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## Effect of some ecological factors on the growth of the copepod *Schizopera elatensis* – a potential food organism for hatcheries

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### Abstract

Copepods are an important link in the marine food web, and, in particular, serve as food for fish larvae. *Schizopera elatensis*, a marine harpacticoid copepod, was reared in the laboratory, and the effect of various environmental factors, i.e., diet, temperature, salinity and height of water column on its growth was determined. *Schizopera elatensis* multiplies at a wide range of salinities, 10 ‰ to 70 ‰, and temperatures, from at least 18° C to an upper limit of 36° C, with an optimum of 25° C, and grows in high densities (300–400 individuals per ml): the lower the water column, the higher the density. The prospective use of *S. elatensis* as a live food organism in mariculture is discussed.

### Introduction

A reliable and abundant source of live food of appropriate size is obligatory for rearing marine fish larvae with a small mouth bore and is one of the major obstacles to the development of mariculture. Despite the large number of potential food organisms, only *Brachionus plicatilis* and *Artemia salina* nauplii are commonly and successfully used in hatcheries (KINNE 1977). Their size, however, which is too large for small-sized larvae, as well as their possible nutritional inadequacy (SCOTT and MIDDLETON 1979, HOWELL 1979, see also KAHAN 1980) is considered to be one of the crucial factors determining larval fate (NASH and KUO 1975). A mixed dietary regime of rotifers and various other live organisms including copepods during the first ten days of life gave better results (BRASOLA 1974, NASH and KUO 1975, NASH 1977, NELLEN et al. 1980). In nature, calanoid (KINNE 1977) as well as harpacticoid copepods (ALHEIT and SCHEIBEL pers. comm., SCHMIDT-MOSER and WESTPHAL 1981) are the most important food of young stages of various fish. This brought FUJITA (1973) to stress the importance of developing mass culture techniques for copepods for marine fish seed production.

Intensive attempts to grow copepods on a large scale have been carried out by many researchers (see KINNE 1977, KAHAN 1980). Nevertheless, copepods have not fulfilled expectations for use in mariculture, except for some species of *Tigriopus* and *Tisbe* which were used to a limited extent, due to the low densities obtained (FUJITA 1977, GIRIN and DEVAUCHELLE 1974; BRASOLA 1979).

This paper is part of a series (KAHAN 1979; KAHAN and AZOURY 1980; KAHAN et al. 1981) concentrating on the biology and growth of copepods as potential food organisms for hatcheries. Some ecological factors influencing mass cultivation of *Schizopera elatensis* were studied.

### Material and methods

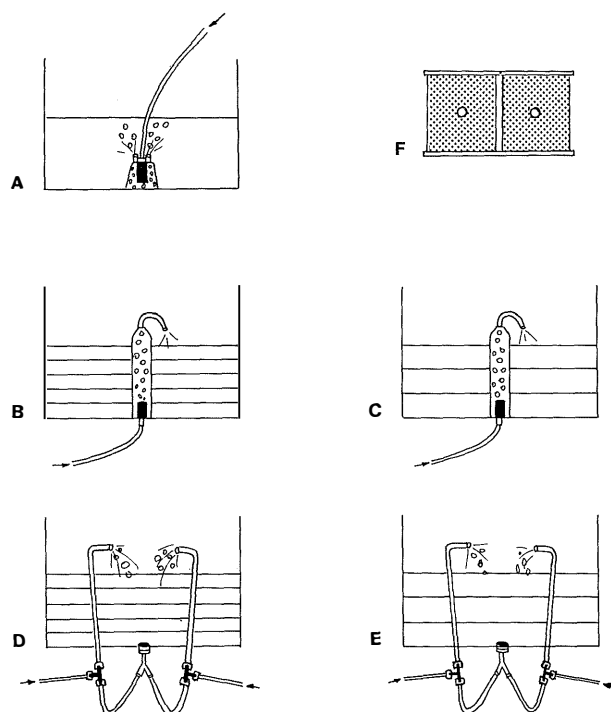
*Schizopera elatensis* was taken in samples from shallow intertidal gravel at the Aqaba Gulf near the H. Steinitz Marine Biological Laboratory, Eilat. It was found to be a new species of harpacticoid copepod of the family Diosaccidae (KAHAN and BAR-EL 1981).

In the laboratory, it was isolated from accompanying organisms and grown on microalgae which developed in light in a controlled temperature room at 18–20° C in cylindrical glass crystallizing dishes. Addition of a wheat grain and a mixture of microalgae, *Dunaliella tertiolecta* and *Phaeodactylum tricornutum*, was found to improve growth. However, better growth was obtained by using other diets, i.e. wheat bran and various vegetables. Fresh lettuce that floated on the water surface was chosen as the main food in the present study as it was found, earlier, to be more convenient (KAHAN 1979). *S. elatensis* was, previously, referred to as the unidentified thalestrid.

