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Citation	北海道大學水産學部研究彙報, 41(1), 13-17
Issue Date	1990-02
Doc URL	<a href="http://hdl.handle.net/2115/24045">http://hdl.handle.net/2115/24045</a>
Type	bulletin (article)
File Information	41(1)_P13-17.pdf



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## Embryonic Development and Larvae of Long Shanny, *Stichaeus grigorjewi* Herzenstein

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### Abstract

Although long shanny, *Stichaeus grigorjewi*, is a common blennioid fish found in Hokkaido waters and is available commercially, no research on its life history has been conducted. The present study describes the embryonic and larval development of the species observed in a rearing experiment.

Eggs and larvae were reared under an average water temperature of 6.1°C and 8.5°C respectively. Eggs were spherical, demersal and adhesive, measuring 1.25 mm in average diameter. The fertilized eggs underwent the typical discoidal cleavage as in other teleost egg. On the 33rd day after insemination, hatching occurred and continued for a week. The hatching rate was as high as 86 percent. The average total body length of the newly hatched larvae was 7.99 mm with 60 to 63 myotomes. Larval mortality increased from the 8th day after hatching and all larvae starved to death by the 13th day, even though nauplii of *Artemia salina* were supplied amply. The average survival time of the larvae was 8.7 days but the growth in total length was only 0.34 mm.

### Introduction

Long shanny is a common blennioid fish distributed in the coastal waters of Hokkaido. Although the annual commercial landing of the species in Funka Bay, southern Hokkaido, rapidly decreased from 2440 metric tons in 1965 to 254 in 1973 (data of ten fishery cooperatives from Sawara to Muroran), this fish has been used as good quality material for paste products. However, no research on its life history has been conducted.

In 1970, the author succeeded in the artificial fertilization and rearing of eggs and larvae of the species. The present paper describes observations on embryonic and larval development.

### Material and Procedure

Ripe adults, a female 457 mm BL and a male 529 mm BL, were obtained from a long line catch on May 14, 1970 in the coastal waters off Yakumo in the western area of Funka Bay. Artificial fertilization was carried out by the ordinary wet method. Water temperature at insemination was about 6°C. About 4500 fertilized eggs were kept in one glass vessel (three dimensions, 22×29×29 cm) filled with 10 liters of sea water. During incubation a vigorous stream of air was bubbled continuously in the vessel to supply the sea water with oxygen and to stir the water, and the water was changed every 2 or 3 days. When hatching occurred, the air

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bubbling was weakened and the water was changed frequently every day. During the peak of the hatching period, each group of 300 newly hatched larvae was transferred to five glass vessels (dimensions of vessel and water volume were the same as in the incubation experiment) without aeration and change of water. In this experiment, eggs and larvae were reared at an average water temperature of 6.1°C (range, 5.2~6.8°C) and 8.5°C (range, 7.5~9.4°C) respectively, taking into consideration the natural conditions of the coastal waters off Yakumo. The specific gravity of sea water varied within the range of 23.6 to 24.8 (15°C) throughout the experiment. As the mouth of larvae was functional and the vent opened at the hatching stage, nauplii of *Artemia salina* were supplied as food from the day after hatching and thereafter. Larvae in one of the five vessels were reared without food for a starvation experiment to assess the survival time only with yolk. The measurements of larvae were carried out in fresh condition by anesthetizing with a 5 mg percent solution of MS 222-Sandoz.

### Eggs and Embryonic Development

The eggs of long shanny were spherical in shape, measuring 1.25 mm in average diameter (range, 1.22 to 1.29 mm) and contained a single, rather large oil globule of about 0.41 mm in diameter, but some eggs had a few small oil globules in addition to the large one. The yolk was heterogeneous: the dark cloudy material formed an obscure outlined mass in it. No conspicuous structure was observed on the surface of the egg chorion. Numerous foamy structures of about 5~30  $\mu$ m in size were scattered over the surface of the yolk. The eggs were light milky white in color and were translucent. The demersal eggs, adhered to each other, and to a slight degree to the glass vessel.

