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A Study on the Possibilities of Obtaining Larva From Native Flat Oysters (*Ostrea edulis* L.) Living in the Black Sea and Larval Metamorphosis Stage

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Abstract: In this research, the possibilities of obtaining larva from oysters (*Ostrea edulis* L.) living in the Black Sea and their metamorphosis stage were investigated. There were no difficulties in obtaining larvae. The results of this investigation indicate that flat oyster (*Ostrea edulis* L.) culture is possible in Black Sea conditions.

Key Words: Oyster, *Ostrea edulis* L., culture, larval development, Black Sea.

Karadeniz'de Bulunan İstiridyelerden (*Ostrea edulis* L.) Yavru Elde Etme Olanakları ve Larval Metamorfoz Süreci Üzerine Bir Araştırma

Özet: Bu çalışmada, Karadeniz'de bulunan istiridyelerden (*Ostrea edulis* L.) yavru alma olanakları ve elde edilen larvaların metamorfoz sürecinin saptanması amaçlanmıştır. İstiridye larvalarının elde edilmesinde bir güçlük yaşanmamıştır. Sonuç olarak, Karadeniz koşullarında istiridye (*Ostrea edulis* L.) yetiştiriciliğinin mümkün olduğu belirlenmiştir.

Anahtar Sözcükler: İstiridye, *Ostrea edulis* L., Yetiştiricilik, Larval gelişim, Karadeniz.

Introduction

The oyster is one of the most commonly cultured demersal marine organisms in the world. It is found in almost all sea areas except the Arctic and Antarctic regions. The flat oyster, *Ostrea edulis* L., is the only oyster species which is found on the coasts of Turkey (1-7).

Today, the annual worldwide oyster harvest, which consists of catching and culturing, is around one million tons, including approximately 200 oyster species. The figure is around 1500 tons, all of which has been obtained by catching and there are no activities regarding oyster culture in Turkey (3, 8, 9).

Ostrea edulis L., whose lower (left) valve is convex and upper (right) valve is flat, lives on firm ground in shallow coastal waters down to a depth of 20 m. The oyster, which is a prominent mollusc in the intertidal zone, like other bivalves, can reach other sea areas in its larval stage. The length of the oyster is around 10-12 cm. *Ostrea edulis*, which is widely distributed in northwestern Europe, can be found in estuaries, and tolerates salinities of up to 23‰. It often occurs in large beds on muddy-sand, muddy-gravel and rocks. Mainly, their prey is made up of phytoplankton such as *Isochrysis galbana*, *Dunaliella tertiolecta* and *Tetraselmis suecica* (2, 10-15).

The reproduction biology of *Ostrea edulis* is very interesting. *Ostrea edulis*, which is a hermaphrodite organism, changes sex, according to the temperature and feeding conditions. After 8-10 months of growth and water temperatures of at least 16°C, *Ostrea edulis* matures, becoming male and releasing sperm. Then it becomes female and begins egg production. The sex variation of *Ostrea edulis*, which cannot be fertilized by itself, continues throughout its life span. Eggs, which themselves are attached to the mantle cavity of the female, are fertilized by sperm released by the male brought in with the feeding current of the female. They subsequently develop into fully shelled larvae before being discharged into the sea, where they undergo the rest of their development. The most noticeable structure of the larva is the velum or swimming organ, a circular lobe of tissue bearing a ring of cilia by means of which the larva both swims and feeds. Also noteworthy are an extensible ciliated foot and black eyespot (Fig. 1) (12, 14).

Phytoplankton, which forms the food of the larva, is caught by cilia, thrown against the base of velum and carried to the mouth. The larva alternates between periods of swimming fairly rapidly upwards and slowly sinking through the water column. If it is disturbed, the