Copepods were grown in artificial sea water prepared from "Instant Ocean" salt mixture, usually at 35‰ unless otherwise stated. Water was not changed during experiments, which were carried out in three types of containers:

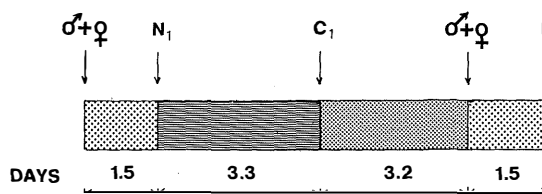
1. Round plastic vessels, Ø 1.7 and 1 cm in height, containing 1.5 ml medium, were used to determine number of nauplii per female and generation time at 28° C and 35‰ salinity. The small (50 µm), individually-layed eggs were difficult to discern among food particles and debris in the culture dish. This was the reason for determining fecundity not by the number of eggs laid per female, but on newly hatched nauplii. Movement of nauplii greatly facilitated their observation under low magnification in small-sized dishes. A young female and male were added to each vessel with a small fragment of fresh lettuce. After nauplii appeared in the vessel the couple was transferred to another dish under the same conditions. Observations were carried out daily except for every seventh day. Similarly, experiments were carried out in order to determine minimal generation time: time elapsed between first appearance of nauplii of one generation to first appearance of same stage of succeeding generation. The study was based on 200 observations.
2. Glass or plastic-covered dishes, Ø 6.5 and 4 cm in height, containing 50 ml medium, were used in order to determine the effect of temperature and salinity on growth. To start an experiment, ten pairs of females and males were added to each vessel with a grain of wheat and a piece of fresh lettuce (2 cm<sup>2</sup>), which was replenished when necessary. For determining copepod growth, three to four samples of 1 ml each were taken from each culture after stirring it thoroughly and distributing the sample in several glass depressions. Counting was carried out with the aid of a dissecting microscope for various developmental stages: nauplii, copepodites and adults. At least nine cultures were tested for each experiment.
  - a. Temperature: Growth at the following temperatures was studied for a period of 30 days at 35‰ salinity: 4, 18, 15, 28, 33, 42° C. The inoculum was from a culture grown at 28° C and 35‰.
  - b. Salinity: Effect of salinity on survival and growth was studied within the range 5 to 100 ‰. The inoculum was from a culture grown at 35 ‰ salinity at 28° C.
3. Glass rectangular aquaria (30 x 16 x 20 cm) were used in upscaling growth experiments in which the effect of aeration, surface area and height of water column were studied. Horizontal surface areas were used within the aquaria with different aeration apparatuses installed at the bottom. Aquaria shown in Fig. 1A contained an aeration stone covered by a 4 cm high cup-shaped chamber with two openings through

which air bubbles emerged. Aquaria shown in Figs. 1B and 1C had a recurved tube 16 cm in height and 3 cm in width, with an opening through which water dripped back to the aquaria. The aquaria shown in Figs. 1D and 1E had no air stones, but, instead, an air lift consisting of two narrow recurved tubes that conducted water back. To increase horizontal surface area, two to four horizontal plastic nets with 1 mm pore size were installed inside aquaria. The nets were kept taut by gluing them with an "H"-shaped glass frame (bars 1 cm in width), Fig. 1F. Similarly, aquaria shown in Figs. 1D, E contained two or four horizontal nets respectively. Lettuce leaves were also placed on these partitions. Growth studies were carried out for each aquarium after inoculating them to a density of 52 copepods per ml.



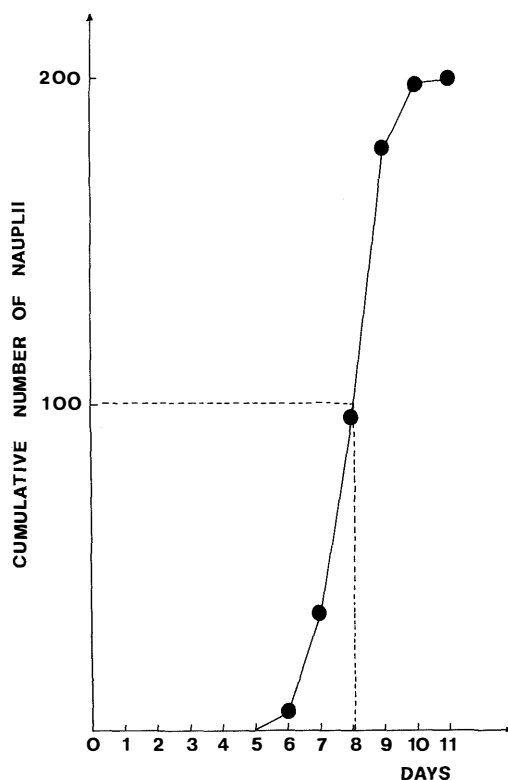
**Figure 1**

A-E: Diagrams of aquaria used with different horizontal surfaces and types of aerations. F: Top view of horizontal partition shown in D and E



