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EFFECT OF DIFFERENT FOOD LEVELS ON THE GROWTH AND SURVIVAL OF LABORATORY-REARED SEA-BASS LARVAE (*DICENTRARCHUS LABRAX* (L.))

by

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ABSTRACT.

Four batches of 7 500 larvae were reared during a period of one month in 150 l tanks at 18° C. The first one was fed on the basis of estimates of the required daily amount of organisms that were made by one of the authors ; the second batch was fed on the average of the estimates made by both authors ; the third and the fourth batches were fed, respectively, with, 20 % above and 20 % below this average.

The densities of food in the tanks varied from 0 to 16 organisms/ml before the daily meal and from 1 to 25 organisms/ml after it. There was no significant difference in the final weight or length of the larvae, although the larvae receiving the highest food level showed a significantly better growth at the age of 15 days. Survival seemed to be better at lower feeding levels. The food conversion rates varied from 34.1 to 15.3 (wet weight) and from 18.7 to 8.4 (dry weight). The best rates were obtained at the lowest feeding level.

RESUME.

Quatre lots de 7 500 larves de bar sont élevés pendant un mois dans des bacs de 150 l, à 18° C. Ces larves reçoivent quotidiennement des quantités différentes de proies vivantes. Le premier lot est nourri sur la base d'une estimation de la quantité de nourriture à donner faite par une seule personne, le second sur la base de la moyenne des estimations faites par deux personnes, le troisième et le quatrième respectivement 20 % au-dessus et au-dessous de cette moyenne.

La concentration des proies dans les bacs varie de 0 à 16 proies/ml avant la distribution des repas et de 1 à 25 proies/ml après. Malgré une croissance plus rapide des larves suralimentées, pendant les deux premières semaines, aucune différence significative ne subsiste en fin d'expérience. La survie finale semble meilleure dans les lots recevant moins de nourriture. Les taux de conversion de la nourriture varient de 34,1 à 15,5 en poids humide et de 18,7 à 8,4 en poids sec. Le meilleur résultat est obtenu pour le lot le moins alimenté.

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INTRODUCTION.

The effect of different feeding levels on the growth and survival of marine fish larvae, reared in the laboratory, has been already investigated for several species. Many authors have tried to maintain a fixed density of prey in the tanks by means of a more or less regular adjustment of the amount provided, without looking closely at the daily food consumption by the larvae (ALDERSON and BROMLEY, 1973 ; HOUDE, 1975, 1976 ; RILEY, 1966 ; SAKSENA and HOUDE, 1972 ; O'CONNELL and RAYMOND, 1970 ; WYATT, 1972). An approach to examining the daily food intake of larvae at different prey concentrations was made by ROSENTHAL and HEMPEL (1970) for *Clupea harengus* and LAURENCE (1976) for *Pseudopleuronectes americanus*.

Results change from species to species. If the prey density is very low, mainly at the early larval stages, the mortality is very high (HOUDE and PALKO, 1970). At higher levels the larvae can tolerate a very wide concentration range (usually from 1 to 6) without catastrophic loss, but the conclusions are contradictory. For some authors there is a positive relation between food density and growth or survival (ALDERSON and BROMLEY, 1973 ; HOUDE, 1975 ; SAKSENA and HOUDE, 1972, *pro parte* ; WYATT, 1972). For others there is no relation at all (HUNTER, 1972 ; HOUDE, 1976 ; O'CONNELL and RAYMOND, 1970 ; RILEY, 1966 ; ROSENTHAL and HEMPEL, 1970). Further, in some cases a higher prey concentration may reduce the survival because of an accumulation of metabolites (SAKSENA and HOUDE, 1972, *pro parte*). However, none of these authors made any calculation of the conversion rate of the food by the larvae. Some information on this point can be obtained from SHELBOURNE (1968) for *Solea solea*.

In a mass production program, the amount of living food to be used is very important because of its high cost and low dependability (GIRIN and PERSON-LE RUYET, 1976 ; HOUDE, 1973). The amount of waste can be reduced, on the one hand by shortening the period before the fish is weaned, and on the other hand by restricting the living food input to the minimal daily requirements of the fish for optimal growth and survival. The first approach was recently investigated in sea-bass by the authors (BARAHONA FERNANDES and GIRIN, 1976). A preliminary investigation of the second approach was made by GIRIN (1976) who compared the effect of different feeding levels, and found that survival was better with the highest feeding scheme, but that the final weight was not significantly different at any food level.

In this paper we report a more detailed investigation of the problem of finding the minimum daily requirements for optimal survival and growth.

MATERIAL AND METHODS.

