemia only.

The turbot larvae showed different food preferences in 1979 and 1980. In the 1979 experiment almost no Brachionus were taken when offered together with nauplii. In 1980 Brachionus played a significant role as a food organism after nauplii were not taken anymore and before copepodids were accepted. The reason for this is unknown. In spite the walls and bottom of the tanks were densely colonized with harpacticoids, these copepods were found only occasionally in the stomachs.

The growth curves of the larvae of both batches are similar, differences are found mainly for the first 10 days, when the 1980 larvae grew with a daily rate of about 30 %. This was probably due to higher rearing temperatures in 1980. Stomach contents were found to be lower in the good growing fish larvae, but the smaller number of larvae investigated have to be kept in mind when discussing these differences. The relatively slower weight increase during the second half of the 1980 experiment was likely caused by the lack of bigger food organisms (Podon) during this year. Podon was also found to be the most important food organism for turbot larvae caught in the North Sea (LAST, 1979).

Comparable growth data are available only from HOWELL (1979) and SCOTT & MIDDLETON (1979), who reared turbot larvae at 17°C and 16°C, respectively. HOWELL reported a length of 8 mm at day 17 compared with 6.9 mm to 9.0 mm in our study. In the growth experiments of SCOTT & MIDDLETON who added different species of algae to the food, the larvae grew to a maximum size of 11.5 mm until day 31. In our experiment mean length then amounted to 19.6 mm (1979) and 22.0 mm (1980), respectively.

Contrary to suvival rates of 50 % and 34 % from day 3 to the end of metamorphosis as we had observed, KINGWELL et al. (1977)

reported an average survival of only 15 % for the period from day 2 to day 20 and JONES (1974) achieved maximum survival of 4.6 % from day 7 until metamorphosis.

The lower survival rates in 1980 may be explained by the stress that was induced by the transfer of the early larvae. Another reason for better survival may have been an earlier bloom of micro-algae in the rearing tank during 1979. The high mortality in 1979, probably caused by difficulties in filling the swimbladder, did not occur in 1980. PURDOM et al. (1972) suggested that difficulties may be due to low oxygen tension, but as the oxygen saturation in the two years were similar in our experiments further studies on this phenomenon are necessary.

Another improvement related to the feeding of natural food organisms to the turbot larvae is the time the larvae need to reach metamorphosis. This lasted 30 - 36 days in 1979 and only 17 - 26 days in 1980 when the mean rearing temperature was almost 4°C higher than in the 1979 experiment. This was much shorter than the period of 45 - 70 days found by JONES (1972) at a rearing temperature corresponding to our temperatures in 1979. We found that metamorphosis seems not to be size dependent.

The present data indicate that the chosen breeding and feeding strategies and the quality of the food organims offered were responsible for good growth and suvival as well as for a short time period the larvae needed to perform metamorphosis. This also may be a prerequisite for good growth rates of the O-group fish after being weaned to non-living diets such as fish meat or pelleted dry feed.

Tab. Ia: Growth and stomach content (average number and dry weight of food organisms · larvae^{-1}) of turbot larvae reared in 1979

days from hatching	larvae invest.	TL (mm)	dry weight (mg)	Brach.	Nauplii	ad. Cop.	Harpact.	Podon	insect larvae (Corethra)	average stomach cont. (ug dry weight)
				stomach content (n)						
3	9	3.38	0.042	-	-	-	-	-	-	-
4	12	3.51	0.040	-	-	-	-	-	-	-
5	20	3.59	0.060	-	4.5	-	-	-	-	1.3
6	9	3.82	0.074+	-	8.8	-	-	-	-	2.6
7	25	4.10	0.101+	-	14.0	0.8	-	-	-	7.0
8	35	4.20	0.116+	-	10.8	0.1	0.15	-	-	4.0
9	7	4.56	0.170+	0.1	39.8	0.7	-	-	-	14.4
10	5	4.63	0.186+	-	20.0	1.6	-	-	-	11.6
12	5	5.02	0.268+	0.6	25.0	0.8	-	0.2	-	13.0
14	4	5.70	0.484+	-	32.5	0.7	-	0.7	-	20.6
17	5	6.90	1.10 +	7.8	3.0	6.6	-	2.2	-	54.3
22	9	10.66	3.76	-	-	8.9	-	12.0	-	175.2
26	5	15.32	11.4	-	-	39.2	-	12.8	-	290.8
30	5	18.52	20.4	-	-	121.4	-	28.0	-	761.0
33	5	21.5	26.8	-	-	62.0	-	-	-	217.0
36	5	22.6	31.6	-	-	42.0	-	5.0	-	207.0
45	4	29.4	70.4	-	-	106.0	10.5	4.3	1	1708.0