**Figure 2**

Duration of life-cycle and different developmental stages of *Schizopera elatensis*



**Figure 3**

Minimal generation time of *Schizopera elatensis*: time elapsed between first appearance of nauplii of one generation to first appearance of same stage of the succeeding generation

Experiments to determine the influence on growth of the height of the water column were carried out with varying volumes of sea water, 0.5, 1, 2, 3, 4, 5 and 6 litres having a height of 1.1, 2.2, 4.3, 6.4, 8.5, 10.5 and 12.6 cm respectively with no aeration. Experiments were carried out in duplicates. Initial densities, the same for all aquaria, were 37 copepods per ml. Lettuce leaves were added when depleted.

## Results

### Fecundity and generation time

Average number of nauplii hatched per female, at 28° C and 35 ‰ salinity, is  $84 \pm 25$  over a period of 12–24 days. First appearance of nauplii is usually two days after copulation. Time lapse between nauplius and first copepodite is about 3 days, and to adult, 3 more days (Fig. 2). Minimal median time elapsed between first generation nauplius to second generation nauplius is 8 days (Fig. 3). Survival of nauplii to adult stage is 72 %.

### Temperature

Fig. 4 shows the effect of the various temperatures on the growth of *S. elatensis*. The optimal temperature was 25° C, reaching a total number of 12,290 copepods per 50 ml (density of  $245 \pm 20$  per ml) after 29 days, and a population doubling time of 3.2 days. However, over a shorter period (1–2 weeks), growth was better at 28° C and 33° C than

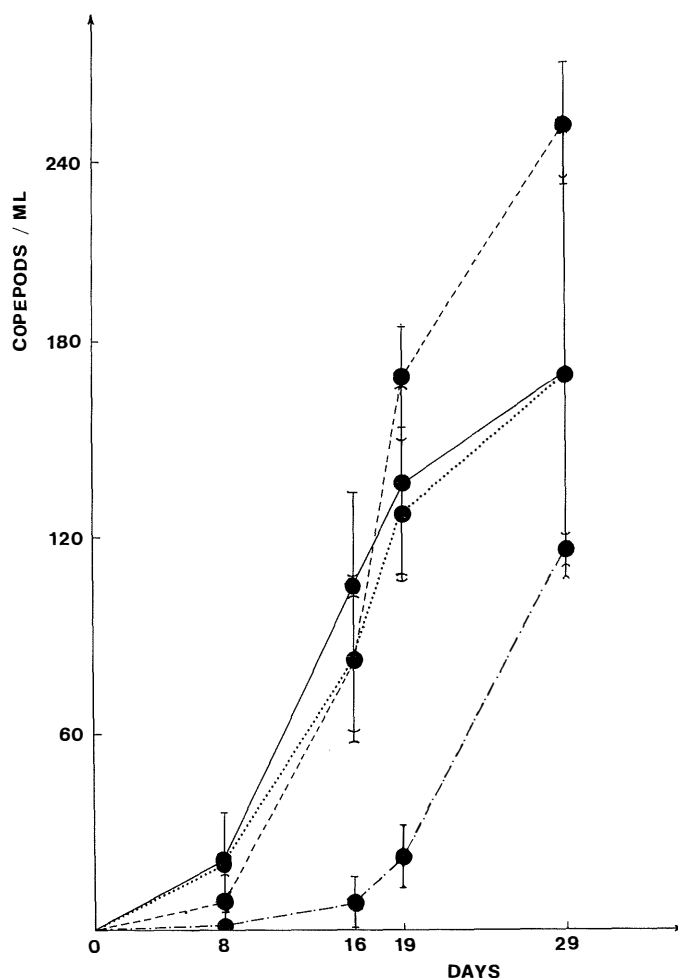


Figure 4

Growth of *Schizopera elatensis* at different temperatures, in 50 ml, at 35 ‰ salinity  
 18° C, ●— ; 28° C ●— ; 25° C ●- - ; 33° C ●.....

at 25° C. Copepods did not survive at the extreme temperatures tested: 4° and 42° C, but could multiply rapidly at 35°–36° C, as was observed in some additional experiments, in which extra attention was needed to control bacterial growth.