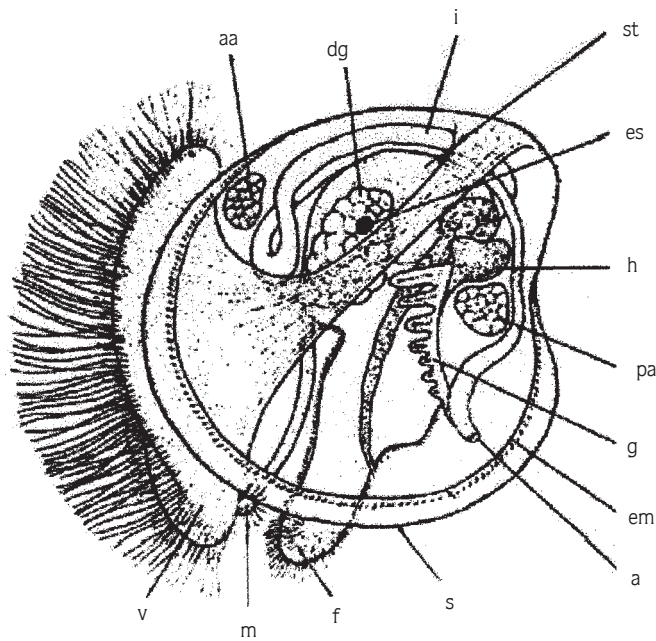
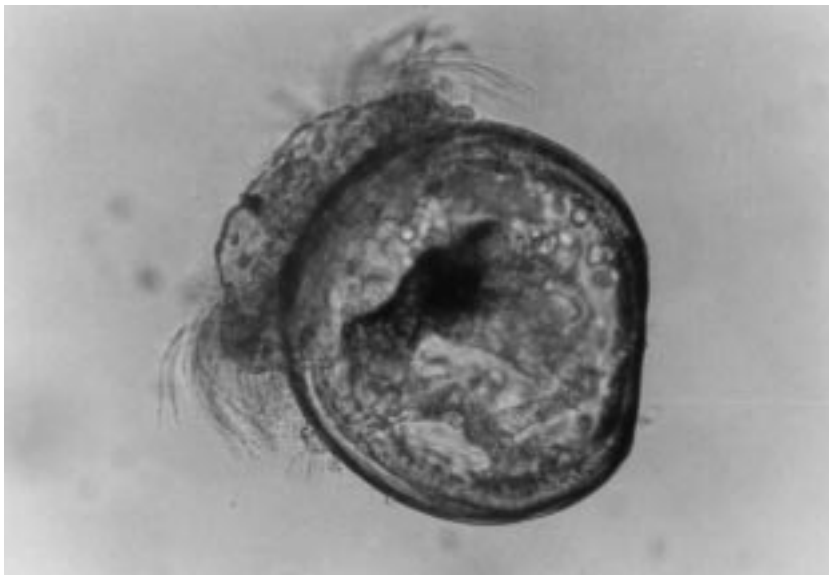


Figure 1. Prominent anatomical features of an oyster larva; aa: anterior adductor muscle, dg: digestive gland, i: intestine, st: stomach, es: eyespot, h: heart, pa: posterior adductor muscle, g: gill, em: edge of the mantle, a: anus, s: shell, f: foot, m: mouth, v: velum (Photographs: original (x200); Fig.: to (15)).



velum can be completely retracted and the two valves of the shell are closed. The closed larva is so efficiently protected that it can be briefly rinsed with toxic chemicals, such as a dilute solution of chlorin, without being harmed (14).

The length of larvae is usually 170-190 μm at liberation, and this increases to 290-300 μm , and exceptionally to 360 μm , at the end of larval development. The most critical stage in the larval development of *Ostrea edulis* is metamorphosis, which is

a transition from pelagic to benthic state. The duration of the free-swimming period is about 10 days; variations in this figure will occur due to differences in temperature and food supply. As the larva approaches metamorphosis, the structure becomes more complex and the shell shape is characterized according to the species. Metamorphosis is a critical stage in the life span, since the ability to move is lost, and the intertidal organs change to suit a sedentary existence. When a satisfactory position is found, the larva settles on its left valve and lives in the same position the rest of its life (2, 5, 10, 12-14).

The morphogenic events related to mechanism of feeding during transition from metamorphosing planktonic larva to benthic adult are complex, but are an important subject of discussion in energy storage (they are responsible for the larva's inability to feed during metamorphosis). The induction of metamorphosis in the larva is followed by the rapid disintegration of the velum, which limits locomotion and feeding. Two or three primordial gill lamellae may be present, which aid respiration but appear not to be functional with respect to retention of food particles. Upon settlement, the velum is completely resorbed, and the poorly defined feeding currents of the gill become organised over a period of 2-8 days. During this transition, energy requirements are met by using lipid reserves stored prior to metamorphosis. It has also been reported that the larval survival rate is correlated with the feeding conditions of the broodstock before spawning season. Metamorphosis is complete at the end of this stage, and *Ostrea edulis* goes on living in this form (14, 16-18).

