Technical development in seed production of the Japanese sea cucumber, *Stichopus japonicus*

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Introduction

The sea cucumber *Stichopus japonicus* is a commercially-important species in Japan as a raw food, contrary to in China. There are three varieties of this sea cucumber: green, red, and black. The former two inhabit the sandy mud bottom of the bay, and are of great commercial importance. The other one can be found both in the open sea and on rocky shores. In Saga Prefecture, located on the northwest part of Kyushu Island, Southern Japan, the commercial catch of *Stichopus japonicus* has gradually decreased from 196 metric tonnes (t) in 1971 to 23 t in 1995 (wet weight). In the whole of Japan, the catch quantity has also decreased from 10 000 t in 1971 to 7000 t in 1995.

For the promotion of starting stock of the sea cucumber, the Sea Farming Center in Saga Prefecture has been producing 500 000 to 1 000 000 juveniles (10–20 mm in body size) per year since 1993. After metamorphosis from larvae, the juveniles grow up to 10–20 mm in size within the first three months, and grow up to an average of about 80 mm (max. 150 mm) size within one year. A protocol for the mass production of juveniles has been developed recently by the Center. Methods of brood stock management and culture of periphytic diatoms have been improved. Periphytic diatoms play two important roles: as a biological cue for the induction of larval metamorphosis, and as a suitable primary food for the juveniles.

Outline of juvenile production

Seed production usually begins with the catch of adult sea cucumbers from the sea in January. These animals are then reared for about three months. Propagation of periphytic diatoms on corrugated plates is also started in February. They are cultured at a density of more than one million cells/cm^2 within two months. In April, the induction of spawning of brood stocks is carried out, and larvae are cultured to a viable stage for about two weeks. After metamorphosis on the plates, the juveniles feed on periphytic diatoms and grow up to 10–20 mm body size within the first three months. These animals are shipped out to be released directly in the sea from July to August.

Brood stock management

Investigation into the maturation of natural sea cucumbers along the coast of the prefecture indicated that the spawning season lies within the March – May period (Fig. 1). As a result, about 100 adult sea cucumbers (300 g each) are caught in January and reared in a 2 t tank with the sea mustard, *Undaria pinnatifida*, at ambient temperature (12–18°C) to control the maturation. In April, the gonad index and the oocyte size in major axis are measured in random sampling to estimate the timing of the spawning induction (Fig. 2). When the gonad index and the oocyte diameter are more than 20 and 140 μm, respectively, spawning should be induced by heating the sea water (+ 5°C) of the tank (Fig. 3). From another investigation, it was found that the optimum density of spermatozoon for insemination is at 5–10 x 10^4 spermatozoon/ml or 1–2 x 10^3 spermatozoon per ovum.

Diatom culture

There are three important steps in propagating the periphytic diatoms on corrugated plates (40 x 32 cm), usually started in February in 15 t tanks (about 1000 plates each) (Figure 4, reprinted from Ito & Kitamura, 1998). The first is the enrichment with the addition of the nutrient salts (ammonium sulphate, superphosphate, Clewat32, and sodium

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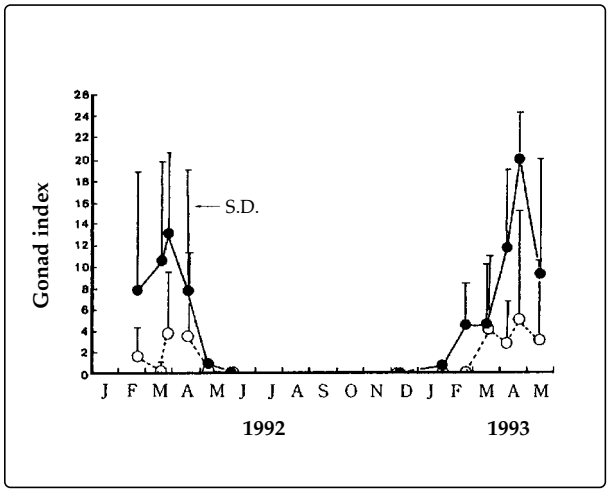