+) Calculated after JONES (1972):

$$\ln W = 4.6695 \ln L - 7.2446 \quad (<7 \text{ mm})$$

$$\ln W = 3.0575 \ln L - 4.3013 \quad (>7 \text{ mm})$$

Dry weights of food organisms:
(own measurements)

Eurytemora affinis nauplii	0.3 ug
Copepodits + adults	3.5 "
Brachionus plicatilis	0.5 "
harpacticoides	2.5 "
Podon spec.	12.0 "
Corethra (insect larva)	1260.0 "

Tab. Ib: Growth and stomach content (average number and dry weight of food organisms • larvae^{-1}) of turbot larvae reared in 1980

days from hatching	larvae invest.	TL (mm)	dry weight (mg)	stomach content (n) Brach.	Nauplii	Copepod.	adult Copep.	Polychaet larvae	Average stomach cont. (ug dry w.)
1	7	2.64	0.050	-	-	-	-	-	-
4	3	3.49	0.042	-	9	-	-	-	2.7
6	3	-	-	-	13	-	-	-	3.9
10	5	5.65	2.33	16	3	1	-	-	11.4
13	5	6.45	4.31	11	-	-	2	-	9.0
18	5	10.2	5.24	13	-	11	2	-	43.0
21	4	12.4	5.23	17	-	-	43	2	230.0
26	4	16.8	12.2	-	-	-	57	-	256.5
31	3	22.0	20.0	-	-	-	116	4	578.0
40	54	27.8	36.2	not examined					

Dry weights of food organisms:
(own measurements)

Adult copepods	4.5 ug
Copepodites	2.5 "
Nauplii	0.3 "
Brachionus plicatilis	0.5 "
Polychaet larvae	14.0 "

Tab. II: Growth of turbot larvae from hatching until end of rearing experiment. The daily growth rates were calculated from dry weights (Tab. Ia + b)

	Growth period (days after hatching)	Daily growth rates (% weight increase per day, after WINBERG, 1960)
	3 - 4	-4.9
	3 - 5	8.7
	5 - 10	23.0
1979	10 - 17	20.3
	17 - 22	21.9
	22 - 30	17.2
	30 - 36	7.2
	36 - 45	8.4
	1 - 4	-5.8
	4 - 10	32.0
	10 - 13	20.0
1980	13 - 21	2.4
	21 - 26	16.0
	26 - 31	9.7
	31 - 40	6.4