The rearing method described by GIRIN and al. (1975) is based on a daily addition (for technical reasons, at noon) of a definite amount of living food to the culture tanks. This amount is estimated from the feeding diagrams of previous experiments, from an estimate of the amount of prey left in the tank from the day before, and from the general appearance and conduct of the population. The estimate of the amount of remaining prey is made by looking through a 250 ml glass beeker containing a sample of the tank-water with some fish in it.

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Successful application of the above method requires much personal experience. The unavoidable difference between subjective estimates made by different people lead to differences in the supply of food, which in turn may well affect the survival or growth of the larvae. We accordingly designed an experiment with five batches, each containing 7 500 larvae from the same spawn and reared in similar physical conditions. One batch was to be fed following the estimates of one of the authors (estimated régime tank 1), the second using the estimates of the other author (estimated regime tank 2), the third the average of both estimates (average regime tank), the fourth 20 % above this average (increased regime tank), and the fifth 20 % below it (lowered regime tank).

An aeration failure in tank 2, two days after the beginning of the experiment, obliged us to change the initial procedure. The solution chosen was to use, for the average calculation, the estimates of both authors for tank 1 which, however, continued to receive only the food level estimated by the author originally assigned to it (fig. 1).

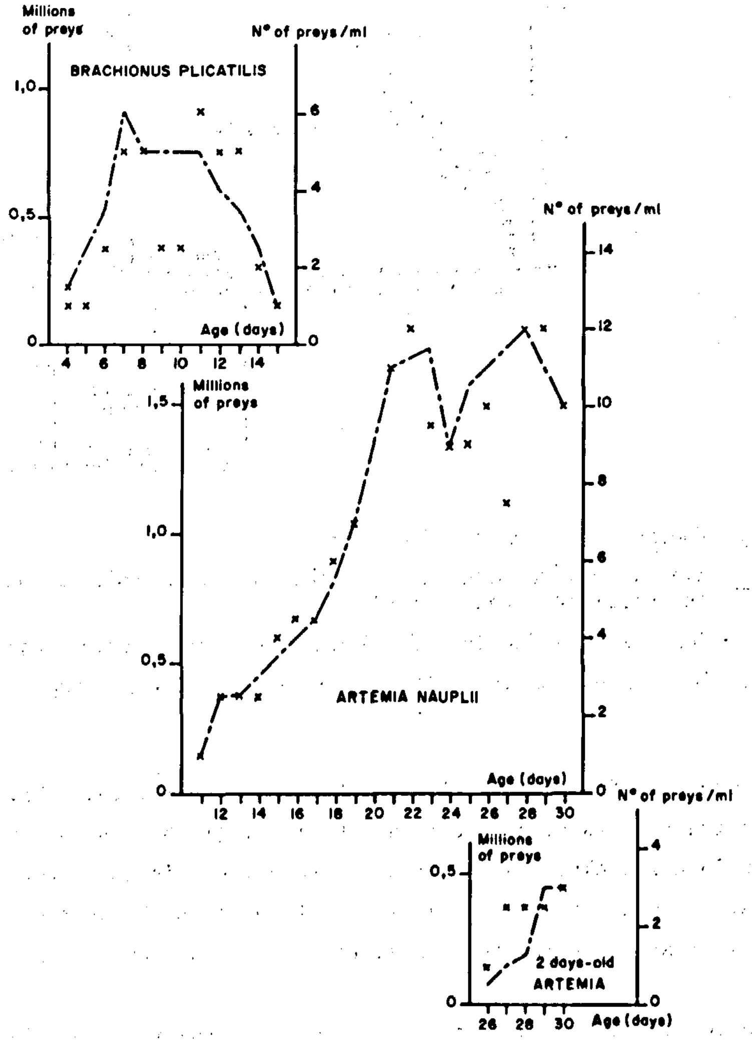

FIGURE 1 : *Feeding schema of the estimated regime tank.*

_._._._._ *Schema used, estimated by one author*

+++++++++ *Estimates made in the same tank by the other author* .../...

No change was made in the design of the experiment for the other three tanks (fig. 2).