Figure 1

Seasonal changes in gonad index of the sea cucumber *Stichopus japonicus*

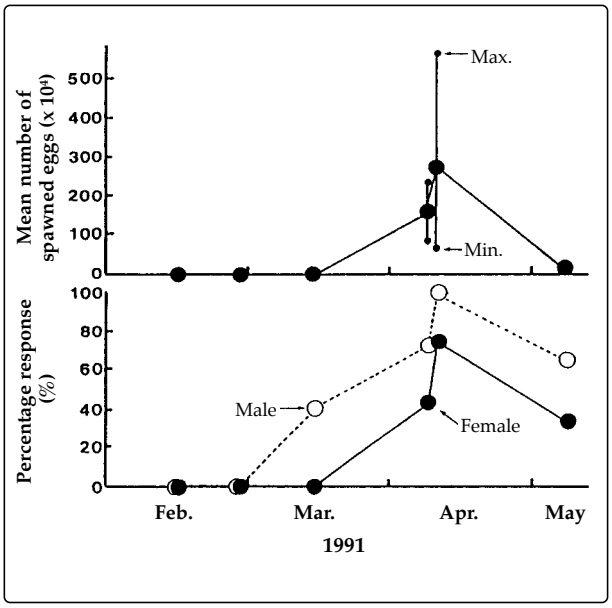

Figure 3

Percentage response of spawning and mean number of spawned eggs per female of the sea cucumber