The objectives of this study were to determine the possibilities of obtaining larvae and to investigate the larval metamorphosis stage.

Materials and Methods

A total of 20 broodstock flat oysters were collected from natural populations in Sinop Bay, Karakum locality, on the north coast of Turkey during the months of June, July and September 1996 (Fig. 2). The collection depth was 15 m. They were maintained in special oyster bags which were lowered to a depth of 5 m from the surface at the time of the trial, May 1997. At the beginning of the trial, they were placed in an aquarium of 80x40x40 cm and were supplied with unfiltered seawater of 18‰ salinity (Fig. 3). They were conditioned to spawn at a constant water temperature of 21°C. Broodstocks were left to spawn larvae naturally. At release, larvae were collected and placed in an other aquarium under the same conditions. Throughout the trial, metamorphosing larvae were observed in the larval aquarium. The seawater of the aquariums was replaced with fresh seawater twice a week so that broodstocks and larvae could feed on the phytoplankton in the fresh seawater. The lengths and widths of larvae were measured (according to the longest distance along the anterior-posterior line of the shell (length) and to the distance from the tip of the umbo to the ventral margin of the shell (width), respectively) under a microscope and photographed (x200, Nikon Lobophop-2A AFX-BX microphotograph apparatus) (19).

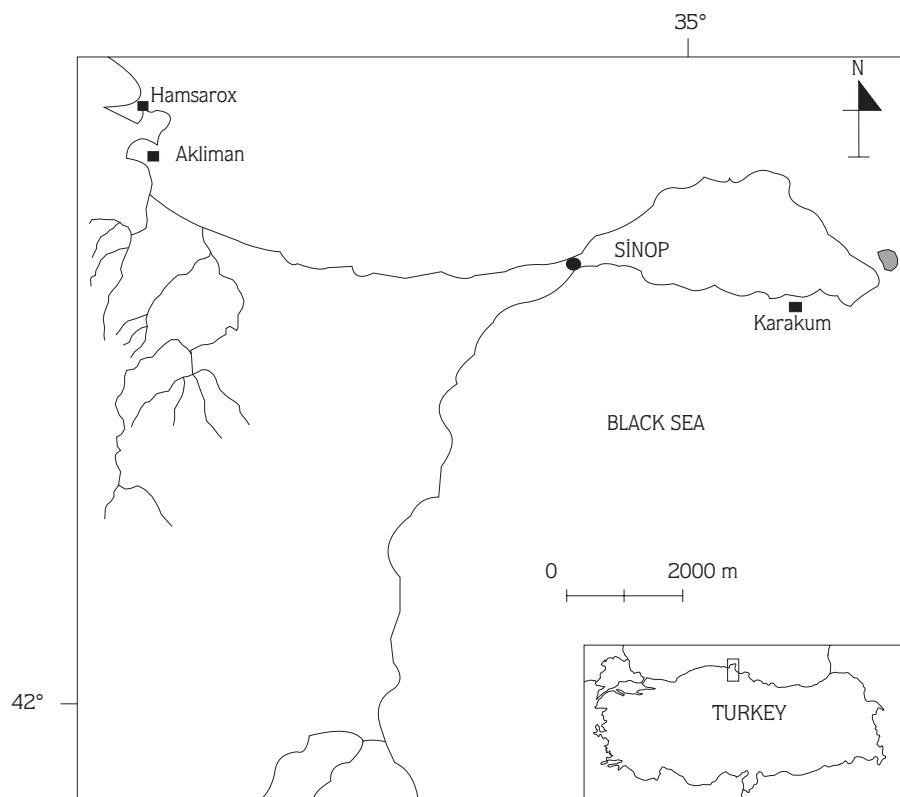


Figure 2. The collecting area of the broodstock oysters.

Results

The moment of liberation of an oyster larva was observed and is shown in Figure 4.

When the larva had been liberated, it could move using the velum. Although the larva shown in Figure 4 has just been liberated, the velum is not visible because when the larva was disturbed, the velum was completely retracted and the two valves of the shell were closed.

An oyster larva able to move in water with its velum and prominent anatomical features is shown in Figures 1

and 5. The internal organs of the larva are visible since the shell is transparent, as shown in Figure 5.