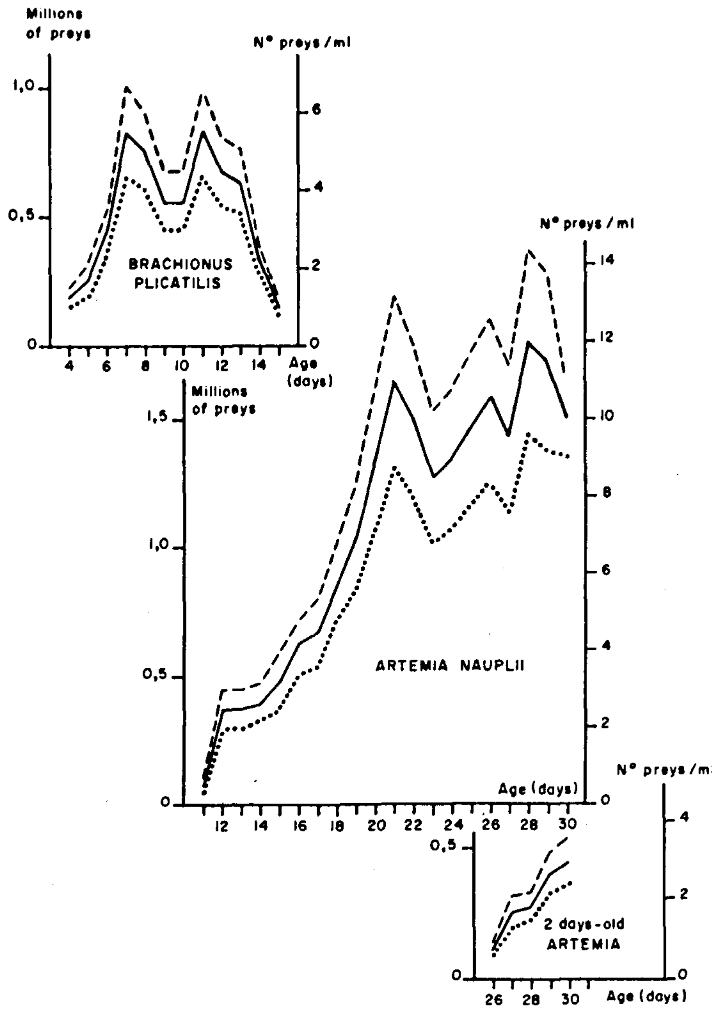

FIGURE 2 : *Feeding schemas at the three differents levels.*

———— *average regime tank (mean of the estimates made by the two authors in the estimated regime tank)*

-------- *increased regime tank (20 % more than the average regime tank)*

.... *lowered regime tank (20 % less than the average regime tank).*

All larvae came from a natural spawn hatched on April 23, 1975. The general rearing method was the same as described by GIRIN *et al.* (1975). The water temperature varied from 18.0 to 18.9° C in all tanks. The oxygen percentage level in the water ranged from 70 to 98 % in the estimated regime tank (tank 1) from 66 to 98 % in the average regime tank, from 73 to 96 % in the increased regime tank and 72 to 99 % in the lowered regime tank.

Ten and 20 days after hatching, an estimate of the total population in each tank was made by gently homogenizing the larvae in the tanks, mixing the water with an agitator and sampling one liter of the suspension for counting.

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The different living-food organisms used were carried to the fish culture room in separate and aerated containers. Their density was estimated from three samples of 1 ml. The mean error of the estimates was 15 %. Any specific desired quantity of prey could thus be given to a tank by homogenizing the contents of one of the containers and removing from it the appropriate volume.

The calculation of the number of remaining organisms in the fish culture tanks was made from three samples of 25 ml : one from the surface, another from the middle and one from the bottom of each tank. The difference between the highest and lowest counts at all levels in the three samples was 30 %.

Dead larvae were not reported because their small size results in a very high decomposition rate that invalidates any count, a problem noted also by ALESSIO (1976), ALDERSON and BROMLEY (1973), O'CONNELL and RAYMOND (1970) and WYATT (1972).

Every five days ten fish were sampled in each tank and preserved in 5 % neutralized formalin for later measurements of length and weight.

RESULTS.

The estimates obtained for survival at the ages of 10 and 20 days and at the final age of 30 days after hatching are plotted in table 1.

	Estimated regime tank		Calculated regime tank		Increased regime tank		Lowered regime tank	
Estimates at 10 days-old (3 samples)	32.7 % 36.9 % 44.6 %	39.0 %	50.6 % 76.9 % 83.3 %	71.8 %	50.6 % 62.5 % 64.6 %	59.5 %	38.7 % 55.4 % 65.4 %	54.4 %
Estimates at 20 days-old (2 samples)	21.3 % 37.3 %	29.5 %	44.2 % 57.3 %	50.7 %	47.5 % 67.7 %	57.3 %	60.6 % 62.2 %	61.4 %
Final survival 30 days-old (counted)	34.9 % (2611 survivors)		22.1 % (1656 survivors)		26.8 % (2011 survivors)		32.8 % (2468 survivors)	

TABLE 1 : *Survival percentages : estimates obtained at 10 and 20 days-old and final survival.*

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This table shows the low validity (wide range) of the estimates made at 10 and 20 days. Improved estimates could be obtained with better homogeneization before sampling, but this would harm the larvae. Other methods, such as using percentages of recaptured, previously coloured larvae, as made by TRANCART and DAOUD (in : BARNABE, 1976) do not seem more dependable than our technique. In any event, one has to choose between information and survival of fish.