Five hours after insemination, the chorion fully swelled and a wide perivitelline space was formed, therefore, the egg diameter increased to 1.59~1.72 mm, that is about 30 percent larger than the original size. The chorion contact points between the eggs protruded considerably, almost all the foamy structures observed on the yolk surface disappeared on site without migrating, and the blastodisc reached its full development.

Seven hours and twenty minutes after insemination, the two cell stage was completed (Fig. 1-A), and subsequent cleavages followed at about two hours fifteen minutes intervals, and by 27 hours after this stage the eggs developed into the prosperous morula stage.

At 97 hours after insemination, the epibolic growth of the blastoderm attained the equatorial plane of the yolk, but the embryonic shield was not yet recognized.

The coloration of the egg chorion became increasingly fulvescent as the development progressed, therefore, embryonic development became difficult to observe in detail. The following observations were conducted through the prominent portion of the chorion where the color was comparatively lighter.

At 160 hours, the blastopore closed completely, the embryo encircled about a half of the circumference around the yolk sphere, and the optic vesicles became outlined in the cephalic region of the embryo (Fig. 1-B).

At 191 hours, optic vesicles were well defined, Kupffer's vesicle fully developed near the posterior end of the embryo, and the lenses and auditory vesicles were

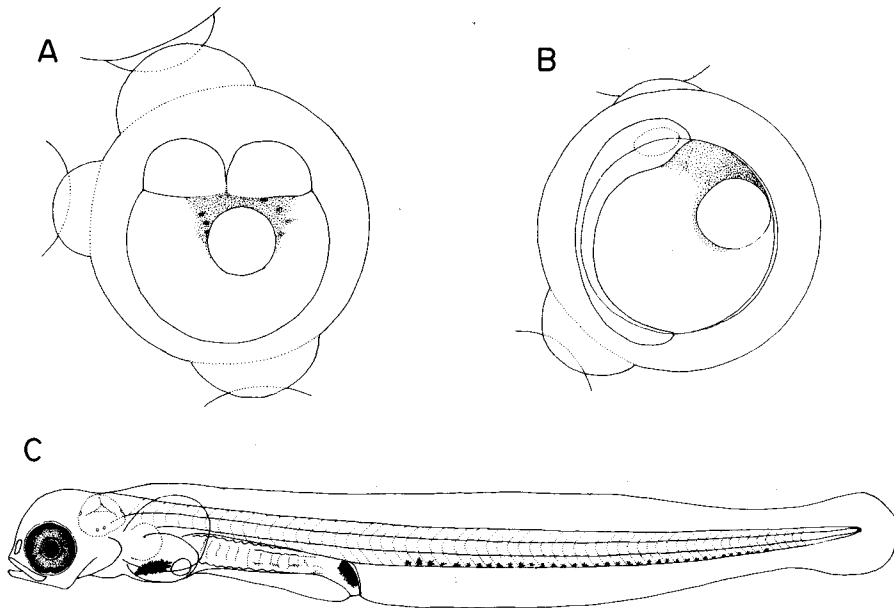


Fig. 1. Drawings of eggs and larvae in *Stichaeus grigorjewi* : A, 2-cell stage, 7 hours and twenty minutes after insemination, top view ; B, Embryo in nearly half-circle, 160 hours, lateral view ; C, Newly hatched larva, 7.96 mm TL, lateral view.

clearly established.

At 263 hours, the Kupffer's vesicle had completely disappeared, the tail started to grow free from the yolk sac and the heart appeared as a slight ventral bulge below the nape.

At about 320 hours, the heart was pulsating slowly and regularly, the embryo showed faint movements by twitching its tail and faint pigmentation appeared on the margin of the optic vesicles.

At 385 hours, the tip of the tail reached the snout encircling the yolk sac as a consequence of the growth of the embryo, the yolk sac decreased in size and a narrow fin fold appeared on the tail.

At 625 hours, the stellate melanophores appeared in one row along the ventral margin of the tail, and other melanophores were apparent on the posterior part of the intestine and on the antero-ventral part of the body cavity. By this stage, the eyes became conspicuously black except for the pupils.