An oyster larva at the beginning of the metamorphosis stage which has entered benthic state, losing its movement ability, and which cannot feed because its velum is absorbed, is shown in Figure 6.

An oyster larva which has absorbed its velum and had partially completed metamorphosis is shown in Figure 7.

An oyster larva which has completed metamorphosis is shown in Figure 8.



Figure 3. Broodstock oysters.

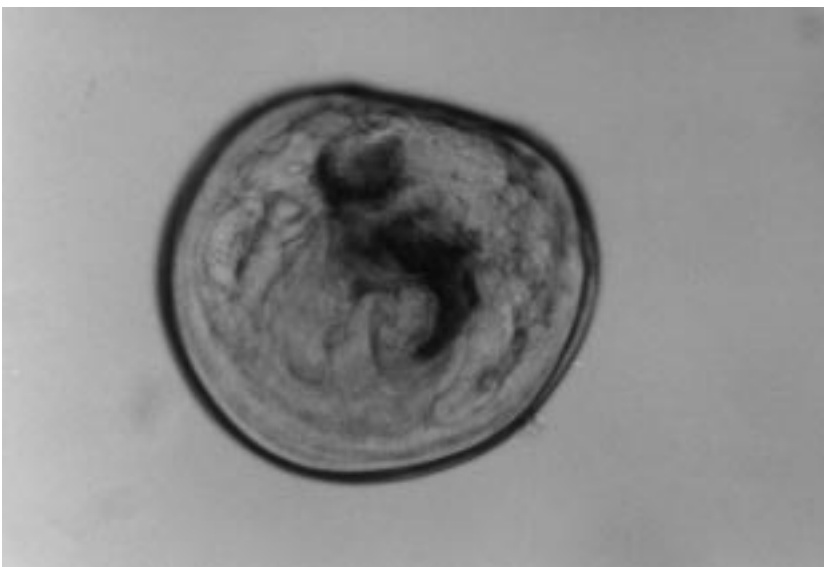


Figure 4. An oyster larva which has just been liberated (original, x200).

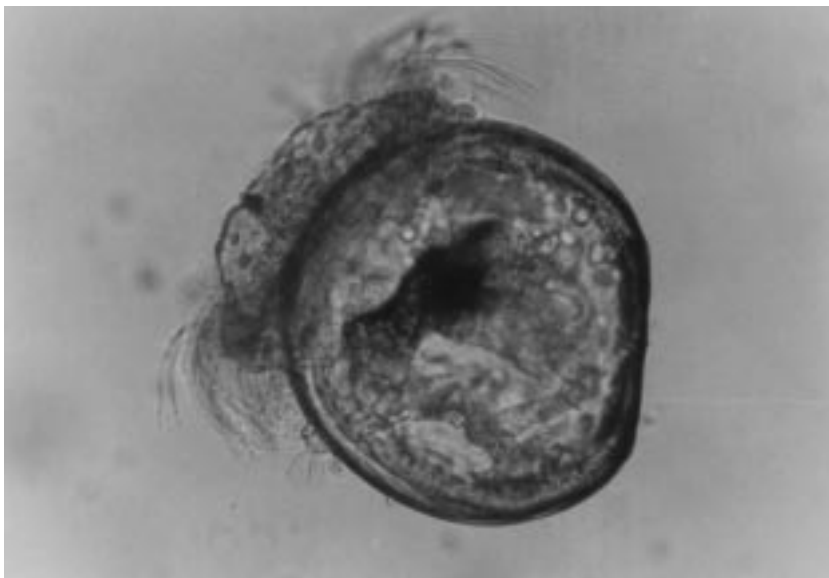


Figure 5. An oyster larva which moves by using its velum (original, x200).