Tests on the survival at the age of 30 days, by means of the normal derivate (SNEDECOR and COCHRAN, 1967), shows no significant difference between the estimated regime tank and the lowered regime tank, but significant differences in all other possible combinations. However, the power of this test is low and the initial 7 500 larvae introduced in each tank are not an exact number but an estimate.

The growth curves in weight and length were plotted in figure 3 from the average values of the samples and their 95 % confidence limits. The analyses of variance in weight

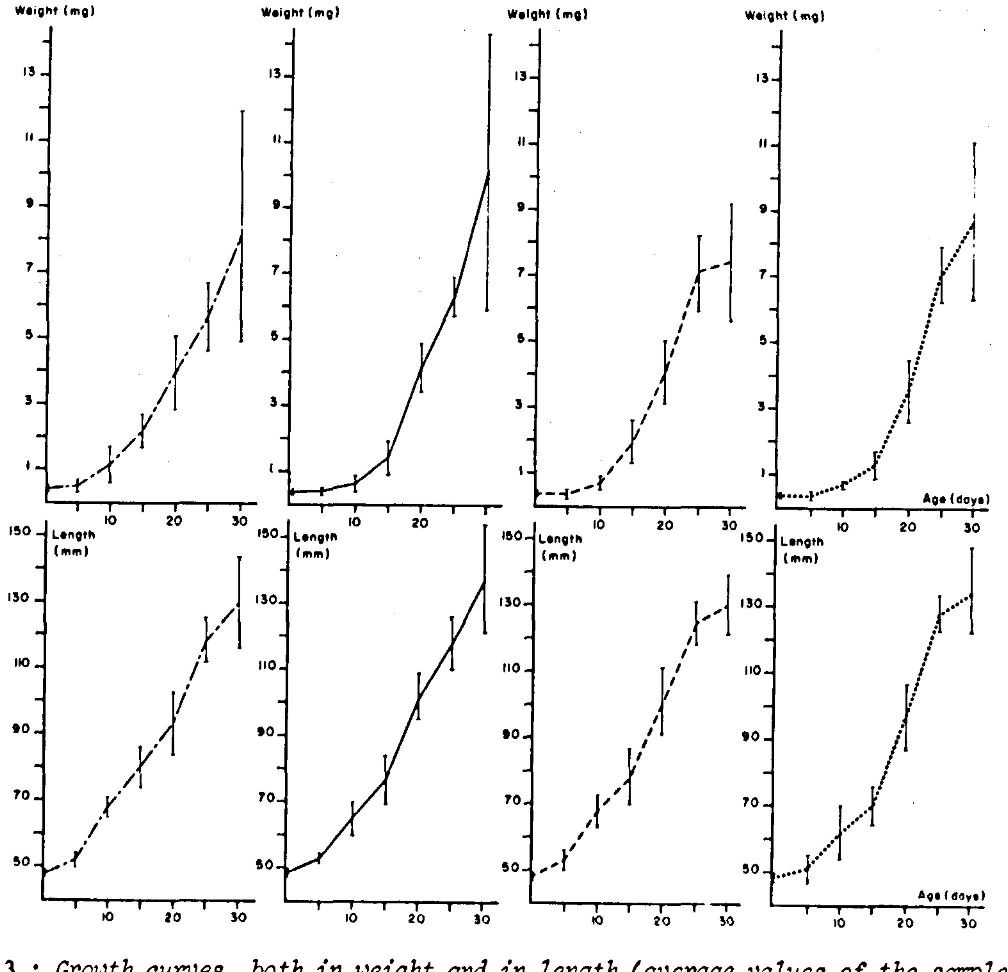

FIGURE 3 : *Growth curves, both in weight and in length (average values of the samples and their 95 % confidence limits)*

_._._._ *Estimated regime tank*
——— *Average regime tank*
------ *Increased regime tank*
...... *Lowered regime tank*

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at the ages of 5, 10, 15, 20, 25 and 30 days were made with orthogonal *a priori* comparison of the estimated regime tank againstall other tanks, of the average regime tank against the increased and lowered regime tanks, and of the increased regime tank against the lowered regime tank. There were no significant differences except between the increased and lowered regime tanks at the age of 15 days.

Figure 4 shows the daily evolution of the number and concentration of the prey in each tank. These curves are approximate because of the mean error introduced by the counting and by some natural mortality of the prey which was not taken into account.

Figure 5 shows the percentage of prey eaten, during a 24 h period, from the daily amount available in the average, increased and lowered regime tanks. The mean percentage of consumption for each type of prey and for all tanks is plotted in table 2.