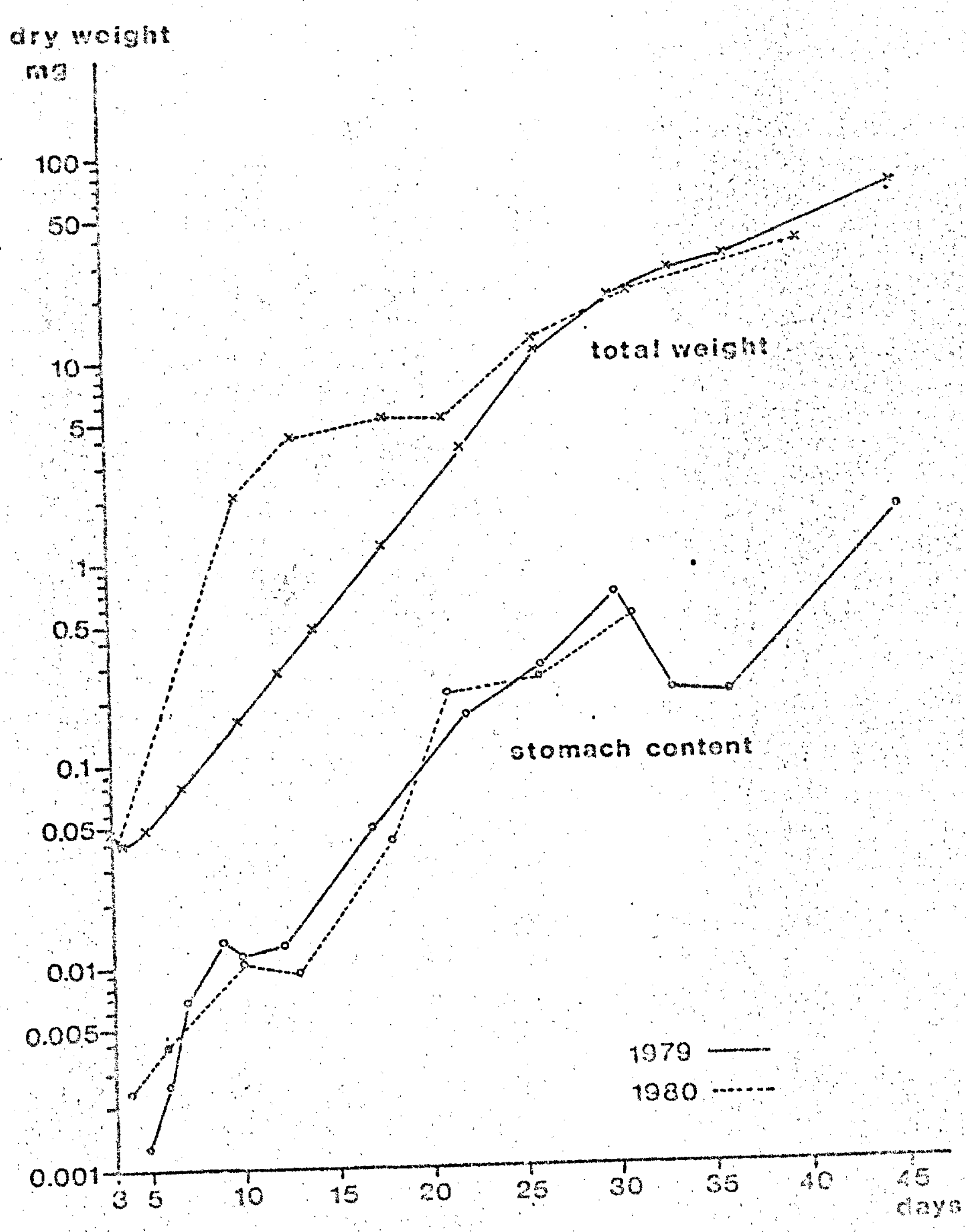
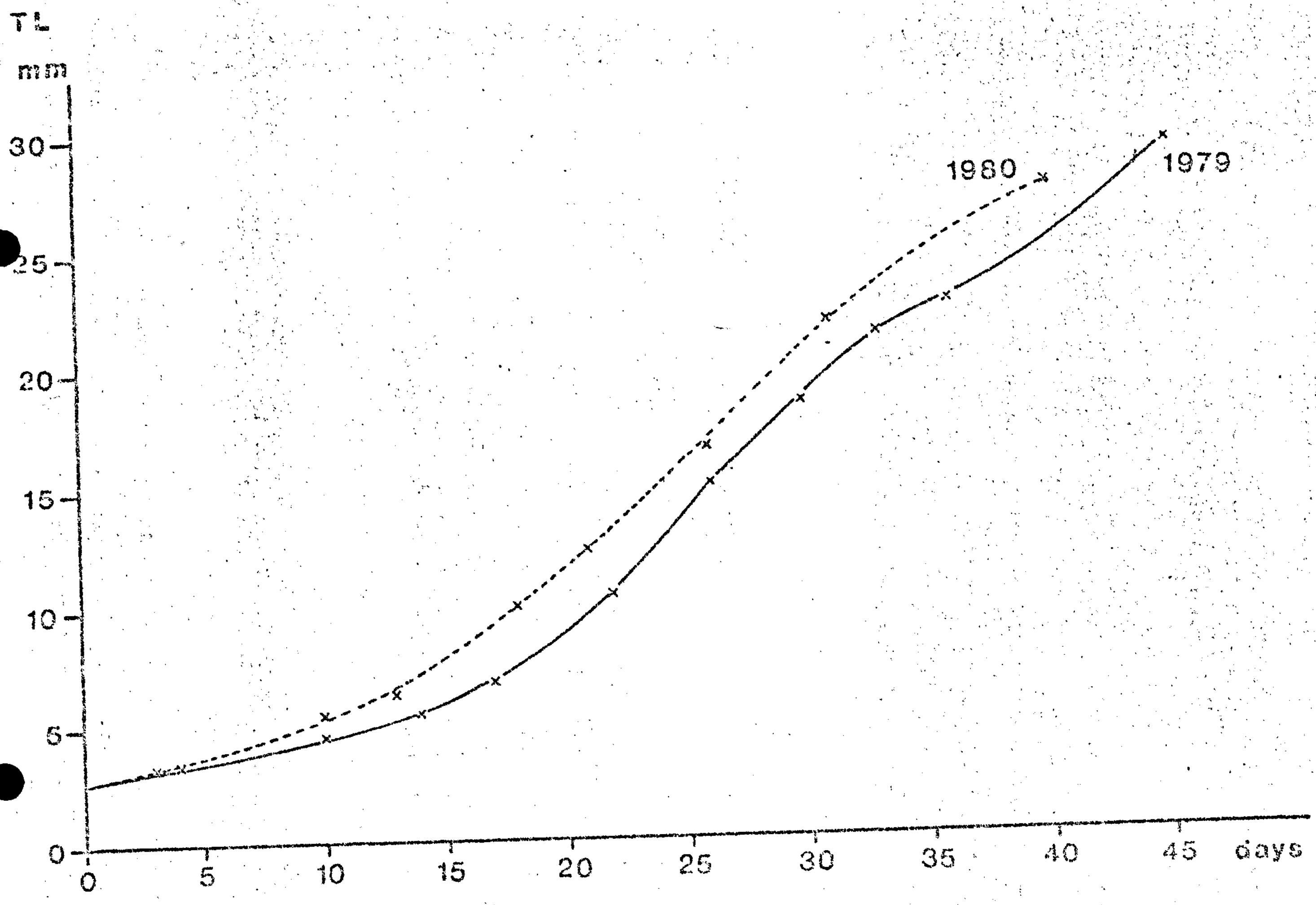