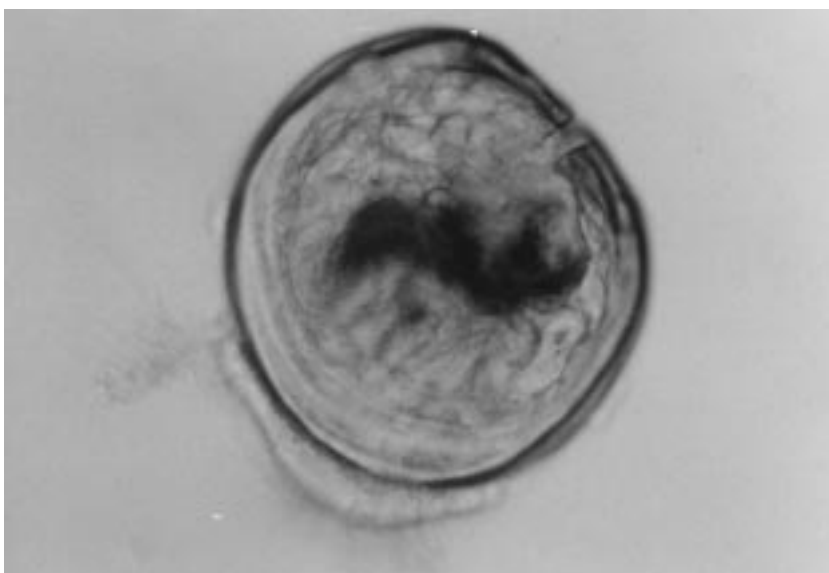


Figure 6. An oyster larva which has settled at the bottom (original, x200).

Variation in the length and width of the larvae during their development are shown in Figure 9.

Discussion

In the present study, the possibilities of obtaining larva from native flat oysters (*Ostrea edulis* L.) living in the Black Sea were investigated, and the larval metamorphosis stage was studied.

As there were no difficulties in obtaining larvae, no breeding methods were used.

The larvae had a lower growth rate than that reported in the literature (14, 16, 18) because of inadequate feeding conditions. It is thus clear that larvae must be fed appropriately with phytoplankton qualitatively and quantitatively so as to allow an optimum larval growth rate.

When the larvae were liberated, their mean length and width were 155.2 x 129.9 μm , whereas after 12 days, they were 174.6 x 167.3 μm . During this stage, proportional length and width gains were 12.5 % and 28.8 %, respectively. According to Figure 9, it was

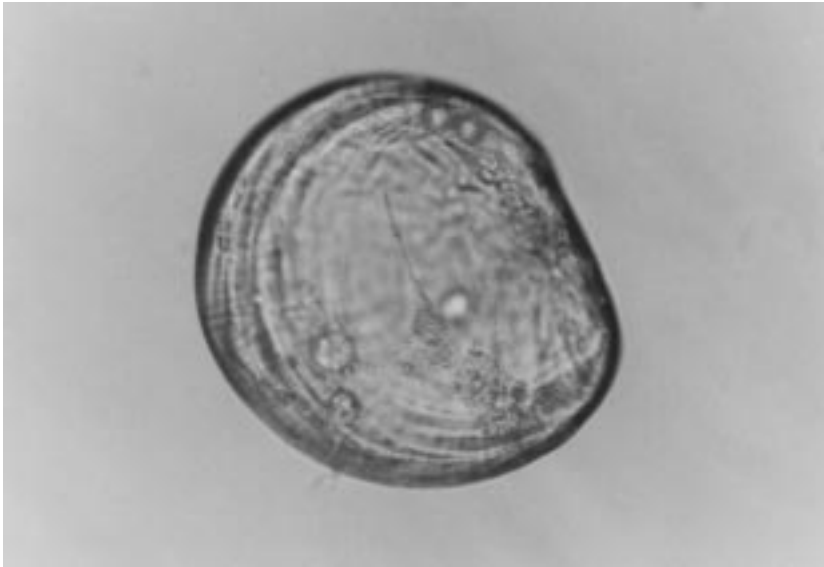


Figure 7. An oyster larva which has absorbed its velum (original, x200).

