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Morphological Changes with Growth in *Liza carinata* (Valenciennes) Egg, Larva and Juvenile as Distinguished from Those of *Liza haematocheila* (Schlegel)

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ABSTRAK

Pertukaran secara morfologi di dalam peringkat telur, larva dan juvena Liza carinata *dengan tumbesaran telah dikaji dalam spesimen yang diternak dan yang ditangkap secara semulajadi.* L. carinata *boleh dibezakan daripada* L. haematocheila *(yang mana mereka bertelur serentak dalam masa dan ruang) dalam peringkat telur, larva dan juvena. Telur menetas dalam masa 45-102 jam selepas penetasan di dalam suhu air 15-21°C, sepadan dengan suhu di tempat peneluran. Larva yang baru menetas mempunyai purata jumlah panjang 2.1 mm. Sirik kaudal "anlage" muncul dalam larva yang mempunyai jumlah panjang 3.4 mm dan peringkat juvena dicapai pada jumlah panjang 8.9 mm.* L. haematocheila *telah dibezakan saiznya yang besar dalam peringkat telur dan larva. Melanofor terletak di atas hujung kepala dan hujung ekor dalam* L. carinata *mungkin juga bertindak sebagai tanda yang boleh membezakan dalam peringkat larva. Unjuran yang lebih panjang di dalam barisan melanofor sisi tengah dalam* L. carinata *membezakan mereka dalam peringkat juvena. Sifat bonjolan dorsal dalam* L. carinata *muncul selepas mencapai jumlah panjang 30 mm, yang mana tidak berlaku dalam* L. haematocheila.

ABSTRACT

The morphological changes in the egg, larval and juvenile stages of Liza carinata *with growth were investigated in reared and wild specimens.* L. carinata *could be distinguished from* L. haematocheila *which is likely to spawn concurrently in time and space in the egg, larva, and juvenile stages. The eggs hatch within 45-102 h after fertilization in water of 15-21 °C, corresponding to the temperature in the spawning ground. A newly-hatched larva averages 2.1 mm in total length. The anlage of the caudal fin appears in the larva of around 3.4 mm and the juvenile stage is attained at a length of 8.9 mm.* L. haematocheila *is distinguished by its larger size in the egg and larval stages. The melanophores located on the top of the head and the rear end of the tail in* L. carinata *may also serve as distinguishing marks in the larval stages. The longer extension of the mid-lateral melanophore row in* L. carinata *distinguishes them in the juvenile stage. The characteristic dorsal ridge in* L. carinata *(which does not occur in* L. haematocheila*) appears after a total length of 30 mm is reached.*

Keywords: morphological change, egg, larva, juvenile, melanophore, spawn, distinguishing mark

INTRODUCTION

Liza carinata (Valenciennes) is a small mullet spread extensively from Japan, Korea, China, and the Malay Archipelago to India and the Red Sea (Fowler 1935; Day 1958; Yoshino and Seno 1984). In Japan, the fish are mainly found along the southern coasts of West Kyushu, but are not commercially important and are treated as garbage fish.

In Ariake Sound, this species occurs with another mullet of the same genus, *L. haematocheila,* which is one of the most commercially important fish in the locality. These two species are likely to spawn concurrently in time and space, at least in a part of the Sound. To investigate the life of the two species it is necessary to distinguish them at early stages of development.

The egg and early stages of *L. haematocheila* have been described in detail (Fujita 1979). However, only a brief description of the morphology of postlarval and juvenile stages of *L. carinata* has been given (Kinoshita 1988), and no effort has been made to distinguish the morphological characters between the two species. Eggs and larvae of both species were obtained by artificial fertilization and larval net surveys in Ariake

Sound (32° 27′ - 33° 11′ N ; 130° 06′ - 130° 37′ E) and neighbouring Chijiwa Bay. Morphological changes with growth in *L. carinata* egg, larva and the juvenile stages as distinguished from *L. haematocheila* were investigated.

MATERIALS AND METHODS

Artificial fertilization of eggs of *L. carinata* was carried out in Chijiwa Bay on April 14, 1988 and that of *L. haematocheila* in Ariake Sound on April 23 and May 5 of the same year. The water temperature of both spawning grounds was around 18°C. The eggs were immediately brought back to the Aquaculture Laboratory of Nagasaki Prefectural Institute of Fisheries and put in water tanks within 2 h 10 min after fertilization. The water temperature of the tanks was then adjusted to ensure that no damage was caused to the embryos by an abrupt change of temperature. Thus the eggs were incubated in water temperatures encountered in the natural spawning ground during the spawning season. The eggs were incubated and the larvae reared in tanks under conditions of ambient room temperature (16-21°C) in the Aquaculture Laboratory. The larvae and juveniles were fed with rotifers, *Brachionus plicatilis* and *Artemia salina* nauplii. Morphological characteristics in individuals raised under artificial conditions and anaesthetized with MS 222 were observed and recorded. The specific differences in morphological characteristics were then confirmed by comparing them with those found in the natural specimens collected in larval nets from the seas in April and May of 1987 and 1988.