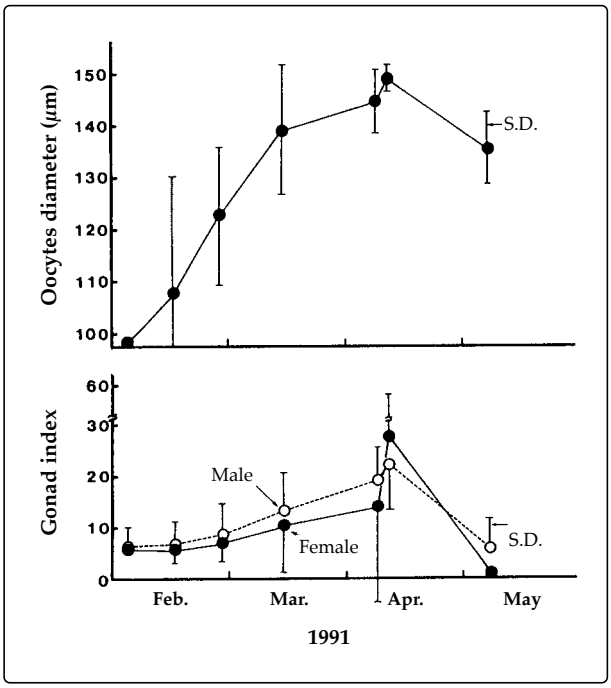

Figure 2

Changes in gonad index and oocyte diameter of the sea cucumber

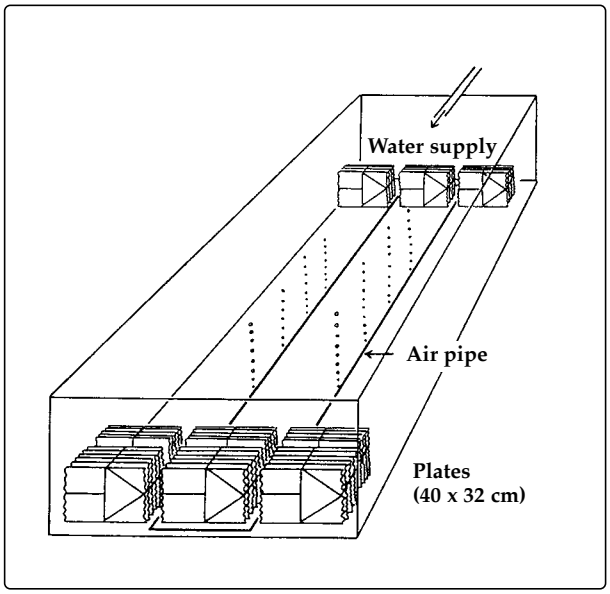

Figure 4

The schematic view of diatom culture in the 15 t tanks

silicate) in the tank under controlled light intensity. Light penetration is controlled by a black mesh sheet placed over the tanks. About 70 per cent of sunlight is shut-off on fine days and 50 per cent during cloudy or rainy days. The second step is washing with high-pressure sea water and reversing the plates once or twice a week. This method can significantly remove the larger diatoms that grow on the plates, thus making the smaller and highly adhesive diatoms dominant, which accelerates the propagation rate of the diatoms. And the last step is the elimination of copepods, which feed on diatoms, using the pesticide trichlorfon (0.5–1.0 ppm). As a result, we can culture small periphytic diatoms, such as *Navicula*, *Amphora*, *Achnanthes*, and *Nitzschia* at a density of more than one million cells/cm² on the plates within two months (see Figure 5, next page, reprinted from Ito & Kitamura, 1998).

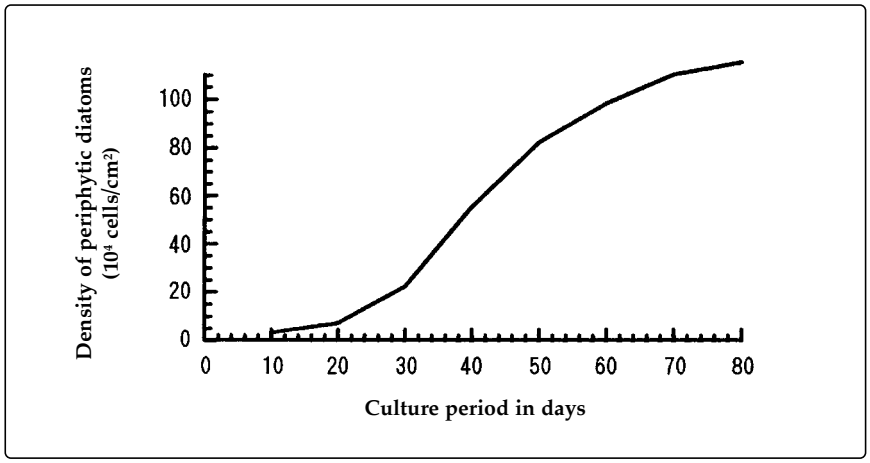

Figure 5

The typical propagation of the periphytic diatoms on the plates

Larval rearing and spat collection

After fertilisation, larvae of the sea cucumber are cultured to doliolaria larvae (Imai & Inaba, 1950) at 20°C by feeding the planktonic diatom, *Chaetoceros gracilis*, for about two weeks (Figures 6 and 7, reprinted from Ito & Kitamura, 1998). The larvae gradually increase in size through the 9th day. On the 11th day, body size of auricularia larvae reaches its maximum size, 900 μm. After that, body size falls to about 500 μm, and they grow up to doliolaria larvae. The induction of metamorphosis should be carried out at the stage of doliolaria. The metamorphosis of doliolaria larvae to juvenile is accelerated by the higher density of periphytic diatoms, and the density should be more than 200 000 cells/cm² to induce more than 50 per cent of larval metamorphosis (Ito, 1994) (Figure 8, reprinted from Ito & Kitamura, 1998). Periphytic diatoms are suitable primary food for the juveniles of this animal as for sea urchins (Tani & Ito, 1979; Kitamura et al., 1993) and abalones (Kawamura & Kikuchi, 1992).

