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Artificial Completion of the Japanese Eel, *Anguilla japonica*, Life Cycle: Challenge to Mass Production

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Abstract：Current eel culture depends entirely on glass eels captured from the wild. However, in recent decades, eel populations have declined. Thus, establishment of the technology for producing sustainable supplies of eel seeds is required. To achieve that end, selective breeding and mass production of glass eels is necessary. This year, we were successful in closing the Japanese eel life cycle. However, we have not as yet established techniques for mass production of glass eels because of various technical difficulties. In this paper, we describe the significance of closing the eel life cycle and the challenges that need to be overcome in order to develop a system of glass eel mass production.

Key words：ell culture, glass ell production, ell life cycle, *Anguilla japonica*

The mysterious life cycle of eels has attracted many researchers. For a long time, no one could find eel eggs or larvae in the habitats where the adults were found; such as rivers, ponds, coastal waters. Early in the 20th century, Schmidt (1922) conducted numerous expeditions and discovered that the spawning area for both the European eel (*Anguilla anguilla* [L.]) and American eel (*Anguilla rostrata* [LeSueur]) was located far offshore in the Sargasso Sea region of the Atlantic Ocean. In 1991, nearly 70 years after Schmidt's (1922) discovery, the Philippine Sea of the western North Pacific was determined to be the spawning area of the Japanese eel, *Anguilla japonica* Temminck & Schlegel. Thus, the life cycle of anadromous eels was gradually uncovered (Tsukamoto, 2009).

Populations of *Anguilla japonica, A. anguilla*, and *A. rostrata* have decreased markedly in recent decades due to overfishing, environmental destruction, or other unknown factors (Casselman, 2003; Dekker, 2003; Tatsukawa, 2003). On the other hand, establishment of Japanese eel culture has maintained the availability of eels to consumers at reasonable prices. However, current eel culture depends entirely on glass eels captured from the wild. A decrease in the availability of glass eels and increase in demand for eels in the marketplace will inevitably lead both to increased prices and decreased natural eel stocks. The vicious spiral could be stopped through the development of techniques to mass produce glass eels through aquaculture.

History of Research for Rearing Eel Larvae and Closing of Their Life Cycle

In Japan, attempts to induce artificial maturation of the Japanese eel started in the 1960s. Yamamoto and Yamauchi (1974) were the first to successfully obtain fertilized eggs and larvae through hormone treatments, and after two-week rearing period the preleptocephalus larvae reached 7mm TL (Yamauchi *et al.*, 1976). Thereafter, other researchers succeeded in obtaining eel larvae (Satoh, 1979; Wang *et al.*, 1980), but suitable larval feeds were not identified.

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Eel larvae cannot be reared with feeds, such as rotifers, that are often used in conjunction with other finfishes. Development of a suitable diet was difficult since it began in the absence of ecological information on larval eels. Even now their food habits in nature have yet to be determined.

Tanaka *et al.* (2001) developed a slurry type diet made from shark eggs. The diet has enabled us to rear eel larvae and led to the first successful production of glass eels in captivity (Tanaka *et al.*, 2003; Kagawa *et al.*, 2005). Since then, we have made an effort to develop a captive broodstock of adult eels reared from the egg stage. That has enabled us to complete the task of closing the life cycle of the Japanese eel (Fig. 1), which was achieved it in 2010. Details of the process will be described elsewhere. We have already produced more than 300 glass eels to an age of 240 days after hatching (dah) and grow them to produce the next generation. Before achieving that breakthrough, all eggs were obtained from adults either captured from natural waters or grown from captured glass eels. We determined that rearing eels from the egg in a controlled environment does not impede oogenesis in adult stage. Closure of the life cycle and selection of individuals to be reared as bloodstock provide the opportunity to produce cultured eels of high quality. Selective breeding for faster growth and higher survival rates is thought to be promising at both the larval and adult stages. Of course, improvement in the taste of cultured eels will also be expected. The next challenge will be to find a way to consistently mass produce glass eels.