RESULTS AND DISCUSSION

Eggs and Embryonic Development

L. carinata eggs are buoyant, nonadhesive, spherical in shape and have a narrow perivitelline space and an extraordinarily large oil globule. These characteristics have been described for mugilid eggs in general (Vialli 1937; Martin and Drewry 1978; de Sylva 1984). *L. carinata* eggs are so buoyant in early stages that the eggs are partially exposed to the air. The eggs gradually become heavier towards the hatching period and sink to the bottom. Such changes in egg buoyancy are known to occur in many fish species (Tanaka 1990). In some mugilids, the eggs are known to sink in late developmental stages, as in *Mugil cephalus*, but not always in *M. capito* (Yashouv and Berner-Samsonov 1970).

The stages of embryonic development of *L. carinata* are presented in Table 1 and *Fig. 1*. The embryonic body starts to form when the germ ring has just passed half of the egg diameter, as judged from a lateral view (*Fig. 1D*). The melanophores appear first on the yolk sac around the embryonic body. Just prior to appearance, uncoloured grains appear on the same areas (*Fig. 1F*). Just after their emergence, the melanophores are only sparsely distributed on the yolk sac and are not present on the oil globule. The oil globule becomes thickly covered with the melanophores on its dorsal surface at a later stage *(Fig.1G)*. The xanthophores are located laterally on the embryonic body and the oil globule and scattered widely on the yolk sac.

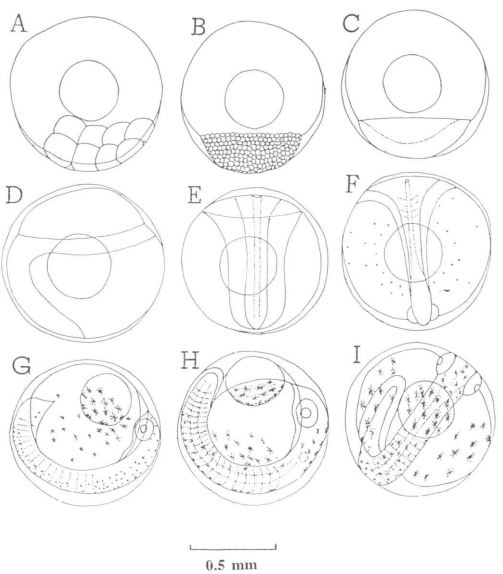

Fig. 1: Embryonic development of L. carinata. *A, eight-cell stage; B, morula stage; C, early gastrula stage; D, formation of embryonic body; E, a little before the closure of the blastopore; F, eye vesicle formation and the appearance of the melanophores; G, tail bud and eye lens formation; H, formation of the tail; I, just before hatching.*

Larvae and Juveniles

The larvae were suspended from mid to lower layers for the first 4 days assuming an upright position with the head down and the tail up. A newly-hatched larva *(Fig. 2A)* averaged 2.1 mm in total length and had 26 (13+13) myomeres. Larvae reach a length of around 2.6 mm in half a day *(Fig. 2B)*, after which growth slows down until the yolk and oil globule are absorbed. The

TABLE 1
Embryonic development of *L. carinata* at various temperatures represented by the times in hours and minutes necessary to reach the respective stages. The eggs were artificially fertilized at 9.50 a.m. on April 11, 1981 and placed in experimental tanks when they were at four-cell stage.

Stage	Incubation temperature (°C)			
	15	17	19	21
Four-cell stage	2:10	2:10	2:10	2:10
Morula stage	5:10	4:40	4:10	3:40
Blastula stage	10:10	8:40	8:10	6:40
Early gastrula stage	18:10	11:40	10:40	9:50
Beginning of embryo formation	26	18	15	14
Closure of blastopore	29	23	18	16
Eye vesicle formation	35	24	19	17
Appearance of melanophores on the yolk sac	37	25	19	17
Kupffer's vesicle formation	39	25	19	17
Appearance of melanophores on the oil globule	40	28	23	20
Appearance of melanophores on the embryo	41	29	24	20
Tail bud formation	42	28	23	21
Appearance of xanthophores	42	30	25	24
Beginning of tail prolongation	45	31	28	26
Disappearance of the Kupffer's vesicle	53	37	31	26
Eye lens formation	51	37	32	28
Beginning of hatching	94	67	53	42
Completion of hatching	102	76	58	45

anus is located slightly posterior of the mid-point of the body until the body begins to grow in height and the anlages of the dorsal and anal fins appear. A large amount of yolk with an oil globule located posteriorly at the bottom of the yolk remains in the newly-hatched larva. The yolk is absorbed within 6 days after hatching, while the oil globule remains for several days more. The day after hatching, black pigments appear on the eyes, and the bud of the pectoral fins emerge. In the three-day-old larva, the mouth is open and the digestive tract is convoluted *(Fig. 2C)*.