Fig. 1: Weight growth (x——x) and stomach contents (o-----o) (mg dry weight) of two generations of turbot larvae in 1979 (full line) and 1980 (broken line)



<u>Fig. 2:</u> Length growth until day 45 after hatching

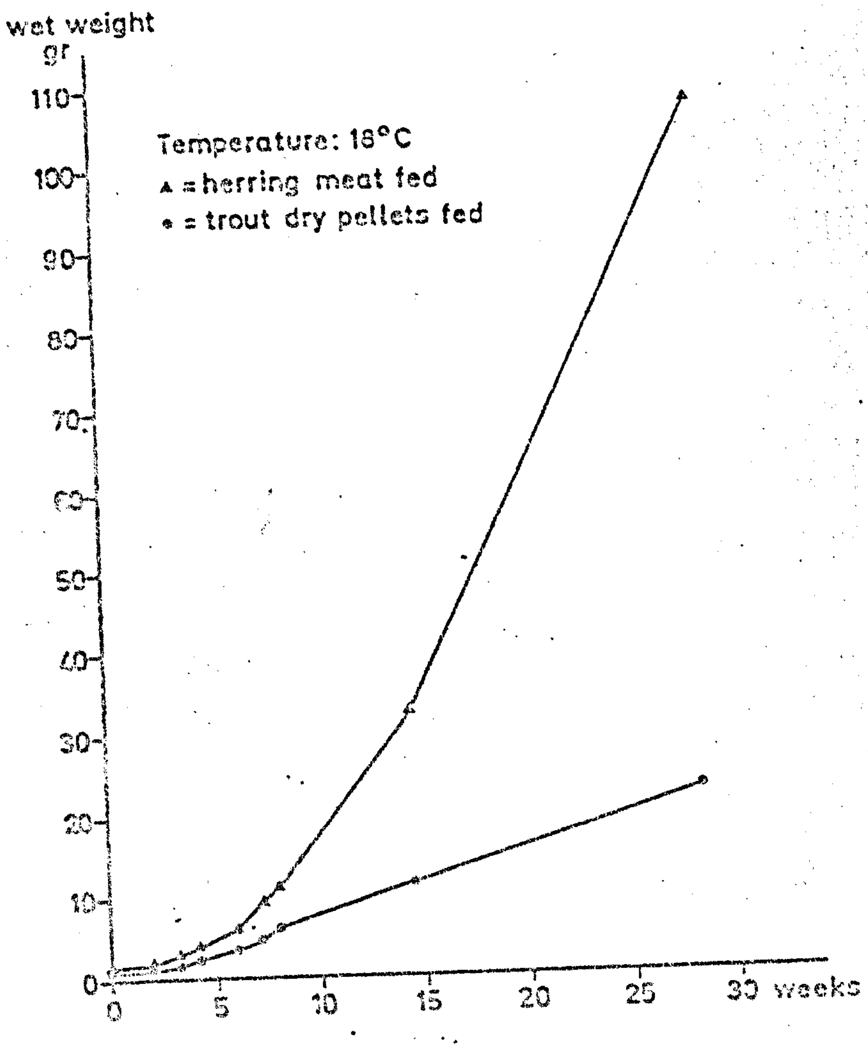


Fig. 3: Weight increase of O-group turbot during 1979 feeding experiments. (Daily rations: herring meat 25 % wet weight of food per total weight of fish, trout feed 10 % dry weight of food per total weight of fish)

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