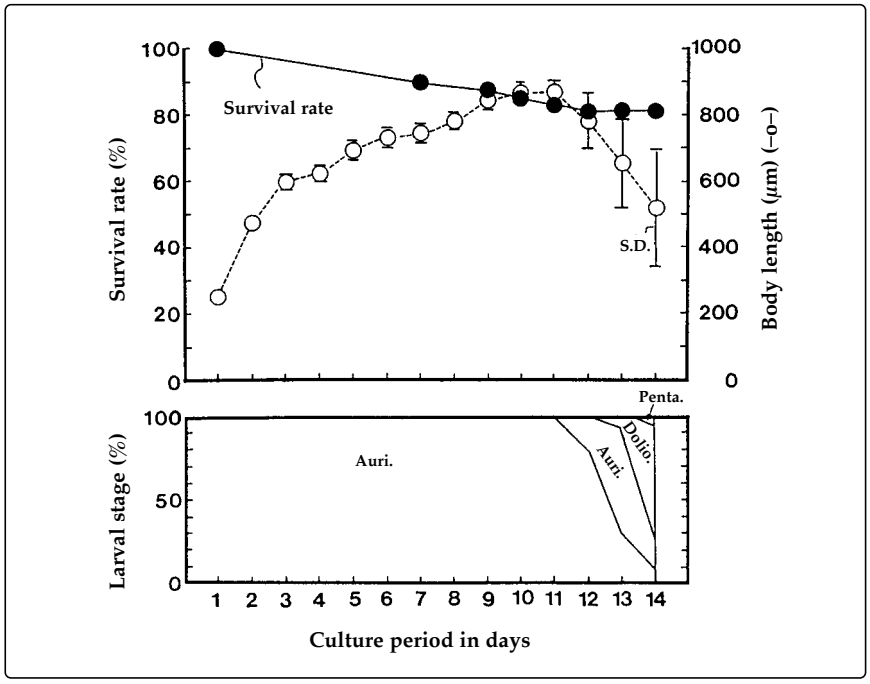

Figure 7

Growth of the larvae and percentage of each larval stage of the sea cucumber

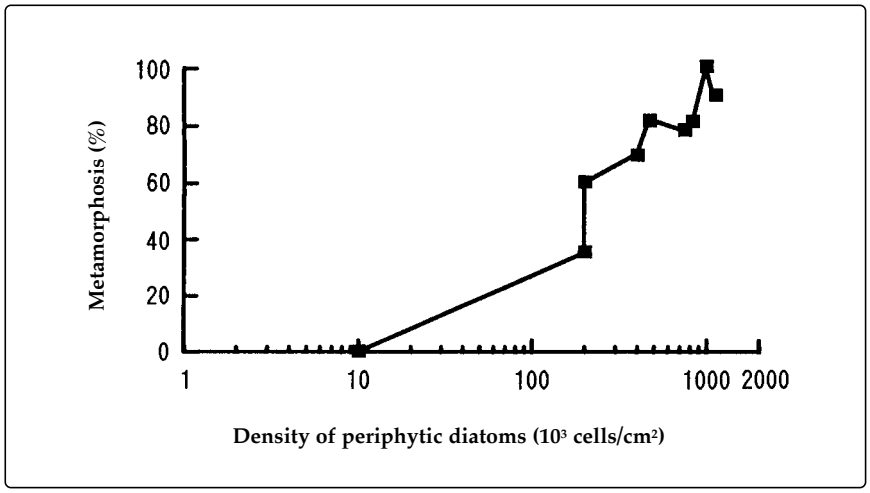

Figure 8

Relationship between the density of diatoms and the metamorphosis ratio

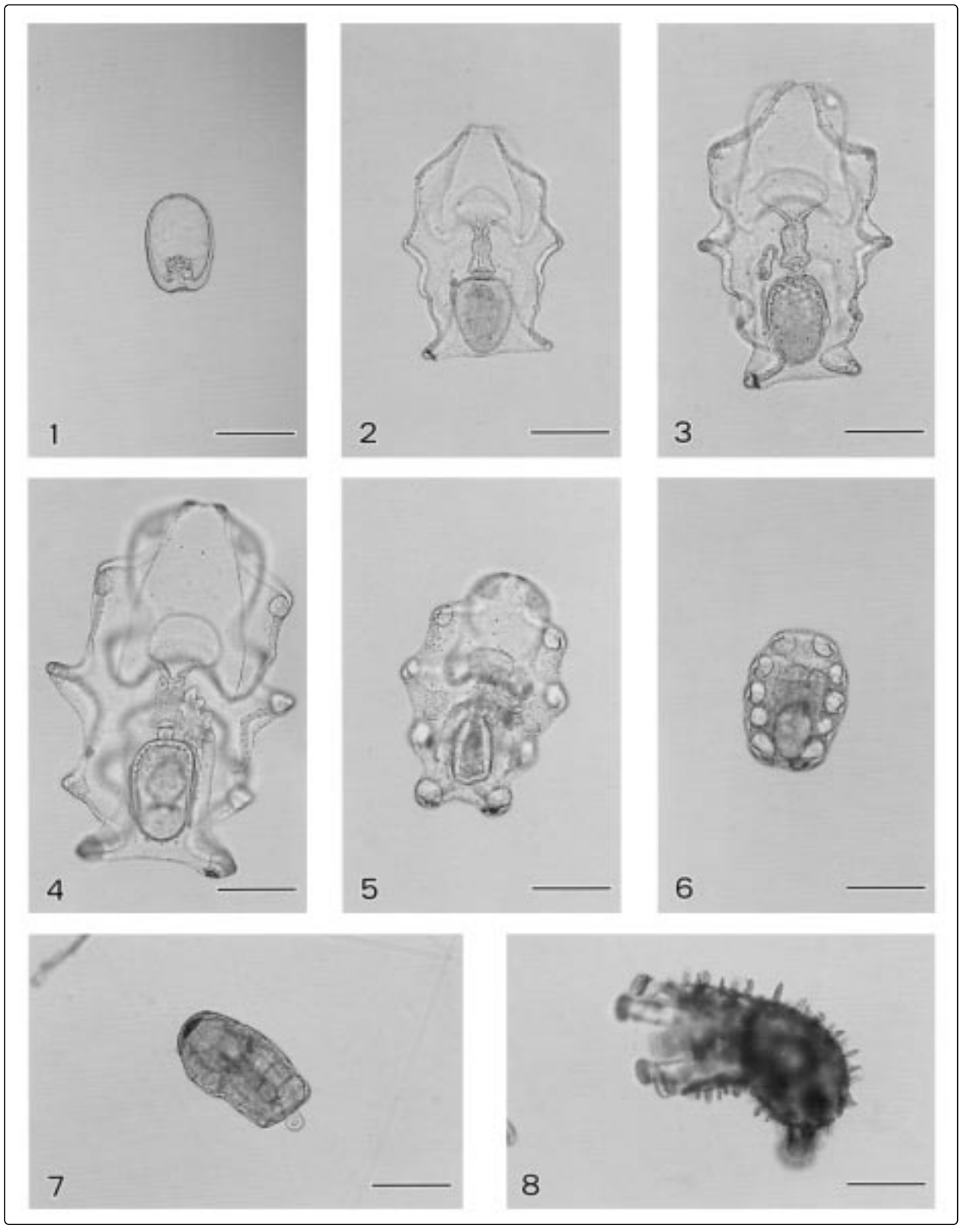

Figure 6

Photographs of the growth stages of larvae of the sea cucumber

1. One-day-old after artificial fertilisation, bar indicates 200 μm
2. Younger auricularia larva (ear-shaped-larva), 3-day-old
3. Auricularia larva, 7-day-old
4. Auricularia larva, 9-day-old, about 900 μm
5. Older auricularia larva, 11-day-old
6. Doliolaria larva (barrel-shaped-larva), 14-day-old
7. Pentacutula larva (five-tentacle larva), 15-day-old; this larva can attach substrata by tentacles
8. Juvenile of the sea cucumber after twenty days from the fertilisation

Rearing of juveniles

After induction of metamorphosis on the plates in April, juveniles feed on periphytic diatoms for about 3 months growing up to 10–20 mm body size in 15 t tanks at 18–26°C (Fig. 9). During this period of juvenile culture, periphytic diatoms are maintained by enrichment with the addition of nutrient salts and by elimination of copepods by the trichlorfonic chemical as mentioned above. It is important to reduce the juvenile density by the addition of other plates to the tank. From July to August, these animals are shipped out to be released directly in the sea. A few of the juveniles are transferred to a diked pond with sandy mud bottom, and they grow up to an average of 80 mm (max. 150 mm) in one year (Fig. 10).