#### Salinity

Fig. 5 shows the growth of *S. elatensis* at various salinities after transfer from 35 ‰. No growth occurred at salinities lower than 5 ‰, and higher than 80 ‰. At these extremities, movement slowed down or stopped, but was restored on return to seawater salinity. The range of salinities in which copepods multiply well is wide: 10 ‰ to 60 ‰. At 2 ‰–10 ‰ and 60 ‰–70 ‰ multiplication is slow.

#### Cultivation in aquaria

No significant differences were obtained in growth of copepods in the 5-litre volume with the various set-ups of partitions and aeration (Fig. 1). Population reached a

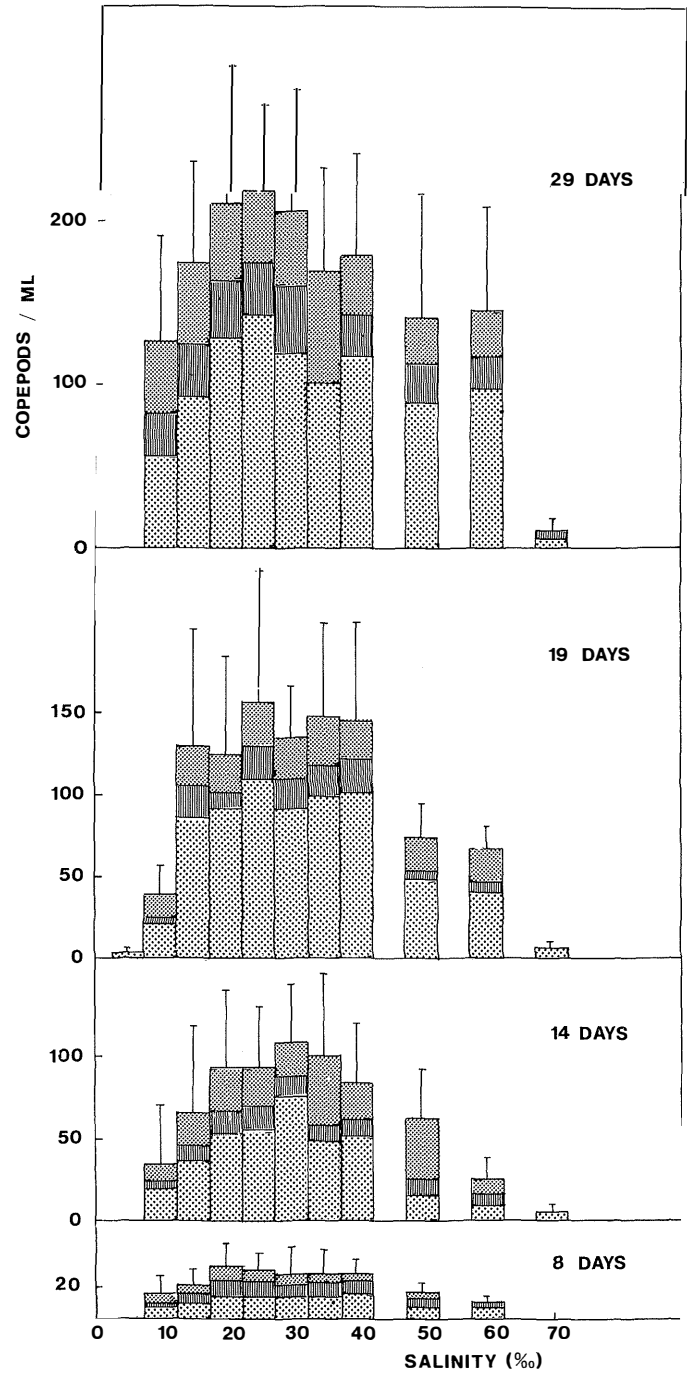





Figure 5

Growth of *Schizopera elatensis* after 8, 14, 19 and 29 days at different salinities, 5 to 70 ‰, in 50 ml at 28° C