The air-bladder is formed within several days after the yolk absorption and the anlage of the caudal fin appears in the larva reaching around 3.4 mm in total length *(Fig. 2E)*. The body noticeably starts to grow in height and the anlages of dorsal and anal fins appear in larva of 4.5 mm. In larva of 4.5 mm *(Fig. 2F)*, the head is round, the body is high, the fin rays of the unpaired fins are under formation, and the pelvic fins are just being formed. In fish reaching around 8.9 mm *(Fig. 2G)*, all the fins are found to be completely formed. The fish reach the juvenile stage before this size, which is slightly smaller than reported by Kinoshita (1988).

Melanophores in a newly-hatched larva are distributed all over the body, dorso-laterally on the yolk sac and dorsally on the oil globule. The xanthophores virtually cover the same locations except in the oil globule, where the melanophores are located dorsally and the xanthophores laterally. As the larva grows, the chromatophores are concentrated in some locations and become sparse on the shoulder and posterior half of the tail. Some melanophores are usually located dorsally and ventrally near the rear end of the tail.

As the larva nears the end of the prolarval stage, the melanophores on the body start to come together on the dorsal and ventral edges and on the mid-lateral line, forming three horizontal lines in lateral view. As the larva grows, melanophores

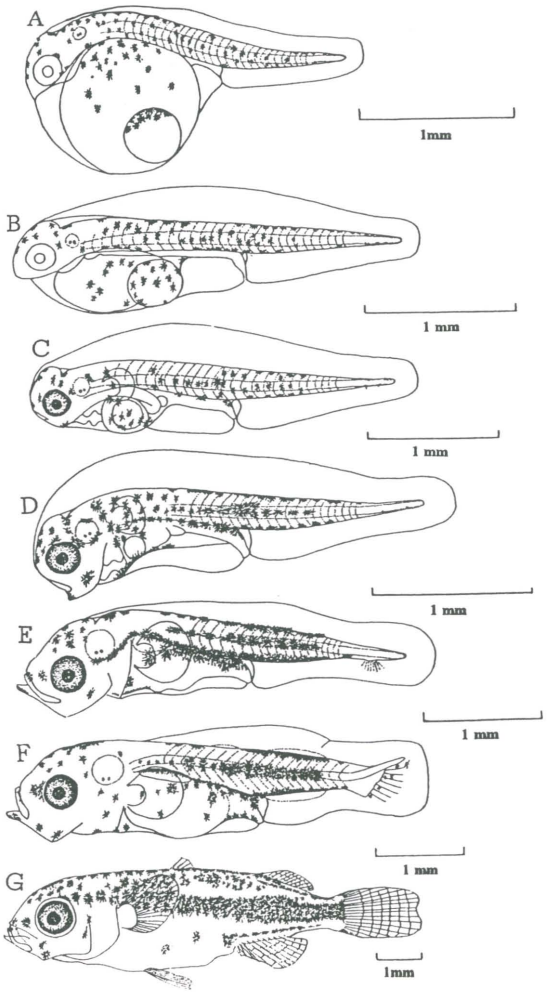

Fig. 2. Morphological development of L. carinata *larvae and juveniles. A, newly hatched, 2.1mm in total length: B, half a day old, 2.6mm; C, 3 days old, 2.9 mm; D, 9 days old. 2.6mm; E, 15 days old, 3.4mm; F, 30 days old, 4.5mm; G, 40 days old, 8.9mm (All drawings and measurements have been made using live specimens).*

covering the body grow thick on the posterior region of the trunk and anterior part of the tail, rendering the myomere uncountable. After the yolk and oil globule are absorbed, the visceral region encasing the air-bladder is covered by thick melanophores. The melanophores are also located on the dorsal surface of the head, the edges of the jaws, the bottom of preopercle and the ventral edge of breast, and these distribution patterns do not change significantly until the larva nears the juvenile stage.

About the time when the caudal fin rays are under formation and the anlages of the dorsal and anal fins appear, the mid-lateral melanophore line starts to broaden, especially on the anterior part of the tail. At the same time, the melanophores on the dorsal sides of the head and body begin to increase.

The guanophores appear sparsely after the postlarva stage is reached and soon become dense all over the body.

Species Differences in the Egg, Larva and Juvenile Stages

The egg and oil globule sizes of artificially fertilized *L. carinata* eggs are shown in Table 2 along with those of *L. haematocheila*. *L. carinata* eggs are smaller in egg size and oil globule size than those of *L. haematocheila*. Although there is an overlap in egg size between the two species, the *L. carinata* egg can be distinguished by the evidently large size of the oil globule, as described by Mito (1988).