Issues to Resolve for Mass Production of Glass Eel

Some technical developments has been accomplished by several researchers with respect to mass producing glass eels in captivity. Tsukamoto *et al.* (2009) discovered that cultured eel larvae have greater buoyancies that captured larvae and that their buoyancy showed ontogenetic change. Okamura *et al.* (2009a) reported that an intermediate salinity (50% of sea water salinity) can yield better growth and survival performance in the early life stages of eel larvae. Okamura *et al.* (2009b) developed a new rearing system using a planktonkriesel instead of a typical tank. Owing to these developments, survival rates of larvae have steadily improved. However, mass production of glass eels has not be realized because of the remaining technical difficulties. Statistics compiled by the Japanese government indicate that more than 21,000 tons of cultured eels are produced annually, which is equivalent to more than 10 million eels (the white paper of fisheries, Japan, 2010). Thus, there is a very long way to go before a significant contribution to the annual total can arise from cultured glass eels. The following difficulties associated with mass production must be overcome:

1) New feeds need to be developed,
2) The rearing process must be simplified,

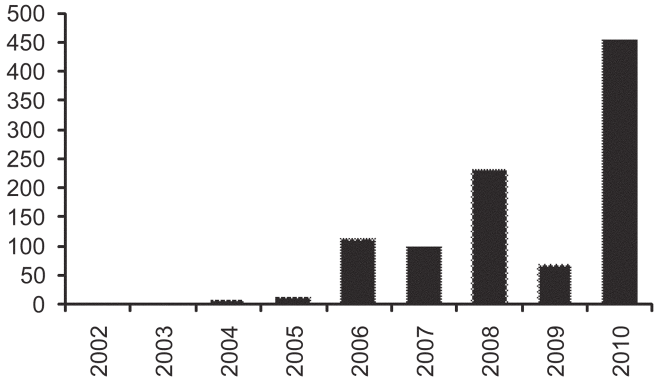

Fig. 1. Number of produced glass eels in Shibushi station. The umber of metamorphosed glass eels between April to the following March of each year (to Nov. in 2010) are represented.

3) The rearing period must be shortened,
4) Diseases must be prevented, and
5) Morphological abnormalities must be prevented.

Today, eel larvae rearing depends entirely on a diet made from shark eggs, but natural shark populations cannot support mass production of glass eel quantitatively. In fact, spiny dogfish, *Squalus acanthias*, from which eel larval diets are formulated, was proposed for inclusion in appendix II of the Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES; Cop15Prop18, proposed by Palau and Sweden). Therefore, we must find effective new dietary formulations. Generally, plant materials are inexpensive and easily obtainable in quantity. But, there are few examples of successful use of plant-based feeds for rearing marine fishes. On the other hand, chicken eggs, as well as eggs from fish and other animals may be acceptable materials because they resemble shark egg in in their composition. Fishmeal, which is used in many fish feeds, including those of adult eels, may have potential. Fishmeal is also available in quantity, though the price is higher than plant protein sources, in general. We have developed an "intestine fullness index" for one of the evaluations of diet effectiveness (Fig.2, Masuda *et al.*, 2010). Using this index, we are searching for new suitable dietary ingredients.

The current larval rearing process is too laborious. Current feeding procedure is as follows (Tanaka *et al.*, 2001): Larvae are kept in the dark except during feeding time, because they swim down and

