

## Reproductive Behavior, Eggs and Larvae of a Lutjanid Fish, *Lutjanus stellatus*, Observed in an Aquarium

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**Abstract** Reproductive behavior and early life history of the white spotted snapper, *Lutjanus stellatus* Akazaki, are described from aquarium observations. Spawning took place between a female and 2-12 males in the early hours of each evening from mid-May to mid-June, 1984.

Six easily distinguishable behavior patterns were involved in the spawning sequence: a) Schooling; b) Searching; c) Nuzzling; d) Rushing toward the surface; e) Spawning; and f) Post spawning. The species was considered to be a group spawner.

Fertilized eggs were spherical, transparent, buoyant and unpigmented. They were 0.80-0.85 mm in diameter, and contained a single oil globule measuring 0.16-0.17 mm in diameter. Hatching took place 30 h after fertilization. Immediately after hatching, larvae were 2.48-2.56 mm in total length and had a large ellipsoidal yolk. An oil globule was situated at the front tip of the yolk. Early life stages of *L. stellatus* were described for the first 3.5 days after hatching.

There is only one direct observation of reproductive behavior and spawning in lutjanids in their natural habitat; *Lutjanus synagris* off southeast Florida (Wiklund, 1969). Bell and Colin (1986) observed spawning of *Caesio teres*, formerly considered a lutjanid, but now included in the Caesionidae, off the East Channel of Enewetak Atoll, Marshall Islands. Thresher (1984) and Grimes (1987) have reviewed the limited field and in-captivity information on spawning in lutjanids and caesionids. The paucity of information stems primarily from logistic problems associated with extended underwater observations and the wide-range of lutjanid habitats.

Reproductive behavior of captive *L. campechanus* from the Gulf of Mexico and *L. kasmira* from Japan, described by Arnold et al. (1978) and Suzuki and Hioki (1979), respectively, provided valuable early life information regarding spawning behavior, eggs and larvae, despite not being from natural habitats.

Leis (1987) reviewed the early life history of lutjanids, with additional reports by Kojima (1988), Mori (1988), Potthoff et al. (1988), Iwatsuki et al. (1989), Soletchnik et al. (1989), and Iwatsuki et al.

(1991). However, knowledge is still incomplete.

The white spotted snapper, *Lutjanus stellatus* Akazaki, is distributed in limited temperate and subtropical regions of the western North Pacific from southern Japan to Taiwan and to the vicinity of Hong Kong (Akazaki, 1983; Shen, 1984; Allen, 1985; Allen and Talbot, 1985). However, little is known of the early life history and ecology of this species.

This paper describes spawning behavior and egg development of aquarium-held *L. stellatus*. Comparisons of eggs and just-hatched larvae are made between the few species of *Lutjanus* for which such information is available.

### Materials and Methods

**Observation of reproductive behavior.** The adult brood stock of 118 *L. stellatus* were captured mainly by hook-and-line fishing around Kashiwa-jima, Hata-gun, Kochi Prefecture during summer months from 1973 to 1980. They were reared in a doughnut-shaped concrete tank at the Yashima Aquarium,

Kagawa Prefecture. The sizes of parental *L. stellatus* were estimated from three preserved specimens, which had died within four months after the spawning period. Two parental males measured 45.8 cm and 47.0 cm in standard length and weighed 3.60 kg and 3.90 kg, respectively. A female was 46.8 cm in standard length and weighed 3.85 kg. Other parental fishes were almost the same size, their estimated ages being from 7 to 15 years, based on their duration in captivity and size when first collected.

The doughnut-shaped concrete tank used for observations of spawning behavior had an inner acrylate circle wall (2 m, 10.4 m and 16.2 m in height, and inner and outer diameter, respectively) for viewing by aquarium visitors. It had a surface area of 121.1 m<sup>2</sup> and water capacity of 242 m<sup>3</sup>. In the upper portion of the outer concrete wall, nine small, paned windows (0.6 m in height × 0.4 m in length) allowed indirect natural moon light to enter from the south-east.