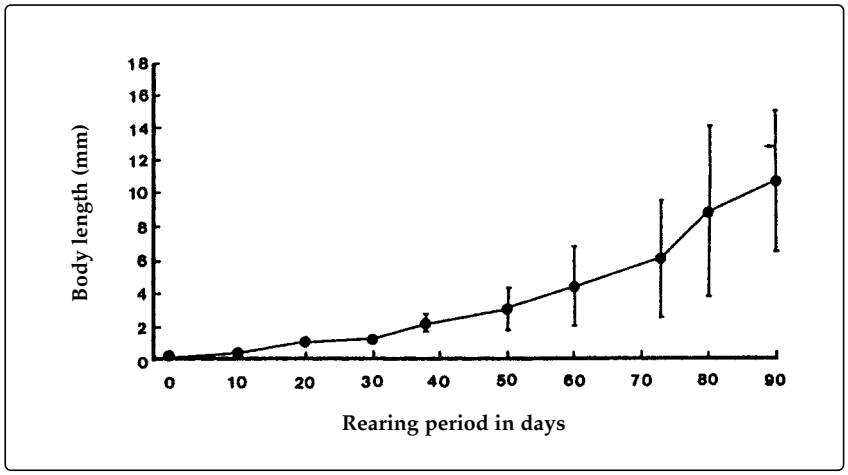

Figure 9

Growth of the juvenile sea cucumber on corrugated plates in the 15t tanks

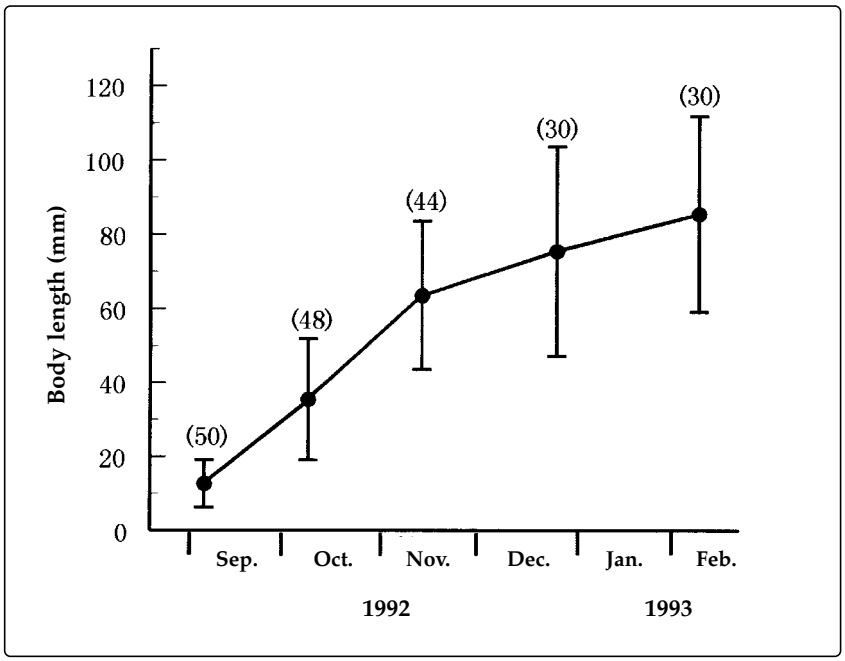

Figure 10

Growth of the sea cucumber in the diked pond

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