	Estimated regime tank	Calculated regime tank	Increased regime tank	Lowered regime tank
Brachionus plicatilis	67.8 %	54.6 %	62.1 %	70.4 %
Artemia nauplii	93.7 %	63.2 %	60.7 %	75.5 %
2 days-old *Artemia*	87.1 %	79.5 %	57.1 %	64.8 %

TABLE 2 : *Mean daily food consumption percentage in each tank.*

Table 3 gives the data for the total amount of prey given to each tank in order to produce 30 days-old larvae, both in total number and in weight.

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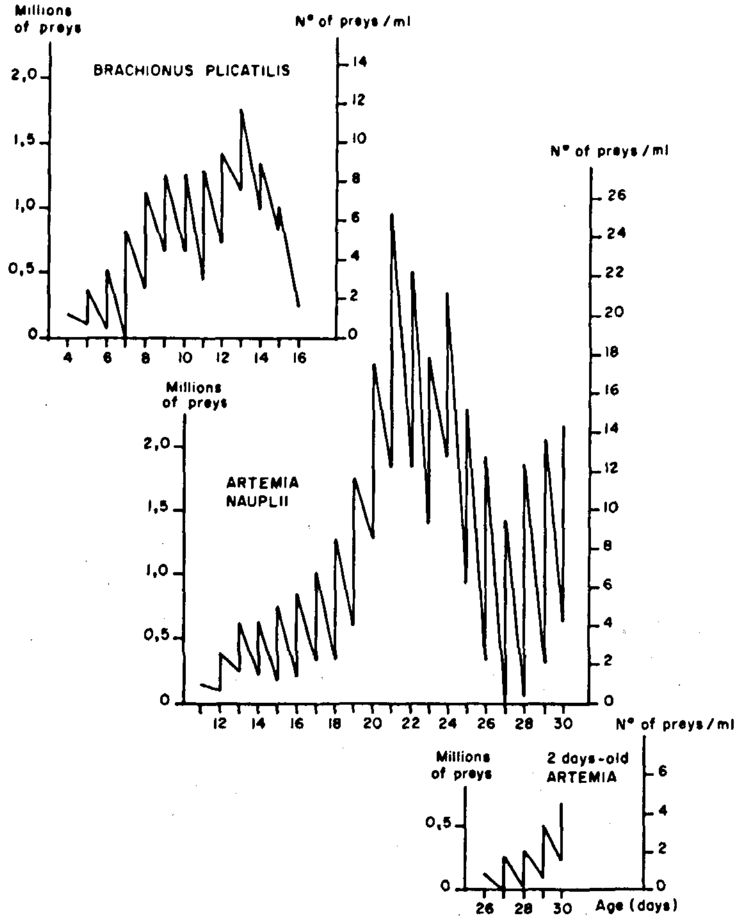