The tank also contained 18 other species of marine fishes primarily from Kochi Prefecture, viz. Serranidae (8 spp.), Carangidae (3 spp.), Lutjanidae (2 spp.), Lethrinidae, Haemulidae, Acanthuridae, Ehippididae and Triakidae (1 sp. each) according to Masuda et al. (1984).

Observations of spawning behavior of *L. stellatus* were made from the end of May to the middle of June, 1984. Serial specimens of eggs and larvae observed during the study were deposited in the Department of Animal Science, Miyazaki University as Cat. No. MUFS 8663–8667.

**Rearing of eggs and larvae.** From mid-May, 1984, eggs of a second species were found in the tank, but were easily differentiated as follows: 1) difference in egg diameter (mean 0.83 mm in *L. stellatus* and 0.76 mm in the other species); 2) different position of the oil globule in the yolk (situated at the anterior tip of the yolk in *L. stellatus*, at the posterior tip of the yolk in the other species). The diagnostic features of the other eggs were consistent with their being from a serranid (Mito, 1963; Ukawa et al., 1966).

Buoyant, fertilized eggs, which were spawned on May 14 and May 28, were used for rearing. Just after spawning, eggs were collected by a net (1.1 m in diameter, 2.7 m in length, mesh aperture size 0.3 mm) which had been previously set up at the surface for ten minutes, and transferred into a small 10 l polyethylene tank. The eggs were subsequently transported to the Kagawa Prefectural Fisheries Experi-

mental Station, and one hundred eggs being placed in each of three 1 l glass beakers, which were kept at 24.0 ± 0.5 °C during incubation.

Illustrations of live specimens were made with the aid of a drawing attachment on a stereo-microscope and many color photographs were also taken in life. All size data are from live specimens. Variance is given as standard deviation (SD).

S type rotifers, *Brachionus* sp., were concentrated and provided as food for the larvae from the second day after hatching to the fourth day, by which time all the larvae had died.

## Results

**Reproductive behavior.** Fertilized eggs of *L. stellatus* were found in the tank every night from May 13 to June 14, 1984, water temperatures ranging from 24.1 to 26.2 °C. However, reproductive behavior in this species was first observed at 20:30 on May 28, when evening observation of the aquarium began.

Spawning behavior was observed from 20:00–23:00, which corresponded to 1–4 hours after sunset. From May 28 to 10 June, we were able to record about 37 spawnings on the basis of reproductive behavior and finding of fertilized eggs.

Fig. 1 shows a diagrammatic representation of the reproductive behavior. Six easily distinguishable behavior patterns were involved in the spawning sequence.

**Schooling.** The fish were usually quiet at night before the spawning time. Each individual stayed just above the bottom, although fishes occasionally swam about slowly in a small school. However, during the spawning period, a general increase in activity occurred. The spawners were active, usually aggregating in a large school of more than about 100 fishes hovering over the bottom, where it was dimly illuminated by indirect natural moon or reflected light through the nine paned windows.

**Searching.** Just before spawning female fishes which had a swollen abdomen and whitish-yellow or lighter-colored body, stayed near the bottom of the school. It became possible to predict which female fish would spawn each day on the basis of the lower position female and its lighter-colored body. Males searched for a lighter-colored female and gathered together around her.