The external morphologies of the larval and juvenile stages, including pigmentation are very similar in mugilids, even between genera. Yashouv and Berner-Samsonov (1970) used the presence or absence of chromatophores on the yolk sac, oil globule and forehead as characters which distinguish the mugilids in early larval stages occurring along the Israeli coast. Some descriptions of eggs and larvae of *Liza* species are available (Natarajan and Patnaik 1972; Fujita 1979; Cambray and Bok 1989), but differences are not definite within genera. *L. carinata* has been considered to be difficult to distinguish from *L. haematocheila* in the larval stage (Kinoshita 1988). We studied the reared specimens to determine the distinguishing features, and then confirmed our findings using wild specimens.

L. carinata larvae are apparently smaller than *L. haematocheila* when the stage is defined by morphological features. Five-day-old *L. carinata* prolarvae which have nearly absorbed the yolk but not the oil globule, measure 2.6 to 2.8 mm in notochord length and 2.8 to 3.0 mm in total length after fixation in formalin solution while *L. haematocheila* larvae at the same stage measure 3.0 to 3.2 and 3.1 to 3.5 mm respectively. *L. carinata* postlarvae which have a straight notochord but have yet to reach fin formation measure 2.7 to 3.0 mm and 3.0 to 3.4 mm, while *L. haematocheila* larvae measure 3.4 to 4.0 and 3.7 to 4.2 mm respectively. The *L. carinata* larvae showing caudal fin rays and anlages of the 2nd dorsal and fins measure 3.6 to 4.3 mm and 3.8 to 4.8 mm, while *L. haematocheila* larvae are 4.2 to 5.1 and 4.4 to 5.5 mm respectively.

TABLE 2
Comparison of the egg and oil globule sizes of the two *Liza* species.

		L. carinata	*L. haematocheila*
	x	0.84	0.96
Egg diameter (mm)	Range	0.78-0.93	0.88-1.02
	SD	0.03	0.03
	x	0.32	0.46
Oil globule diameter (mm)	Range	0.27-0.37	0.44-0.49
	SD	0.03	0.01

In prolarva and postlarva stages, the melanophores on the top of the head are distributed irregularly in *L. haematocheila (Fig. 3C and D)*, while in *L. carinata* a comparatively widely branched melanophore is distributed at the posterior end of the cranium *(Fig. 3A and B)*. Up to the stage of caudal fin formation the tiny melanophores located on the rear end of the tail in *L. carinata* may also serve as a distinguishing feature from *L. haemotocheila* larva, which does not have a melanophore in that position.

The criteria used to distinguish the larvae can not be applied to the juveniles, for the melanophores on the head increase in *L. carinata* and their distribution becomes irregular. In *L. carinata* juveniles *(Fig. 3E)*, the mid-lateral melanophore row extends farther ahead than in *L. haematocheila (Fig. 3F)*, although the width of the row and melanophore density vary according to the rearing conditions. The characteristic dorsal ridge in *L. carinata* appears after reaching 30 mm in total length. This ridge does not occur in *L. haematocheila.*

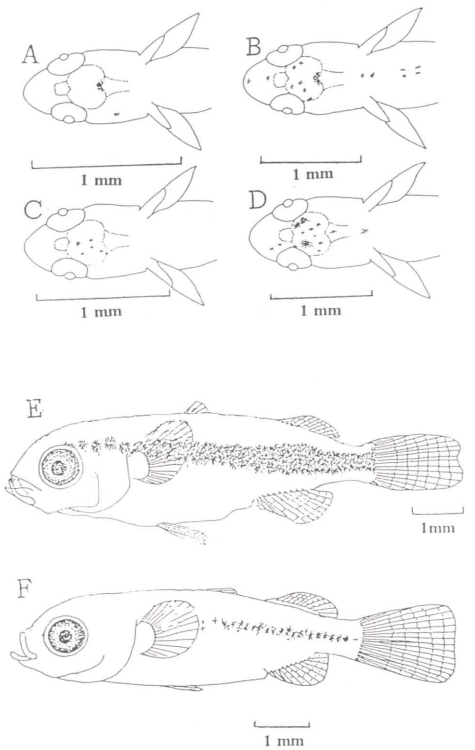

Fig. 3. Comparison of the pigmentation of L. carinata *and* L. haematocheila, *showing the points to distinguish the two species. A to D, anterior dorsal views of the both species; A and B,* L. carinata; *C to D,* L. haematocheila; *A, 2.8 mm total length; B, 4.3 mm; C, 3.4 mm; D, 5.1 mm; E to F, lateral views showing the midlateral melanophore row; E,* L. carinata, *8.9 mm and F,* L. haematocheila, *10.1 mm. (All drawings and measurements have been made using specimens fixed in formalin).*

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