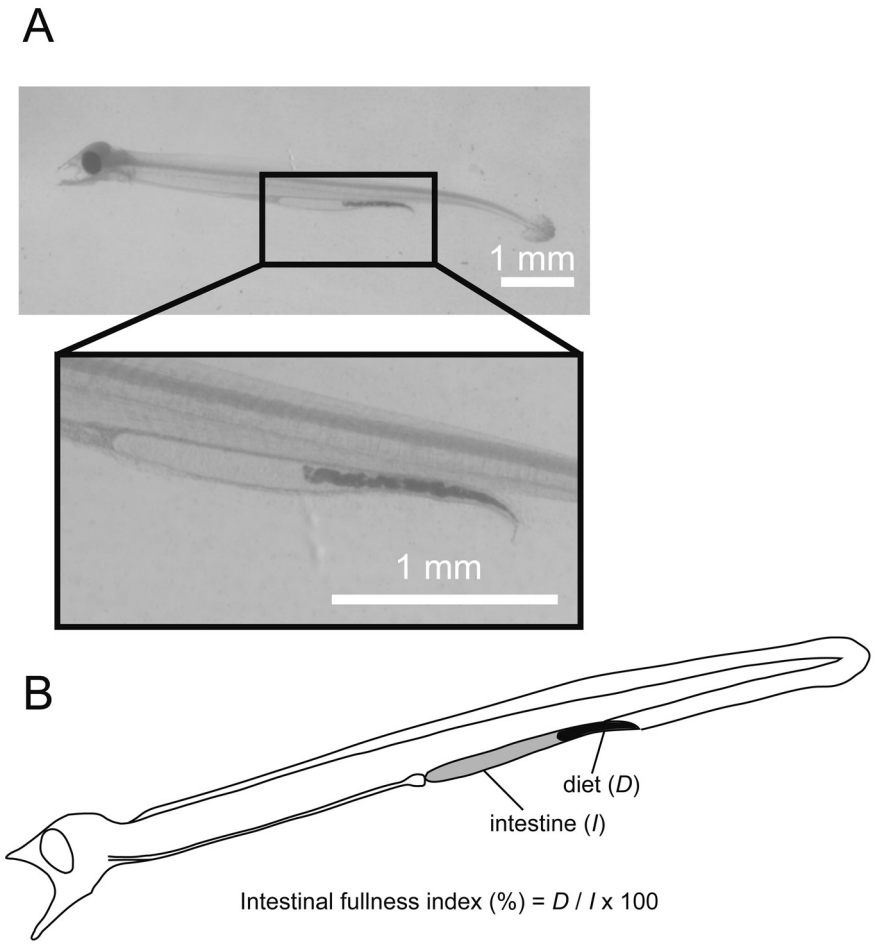

Fig. 2. A Japanese eel larva that had ingested milk (A). Intestine (gray and black) and diet (black) for calculating the intestinal fullness index(B). This figure is modified from Masuda *et al.* 2010.

push persistently on the bottom of tank in the light resulting in jaw breakage. At feeding time, we turn on a light and put the slurry diet on the bottom of tanks (Fig. 3). The larvae swim to the bottom to feed. Before putting in the feed, we must stop the water flow because they are not good swimmers. After allowing the larvae sufficient time to feed, a hose is used to flush any remaining feed from the tank, since normal water flow is not sufficient to do the job. Finally, we turn off the light. Using the described procedure, it is unavoidable for larvae to contact with walls of the tank. The inevitable mortalities can accumulate on the tank bottom, leading to bacterial growth that may induce infectious disease. Thus, we must remove dead larvae by hand. In addition, we must prepare clean tanks and transfer the larvae to them each day.

Developing a feed that is suspended in the water column may be a solution to the problems associated with feeding on the tank bottoms. A colloid-type diet might be suitable (Masuda *et al.,* 2010). When fed milk, a typical colloid-type diet, larvae survived until 26 dah.

The period of time required for rearing eel larvae from hatch to metamorphosis is too long. Until 2009, the rearing time from hatch to completion of metamorphosis had been from 153-754 dah in Shibushi station (Fig. 4). In nature, on the other hand, larvae are thought to metamorphose into glass eels from 100-160 dah (Arai *et al.,* 1997). Thus, the larval period in captivity is not only too long, but also various much more widely than is the case for wild larvae.

The long rearing period complicates the research because of the time involved to complete each experiment. Furthermore, keeping various larval stages at the same time is apt to make the rearing environment unsuitable. In 2010, our new rearing system produced accelerated larval growth, and glass eels were obtained beginning 131 dah (Masuda