**Nuzzling.** Typically, a male started his courtship behavior toward a selected female by pecking and

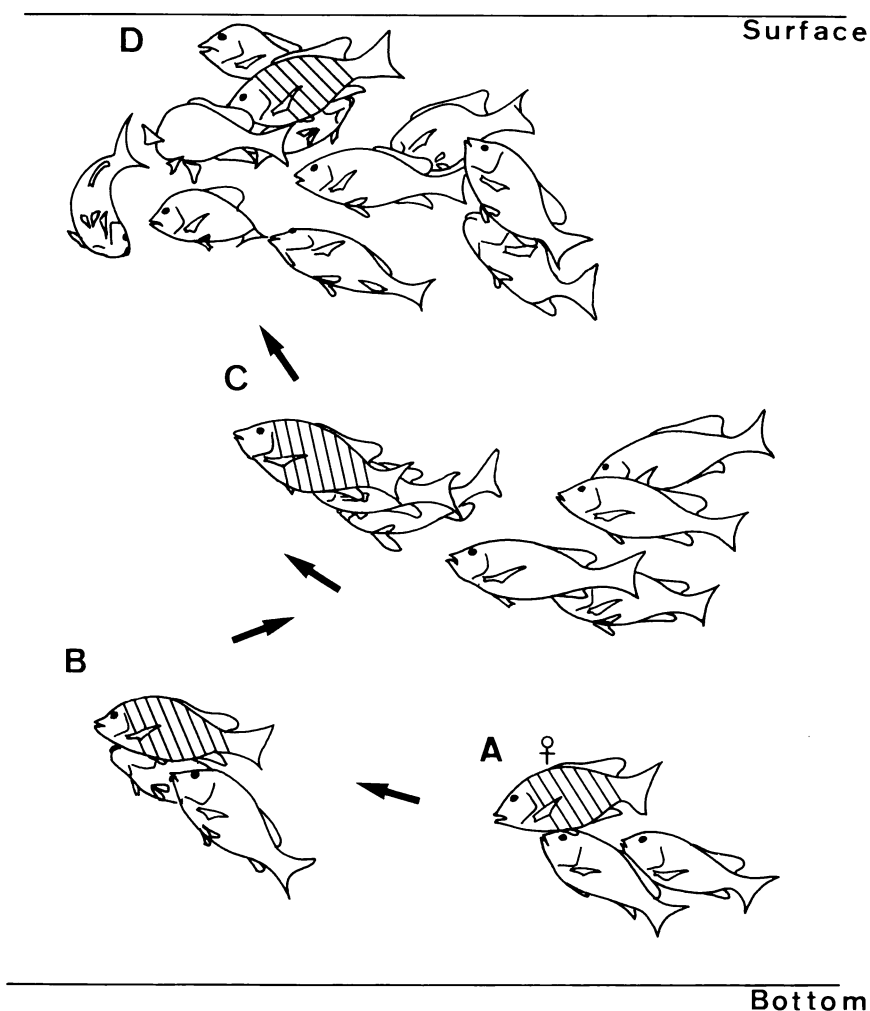


Fig. 1. Diagrammatic view of the courtship and reproductive behavior of *Lutjanus stellatus* in the aquarium.

pushing her swollen abdomen with his snout (Fig. 1A). Each male from the small school was successively attracted to the female.

**Rushing toward the surface.** A few males pushed the selected female upward in the water, quickly ascending together in a spiral (Fig. 1B) to about the middle layer of the tank, before all fishes returned individually to the bottom. Such ascending movements were observed several times, the pattern being repeated 10 to 15 times over 2 to 3 hours. As the spawning time approached, ascending movements became more rapid and ceased higher in the water column.

**Group Spawning.** Between 5 and 10 males participated in the spiral and ascending movements (Fig.

1C). Eventually, all the involved males quickly ascended spirally to just beneath the surface of the water (Fig. 1D). Group spawning, in which a female was pushed up by 2 to 10 males, usually occurred but rarely some lone females ascended spirally. In such cases, from 5 to 10 males rushed upward after the female and spawned.

**Post spawning.** After spawning, each adult returned individually to the bottom, slightly apart from the school. However, some females and males joined another school for group spawning.

Different females with 2–10 males spawned successively at 5–10 minute intervals. After 23:00, spawning was not observed. Although the same female spawned a few times on the same night, it could not