Fig. 4 b

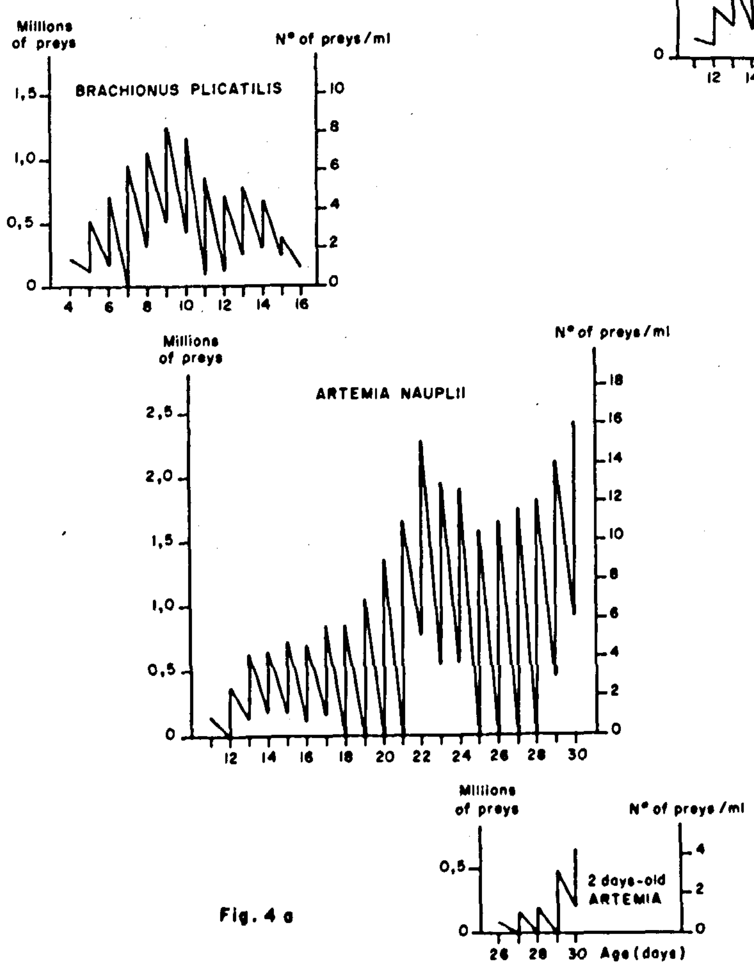

Fig. 4 a

FIGURE 4 : *Evolution of the food level in each tank, both in total number of preys and in concentration*

a) Estimated regime tank *b) Average regime tank*

c) Increased regime tank *d) Lowered regime tank* .../...

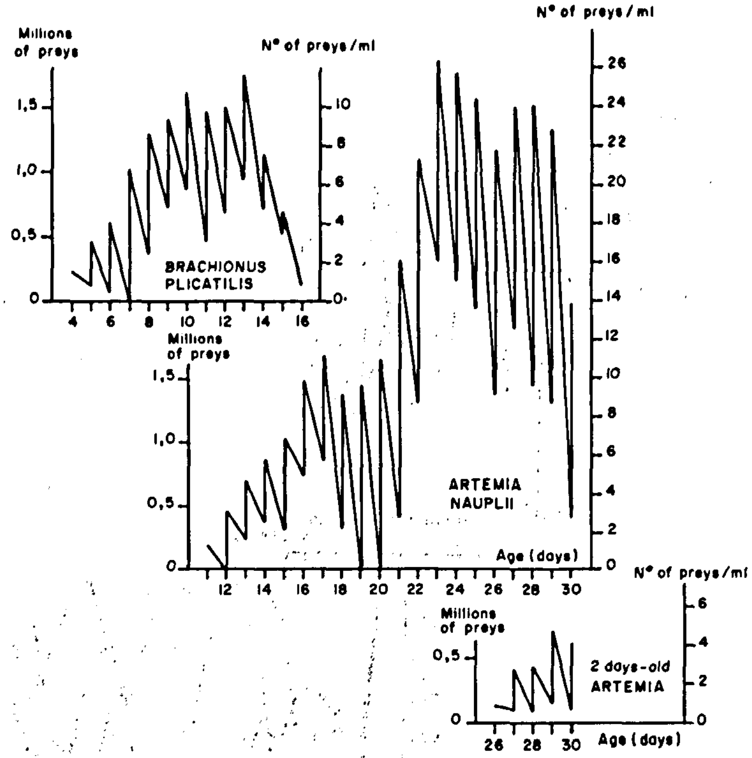
Millions of preys
N° of preys/ml
BRACHIONUS PLICATILIS
ARTEMIA NAUPLII
2 days-old ARTEMIA
Age (days)

Fig. 4 c

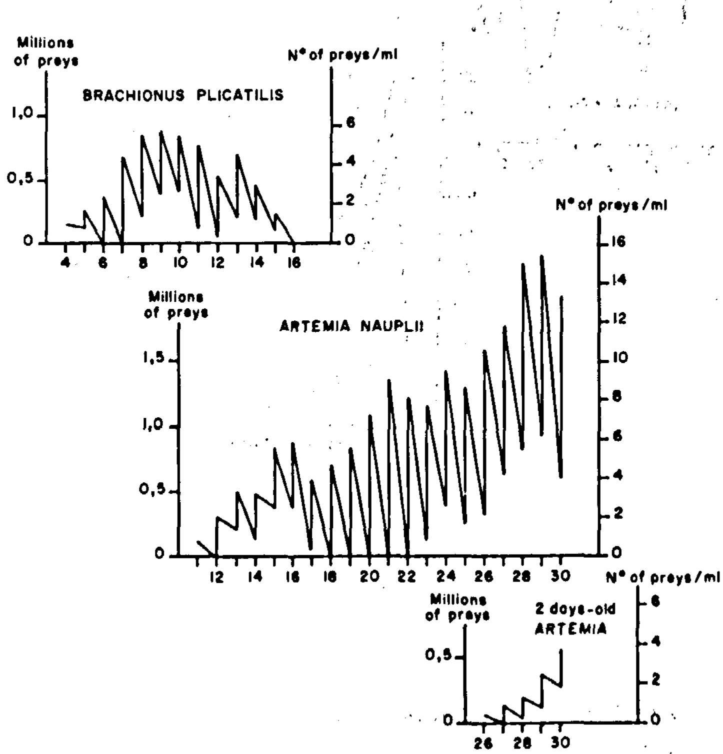
Millions of preys
N° of preys/ml
BRACHIONUS PLICATILIS
ARTEMIA NAUPLII
2 days-old ARTEMIA