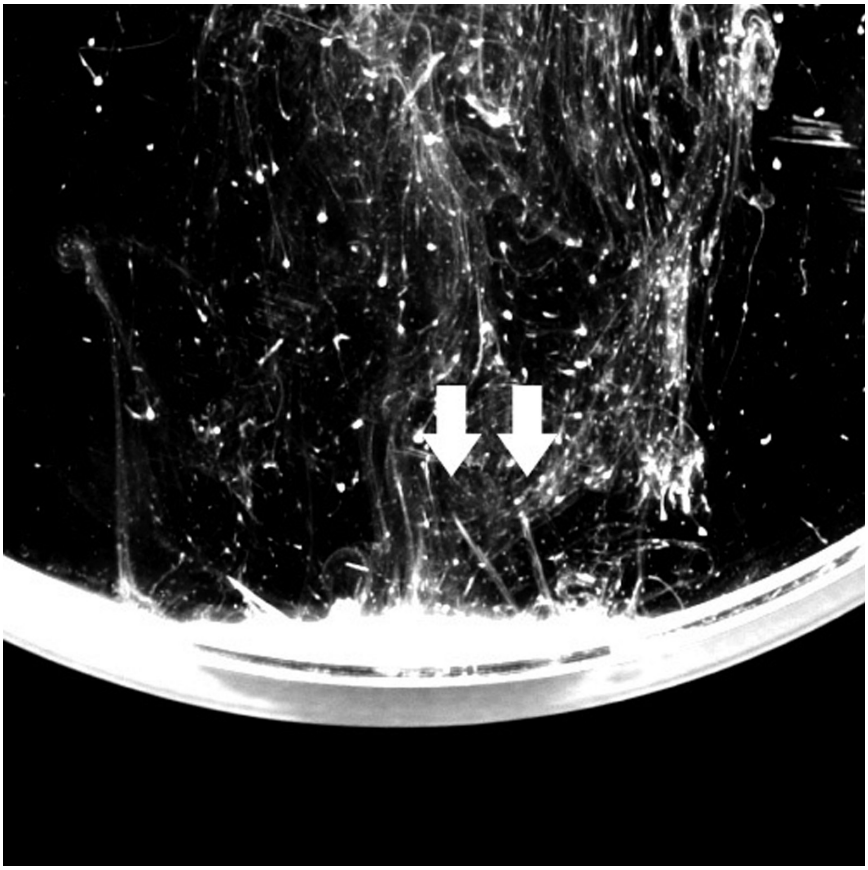

Fig. 3. Eel larvae (6 dah) eating the slurry-type diet. Larvae are pointed out by arrows.

et al., 2011). Details of the experiment will be published in near future. We hope that the results obtain in 2010 will contribute to reducing the costs of labor and energy associated with glass eel production. Moreover, shortening of rearing period can contribute to reducing the risk of diseases, morphological abnormalities, and other deleterious factors.

The danger of diseases associated with larval rearing has recently emerged as an issue. As improvements in rearing technology lead to increases larval survival rates, the relative impact of diseases increases (Fig. 5). From our observations, it seems that a disease kills larvae in a short time; only several hours in some cases; and sometimes leads to mass mortality. However, specific pathogens have not been identified. We have no idea if we' re dealing with a single or multiple pathogens.

Finally, morphological abnormalities are a problem, though they are not always fatal. For example, larvae with broken jaws, the most critical abnormality, can eat the slurry-type diet, grow, and metamorphose into glass eels. However, juveniles with broken jaws cannot eat any type of food and die of starvation. Curved larvae, folded glass eels, and broken necks of glass eels are often seen (Fig. 6). Generally, such larvae and juveniles are hard to feed in spite of having normal jaws. Thus, they are at serious disadvantage with respect to survival and their growth is always poor.

Conclusion

We can produce glass eels from eggs, though the cost is very high. The next steps are to solve the remaining problems hindering mass production of high quality eel larvae. Having found a way to rear eels throughout their life cycle, it might be possible to find other added value, such as "eels for enjoying their style and color." Before that, it will necessary to establish techniques for mass production of glass eels at a reasonable cost and supply them to aquaculturists for growout. Ultimately, the hope is to supply the market with high quality and eels that meet consumer expectations in terms of flavor and cost.

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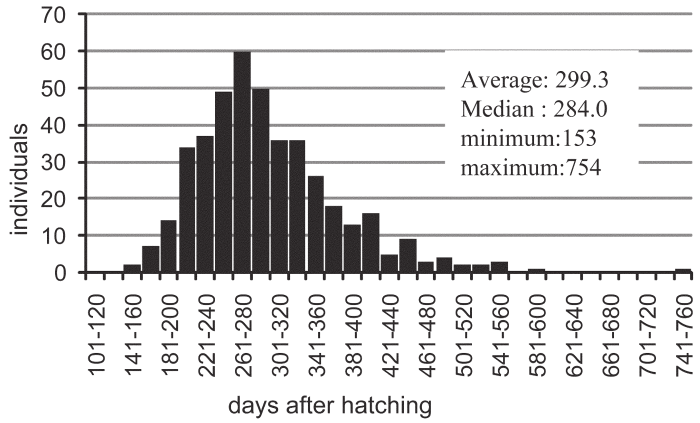

Fig. 4. Days after hatching to complete metamorphosis of eel larvae in Shibushi Laboratory.(Sum of data between April 2007 to July 2010).