Seven hundred and ninety-seven hours after insemination, at the 33rd day, hatching occurred and continued for a week. The incubation time required to 50 percent hatched was about 36 days and the temperature summation was 220°C. The hatching rate was as high as 86 percent.

#### Larvae

The average total length of the newly hatched larvae was 7.99 mm (range, 7.7 to 8.4 mm). The oval yolk sac measured about 0.72 mm along its horizontal axis,

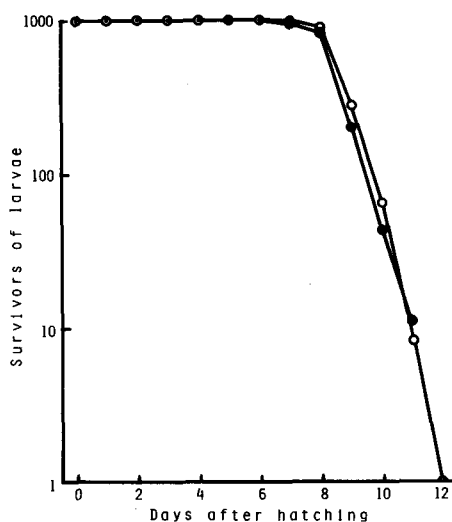


Fig. 2. Survival curves in larval stage, original numbers being 1,000. Open circles and closed circles represent fed group and non-fed group respectively.

and it contained an oil globule in it. The number of myotomes was about 17~18+43~45=60~63 (the number of vertebrae excluding urostyle was 59~60 by the author's observation on the adult specimens collected off the coast of Shikabe, Funka Bay).

The body of the larvae was elongated in form and the vent opening was located at a position about two-fifth of the total body length from the tip of the snout (Fig. 1-C). Eyes and auditory vesicles were large and averaged 454  $\mu\text{m}$  and 431  $\mu\text{m}$  in horizontal diameter respectively. The mouth of the larvae was functional. A transparent fin fold originated on the nape and extended to the postero-ventral position of the yolk sac surrounding the tail, with the discontinuity at the vent. The characteristic pigmentations appeared on the body; a pair of large dendritic melanophores were apparent on the antero-ventral part of the body cavity forming an inverted V-shape pattern in ventral view, a large dendritic melanophore on the rectum part of intestine, and 34 to 36 small stellate or dendritic melanophores lay in one row on the ventral margin of the tail. Larvae were able to swim about actively immediately after hatching and exhibited the phototactic behavior.

Nine days after hatching, the total length of the larvae measured 8.33 mm on the average (range, 8.1 to 8.5 mm). A small yolk sac containing a minute oil globule was found at the anterior part of the body cavity. The body form, the number of myotomes and pigmentation pattern were essentially the same as in the newly hatched larvae.

In the fed experimental group, the larval mortality rate began to increase from the 8th day after hatching and all larvae starved to death by the 13th day without taking food. Therefore, the survival curves for larvae in the fed group showed a similar pattern to the non-fed group (Fig. 2).

### Discussion

Eggs of long shanny are demersal and adhesive. The fertilized eggs undergo a typical discoidal cleavage as in other teleost egg. However, peculiarities in the development the author observed are the swelling of the egg chorion at the contact points between eggs and the change in color of the egg chorion during the course of development. Prominent adhesive of egg chorion was reported in a blennioid fish, *Dictyosoma burgeri* by Shiogaki and Dotsu (1972). This probably contributes to egg survival by welling the water flow through the gap between the eggs, but the ecological significance of the change in color of the chorion is unknown.

A high rate of mortality occurred during the yolk absorption and none of the larvae survived beyond the yolk-sac stage, even though an ample diet was supplied. This fact suggests that the nauplii of *Artemia salina* is not a suitable food for long shanny larvae in rearing experiments, presumably because they were too large to be eaten. The larvae were able to survive for 8.7 days on the average under a water temperature of about 8.5°C without external nutriments, but the growth increment of total body length during this period was only 0.34 mm.

### References

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