Fig. 4 d

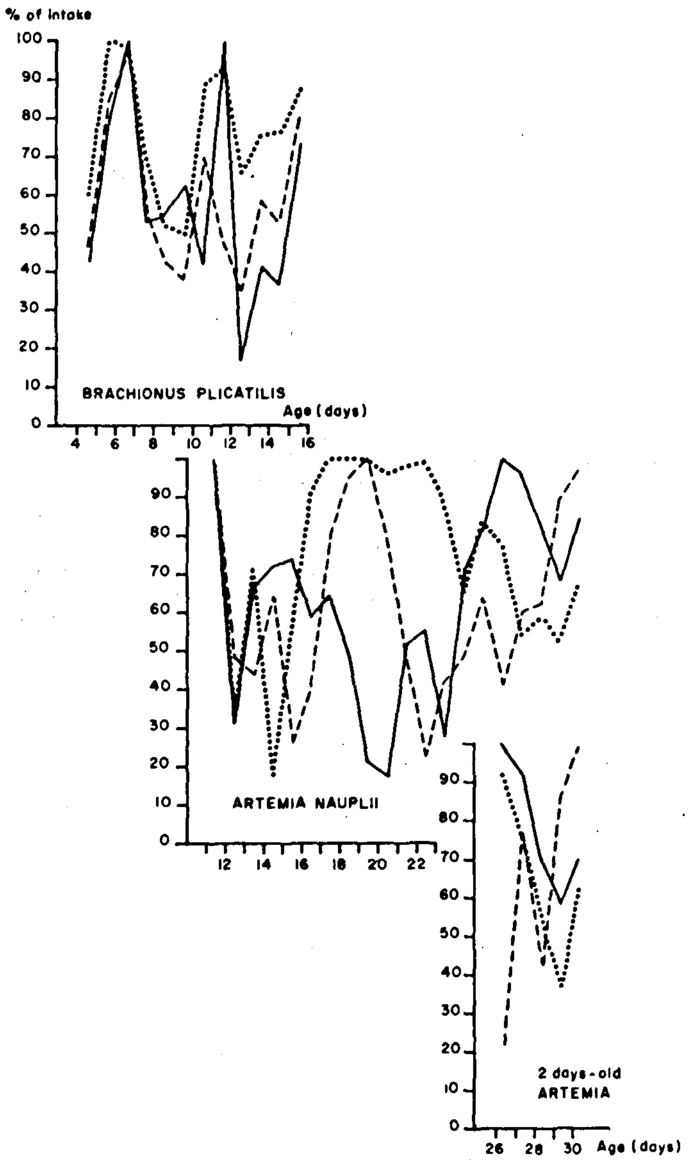

FIGURE 5 : *Daily percentage of the available preys eaten in 24 h.*

——— *Average regime tank*

------ *Increased regime tank*

...... *Lowered regime tank*

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		ESTIMATED REGIME TANK	CALCULATED REGIME TANK	INCREASED REGIME TANK	LOWERED REGIME TANK
Total number of given prey (x 10^3)	*Brachionus plicatilis*	6 675	6 225	7 470	4 980
	Artemia nauplii	22 200	21 712	25 860	17 370
	2 days-old *Artemia*	1 312	1 515	1 737	1 212
Wet weight of given prey (mg)	*Brachionus plicatilis*	16 020	14 940	17 928	11 952
	Artemia nauplii	295 926	289 428	344 714	231 542
	2 days-old *Artemia*	59 141	68 266	81 784	54 613
Total wet weight of given prey (mg)		371 087	372 633	444 425	298 107
Dry weight of given prey (mg)	*Brachionus plicatilis*	1 735	1 618	1 942	1 295
	Artemia nauplii	41 514	40 602	48 358	32 482
	2 days-old *Artemia*	4 371	5 045	6 044	4 036
Total dry weight of given prey (mg)		47 620	47 266	56 344	37 813

TABLE 3 : *Total amount of prey offered to each tank, both in total number and in weight.*

Table 4 gives the total number and weight of prey needed for the production of one 30 days-old larva in each tank. It was calculated both from the final survival data (table 1) and from the amount of food provided (table 3).