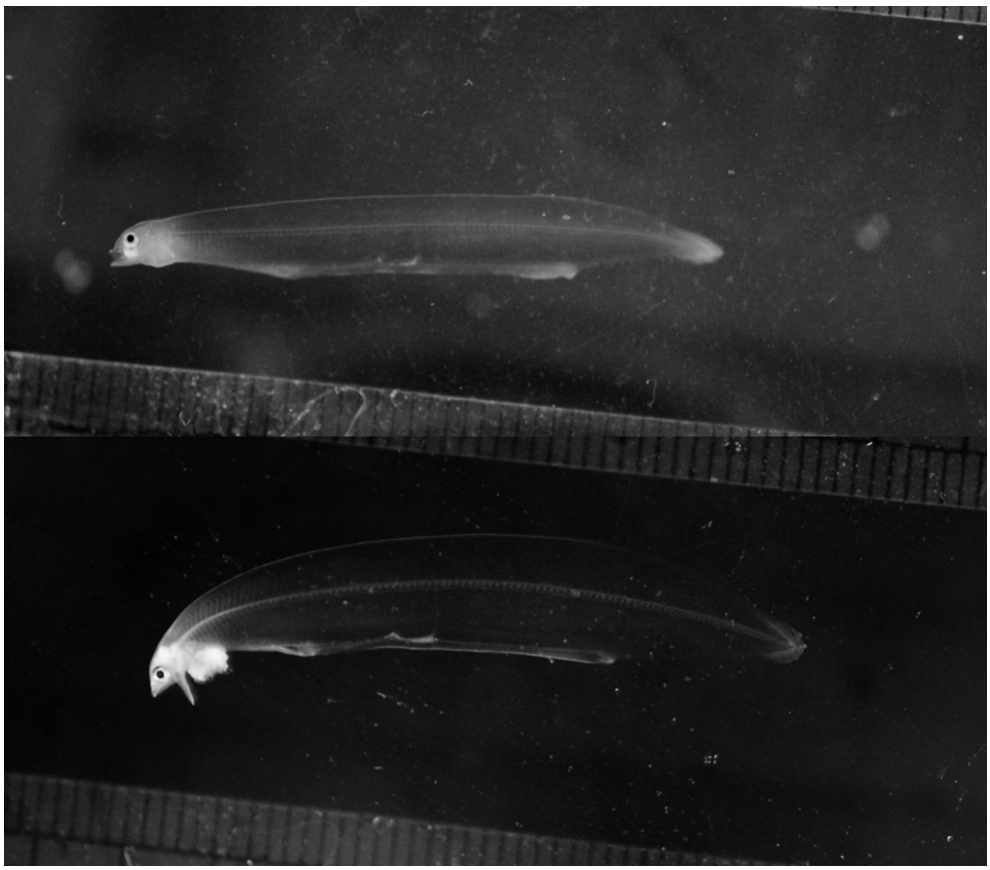

Fig. 5. Larvae with pathology. Upper: Larvae (90 dah) with white head and tail. Lower: Larvae (124 dah) with broken throat.

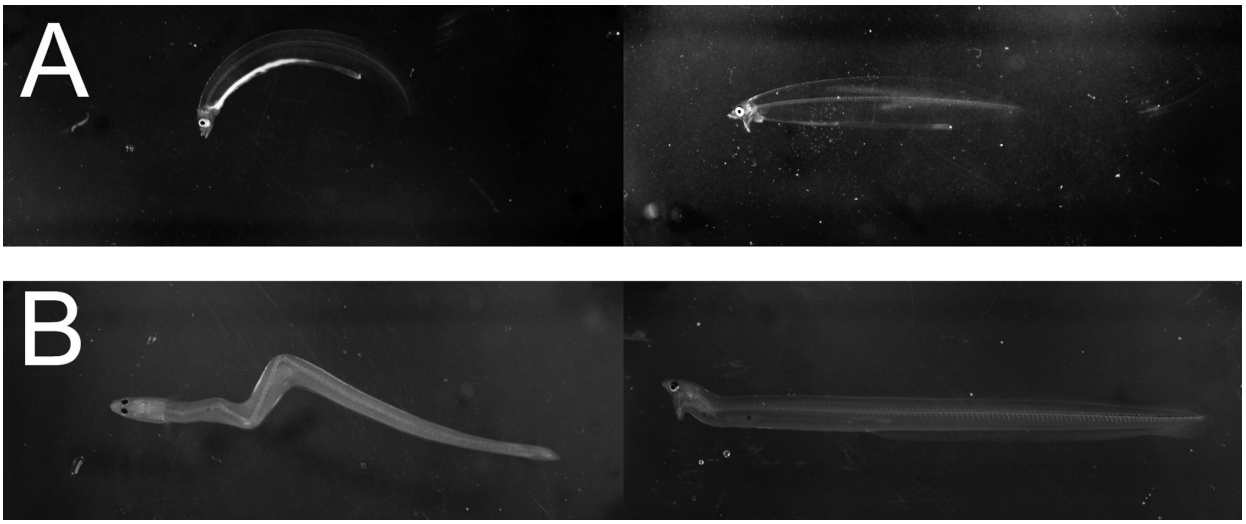

Fig. 6. Larvae with morphological abnormalities. A: Curved larva (86 dah, left) and larvae with broken jaw (80 dah, right). B: folded glass eel (left) and glass eel with broken jaw (right).

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