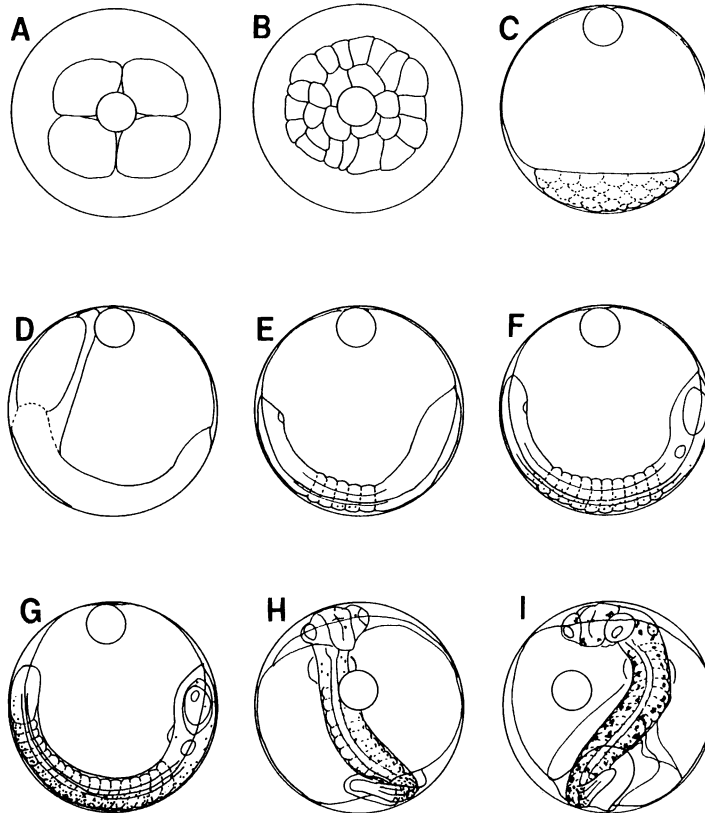


Fig. 2. Embryonic development of eggs of *Lutjanus stellatus*. A) 4-cell stage, 55 min after fertilization; B) 32-cell stage, 1 h 40 min; C) Morula stage, 2 h 50 min; D) Embryonal body appears, 10 h 25 min; E) 5-myotome stage, 12 h 10 min; F) 10-myotome stage, 15 h 45 min; G) 18-myotome stage, 18 h 30 min; H) 24-myotome stage, 26; and I) 25-myotome stage, 29 h 50 min.

be determined if the same female spawned successively every night or not during the spawning period.

**Eggs and embryos. Morphology.** Fig. 2 illustrates the embryonic development in this species. The fertilized eggs were spherical, transparent, buoyant and unpigmented.

Eggs ranged from 0.80 to 0.85 mm in diameter (mean =  $0.83 \pm 0.02$  mm, N = 36), and contained a single oil globule measuring 0.16–0.17 mm in diameter (mean =  $0.16 \pm 0.003$  mm, N = 36). The perivitelline space was narrow, the chorion clear and unsculptured, and the yolk homogeneous and unsegmented.

Eggs developed in a manner typical of pelagic teleosts as follows: fertilized eggs reached the 2-cell stage in about 40 min, the 4-cell stage in 55 min (Fig. 2A), the 32-cell stage in 1 h and 40 min (Fig. 2B), the morula stage in 2 h 50 min (Fig. 2C), and the

gastrula stage in 6 h 40 min after fertilization.

After 10 h 25 min after fertilization, about 3/4 of the yolk was covered by the blastoderm and the embryonic body had appeared (Fig. 2D). At the 4-myotome stage, Kupffer's vesicle could be observed. The 5-myotome stage (Fig. 2E) could be observed after 12 h 10 min.

After 15 h 45 m the 10-myotome stage (Fig. 2F), including auditory vesicles and lenses in the eyes could be observed. In the 18-myotome stage (Fig. 2G), reached at 18 h 30 min. Kupffer's vesicle had disappeared and the heart appeared.

After 26 h, 26 myotomes were counted on the embryonic body and the posterior tip of the caudal had already become separate from the yolk. Rudiments of the pectoral fin were also observed (Fig. 2H).

After 29 h 50 min the embryo occupied 5/6 of the

yolk circumference, the posterior half of the body being separated from the yolk (Fig. 2I). Hatching started by 30 hours after fertilization with 80% of the eggs being hatched by 33 hours after fertilization.

**Pigmentation.** The embryo was lightly pigmented, with a fairly constant pattern of melanophores. The latter were observed only on the embryonic body.