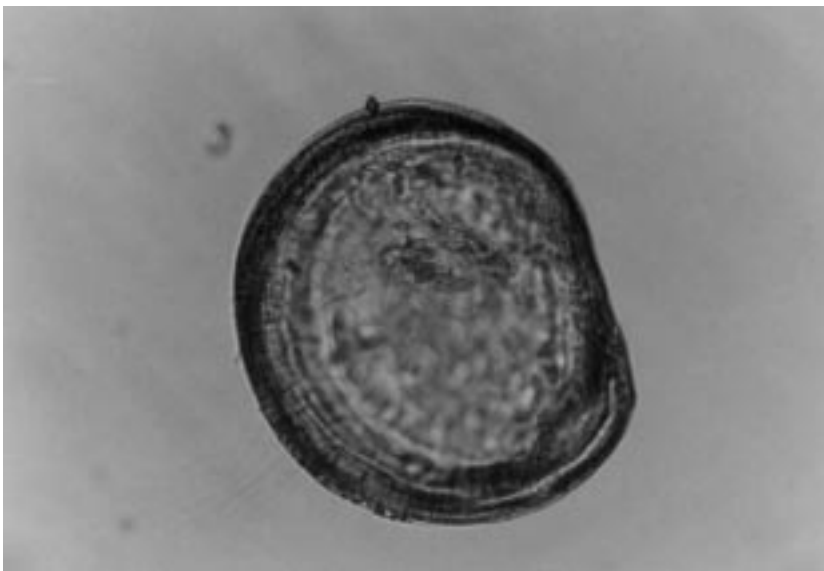


Figure 8. An oyster larva which has completed metamorphosis (original, x200).

observed that the larvae were unable to feed during metamorphosis because of the disintegration of the velum, which enables larvae to move and feed (day 4-7). In this stage, the energy requirements of the larvae were met by lipid reserves stored prior to metamorphosis. When the metamorphosis stage was complete (day 10), the larval growth rate increased (16-18).

Only 20 adult oysters were collected in 15 diving operations because the natural oyster population of the Black Sea is low. In addition, there are dangers which

threaten oyster stocks in the Black Sea. The most serious threat to the oyster stocks in the Black Sea is the oyster drill snail, *Rapana venosa*. As a mass bottom-living carnivore, the snail preys on oysters, mussels and other bivalves and can cause extensive damage to oyster beds, feeding on the sedentary oysters (20). Furthermore, it has also been reported that the local hydrography with low salinity, temperature and unknown events results in low and variable recruitment of juvenile oysters on natural oyster beds, although adult *Ostrea edulis* survives and grows well (21).

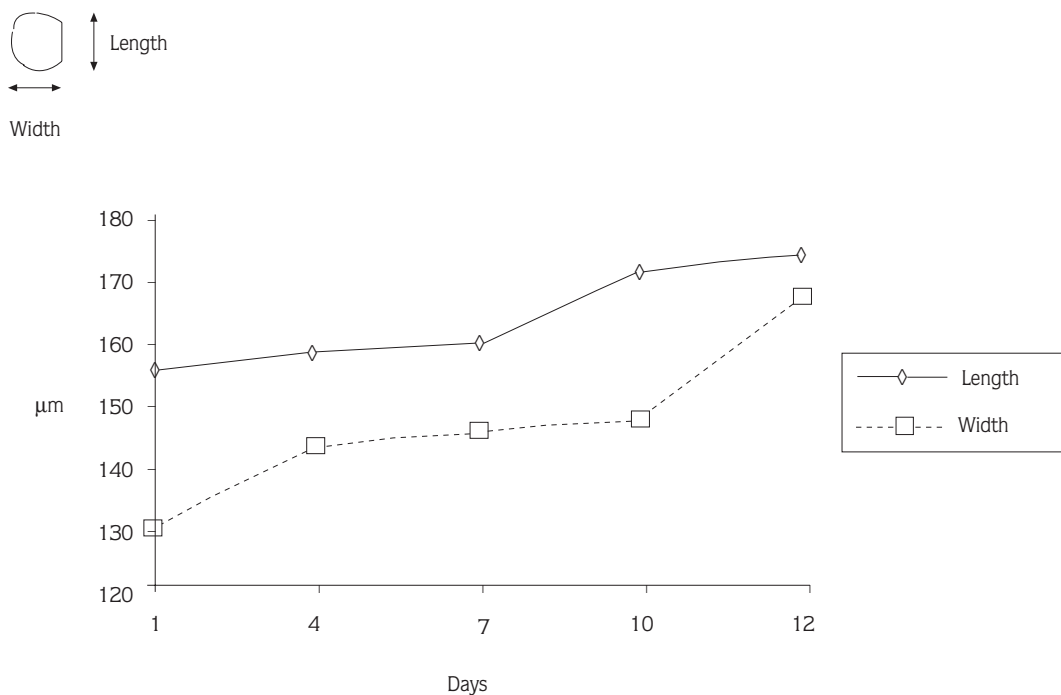


Figure 9. Variations in the length and width of the larvae during development.

According to the results of this study, flat oyster (*Ostrea edulis* L.) culture is possible in Black Sea conditions. The necessity of supporting and developing

oyster culture is based on the fact that oyster culture has commercial importance and that it supports limited natural stocks in the Black Sea region.

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