		ESTIMATED REGIME TANK	CALCULATED REGIME TANK	INCREASED REGIME TANK	LOWERED REGIME TANK
N° of prey needed to produce one 30 day-old larva.	*Brachionus plicatilis*	2 556	3 759	3 715	2 018
	Artemia nauplii	8 502	13 111	12 859	7 038
	2 days-old *Artemia*	503	915	902	409
Total wet weight of prey needed to produce one 30 day-old larva (mg)		142	225	221	121
Total dry weight of prey needed to produce one 30 day-old larva (mg)		18	24	28	15

TABLE 4 :*N° of prey to make one 30 day-old larva, both in total number and in weight.*

The mean increase of the weight for each batch of larvae and the food conversion rate for each tank (including consumption by larvae which died during the experiment) are plotted in table 5. The food conversion is given by the equation :

$$\text{Food conversion rate} = \frac{\text{Weight of the food offered}}{\text{Average weight gain of the individual larvae}} \times \%\ \text{survival}$$

		ESTIMATED REGIME TANK	CALCULATED REGIME TANK	INCREASED REGIME TANK	LOWERED REGIME TANK
Mean weight at age 0 (mg)	Wet	0.30			
	Dry	0.09			
Mean weight at age 30 (mg)	Wet	8.13	10.13	7.44	8.66
	Dry	1.92	2.51	1.82	8.37
Mean weight gain (mg)	Wet	7.87	9.87	7.18	8.37
	Dry	1.78	2.36	1.68	1.95
Conversion rate	Wet	19.2	25.1	34.1	15.3
	Dry	11.0	13.5	18.7	8.4

TABLE 5 : *Mean increase of wet and dry weights in each tank and the corresponding conversion rate.*

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Both the weight of food and the weight of larvae are expressed as dry weight for the calculation of the dry food conversion rate and as wet weight for the calculation of wet rate.

DISCUSSION.

The lack of replicates in the experiment, and the low validity of the population estimates in the tanks restrict possible analyses of the survival data. However, it seems that a higher feeding level might enhance survival during the first-feeding period, and have a negative effect later.

The accuracy of the information on growth is much better. A higher feeding level results in a significantly better growth at the age of 15 days, but there is no remaining difference at the end of the first month of life.

Taken together, the results tend to show, within the food-level range used, that the larvae benefit from a high feeding level in the beginning, but that this has not much influence on its later life in the given conditions. There is thus apparently no point in keeping the food level high at this stage. A similar conclusion was reached, for various species of fish larvae, by HOUDE (1975), HUNTER (1972), RILEY (1966), ROSENTHAL and HEMPEL (1970), SAKSENA and HOUDE (1972, *pro parte*), O'CONNEL and RAYMOND (1970), and WYATT (1972).

Our data fit well with the general idea that newly feeding larvae have a low predatory efficiency. The statistical chance of larvae and prey meeting would have a high influence on the amount of food eaten, and thus on growth and survival (ALDERSON and BROMLEY, 1973 ; LASKER, 1976 ; LAURENCE, 1976).

In a more advanced stage of larval development, when the larva has changed into a much more efficient predator, an excess of food may be more dangerous than useful. This danger may come from an accumulation of metabolites in the water (HOUDE, 1975 ; O'CONNEL and RAYMOND, 1970 ; SAKSENA and HOUDE, 1972).

In addition, particularly when the culture is made without algae, which was the case in this experiment, overfed larvae have more chances of eating *Artemia* which remained in the tanks for some time and have a low nutritional value (PAFFENHOFER, 1967).

The experiment reported in the present paper, and the previous experiment reported by GIRIN (1976), in which the feeding levels covered a wider range but involved smaller total quantities, lead to the conclusion that in the particular case of sea-bass the calculated estimates fulfil rather well the essential food requirements of the larvae. The unavoidable subjectivity of the method has only a very small effect on the results of the culture, inasmuch a difference of ± 20 % is hardly reflected in the survival data and has no effect on the growth achieved after 1 month.

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It seems that the larvae eat more whenever they have more available food, but do not grow faster. On the other hand, a useless excess of food will result in considerable waste and in lower efficiency : in this experiment, the food conversion rate was roughly twice as good at the lowest feeding level as it was at the highest.

Information on food conversion rates of marine fish larvae is very rare in the literature. Accounting for daily mortality is quantitatively impossible, and, without this information, one can calculate only a gross value for the food conversion rate from one age to another. The only such data available, were obtained by SHELBOURNE (1968) on sole (*Solea solea*) and at a temperature of 10 to 14° C with a survival of 42 and 44 %. SHELBOURNE calculated wet conversion rates of 13.1 and 12.6, from newly hatched to 4 month old fish. In our experiment, the lowered regime tank, with a 33 % survival, gave a wet food conversion rate of 15.3.

In conclusion, our data lead to the recommendation of designing feeding schemes with a high food level (*Brachionus plicatilis*) for the initial feeding period and thereafter with daily amounts of *Artemia salina* strictly restricted to the intake capacity of the larvae.

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