nauplii  ; copepodites  ; adults 

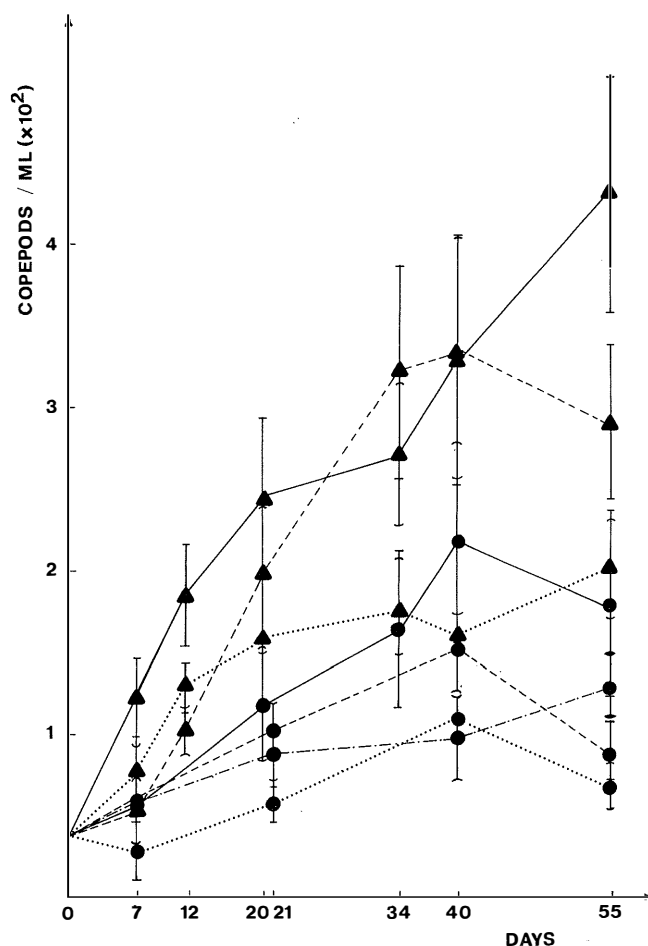


Figure 6

Growth of *Schizopera elatensis* at 25° C and 35‰ salinity in aquaria with same base area (30 x 16 cm) at various volumes (in litres)

0.5 ▲ — ; 1 ▲ - - - ; 2 ▲ .....  
 3 ● — ; 4 ● - - - ; 5 ● ..... ; 6 ● .....

plateau in all sets at about 70 individuals per ml. On the other hand, in experiments with different volumes (Fig. 6) the highest density of copepods was reached at the lowest volume,  $412 \pm 73$  individuals per ml after 55 days at 0.5 litre, decreasing with increasing volume to  $66 \pm 14$  individuals per ml at 6 litres.

### Discussion

Various criteria have been used for selecting copepod species for mass culture: hardiness, movement, diet, growth, etc. (IKEDA 1973, KITAJIMA 1973, OMORI 1973). Most of these criteria are met by *S. elatensis*.



It is an exceptionally hardy copepod, enduring a wide range of salinity, temperature and pH fluctuations (4–10) as was established in preliminary studies in accordance with the extreme, fluctuating conditions of its natural environment, the shore of Elat. The copepodites and adult stages are epibenthic, distributed throughout the water column and thus available to fish larvae, unlike other harpacticoids, which are mostly benthic or surface dwelling. *S. elatensis* thrives on a variety of diets, including market vegetables. As a non-selective feeder, it could be enriched with growth factors beneficial to fish larvae, a subject for further study.

*S. elatensis* is close in size to the smallest of the copepods listed by IKEDA (1973) (nauplii 50  $\mu\text{m}$ , adults 500  $\mu\text{m}$ ) and, accordingly, has one of the shortest median generation times (8 days) and a population doubling time of about 3–6 days. Furthermore, it lays eggs individually, a character that has been considered advantageous (CORKETT and ZILLIOUX 1975). On the other hand, its fecundity seems to be inferior, as one female produces 84 nauplii, while most species of copepods listed by IKEDA lay 100 or more eggs.

Density of organisms is an important factor determining size and therefore expense of containers and installations. The densities obtained in this study for *S. elatensis* are higher than those previously reported for growth of various other copepods (see KINNE 1977, ROTHBARD 1976, HOPPENHEIT 1978, GOPALAN 1977, KAHAN 1979, WALKER 1979, KAHAN and AZOURY 1980, and UHLIG 1980). In larger volumes, however, lower densities were obtained, similar to what has been noticed with *Tigriopus japonicus*, recommended as the most promising copepod species for mass culture (KITAJIMA 1973, FUJITA 1977). In our experiments, increasing the surface area by installing partitions in the aquaria or aerating by various methods did not improve densities. However, density was augmented by lowering the water column. This could be attributed to a rise in oxygen concentration due to an increase in the ratio of surface area to volume and not to aeration which creates turbulence. The latter probably interferes with copepod growth.

WALKER (1979) and FAVA and CROTTI (1979) indicated the importance of water renewal in excluding growth inhibiting factors. Since in this work water was not changed, further improvements in copepod growth may be achieved.

### Acknowledgments

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