At the 5-myotome stage, the first two melanophores appeared, along the dorsal side of the 2nd and 3rd myotomes (Fig. 2E). By the 7-myotome stage, the total number of melanophores had increased, in keeping with the increase in myotomes.

By the 10-myotome stage (Fig. 2F), 2–5 melanophores had appeared in each dorsal myotome, totaling about 40 melanophores in all. No melanophores were observed on the head or posterior caudal region.

Melanophores had increased in number by the 18-myotome stage (Fig. 2G), with 3–8 melanophores on the dorsal surface of each myotome. In addition, melanophores had appeared on the head and caudal region.

The 24-myotome stage (Fig. 2H) had become slightly punctate, forming two lines on the dorsal surface side of the body. At the 25-myotome stage (Fig. 2I), melanophores had decreased in number, but had clearly developed into punctate or dendritic structures.

**Larvae. Morphology.** Larvae were deep to slender-bodied, laterally compressed and had 24–26 myotomes (9+10+15–16) at 3.5 days of age.

The just-hatched larvae were transparent and ranged from 2.48 to 2.56 mm (mean  $2.52 \pm 0.03$  mm,  $N=7$ ) in total length (TL; including the anterior portion of the yolk protruding beyond the snout of the larva). From the anterior tip of the snout to the posterior portion of the caudal fin measured  $2.37 \pm 0.02$  mm ( $N=7$ ). The head was slightly deflected at the anterior end of the ellipsoidal yolk (about 1.23 mm length at the longest diameter and 0.57 mm at shortest diameter).

The surface of the whole body was uneven, small, very short lines being observable under the correct incident light angle. The anterior portion of the yolk extended beyond the snout of the larva and an oil globule was situated close to the anterior inner margin of the yolk. The number of myotomes was  $10 + 16 = 26$ . The anus was not contiguous with the posterior end of the yolk.

Six hours after hatching, larvae were 2.78 mm TL

and had  $10 + 16 = 26$  myotomes (Fig. 3B). The yolk was about 1.20 mm in length. The anus was situated at about the midpoint of the larva. The small lines over on the body surface, observed in just-hatched larvae, were still observable but had formed small, granule-like spots (Fig. 3B–F). The snout was also formed.

Twelve hours after hatching, larvae were about 2.90 mm TL and had  $10 + 16 = 26$  myotomes (Fig. 3C). A small yolk remained, measuring about 1.20 mm in length. The granules on the body surface were unchanged, but the dorsal margin of the temporal region had become thickened.

Thirty hours after hatching, larvae were about 3.42 mm TL and had  $9 + 16 = 25$  myotomes (Fig. 3D). The remaining yolk measured 0.60 mm at the longest diameter. The anus was situated  $4/9$  of the total length from the head. The yolk had been absorbed considerably that remaining being anteriorly situated. Granular spots on the body surface had increased and the eye lens had cleared. Pectoral fin rudiments had become more clear.

Fifty-four hours after hatching, larvae were 3.46 mm and had  $9 + 16 = 25$  myotomes (Fig. 3E). The yolk sac was barely retained. The mouth and anus had opened, and the eye lens become blackish in color.

Ninety-hours after hatching, larvae were about 3.40 mm TL and had  $9 + 15 = 24$  myotomes (Fig. 3F). Little yolk remained.

Larval mortality rate was high and the last individual died in 4 days after hatching. No remarkable growth or changes in shape were observed during this period.

**Pigmentation.** Larvae were pigmented, with fairly constant melanophore patterns.

Immediately after hatching, larvae had many melanophores along the body axis from the head to the caudal section (Fig. 3A). Melanophores on the eye constituted very small spots but those on other parts of the body were dendritic. Abdominal melanophores were situated the more or less at the dorsal surface, but those on the caudal region were present both dorsally and ventrally.

By 6 hours after hatching (Fig. 3B), the melanophores on the dorsal surface had moved mostly towards the lateral and ventral surfaces of the body, some having become slightly expanded in shape. Caudal melanophores had slightly increased in number. A few melanophores had appeared about the center of the yolk.

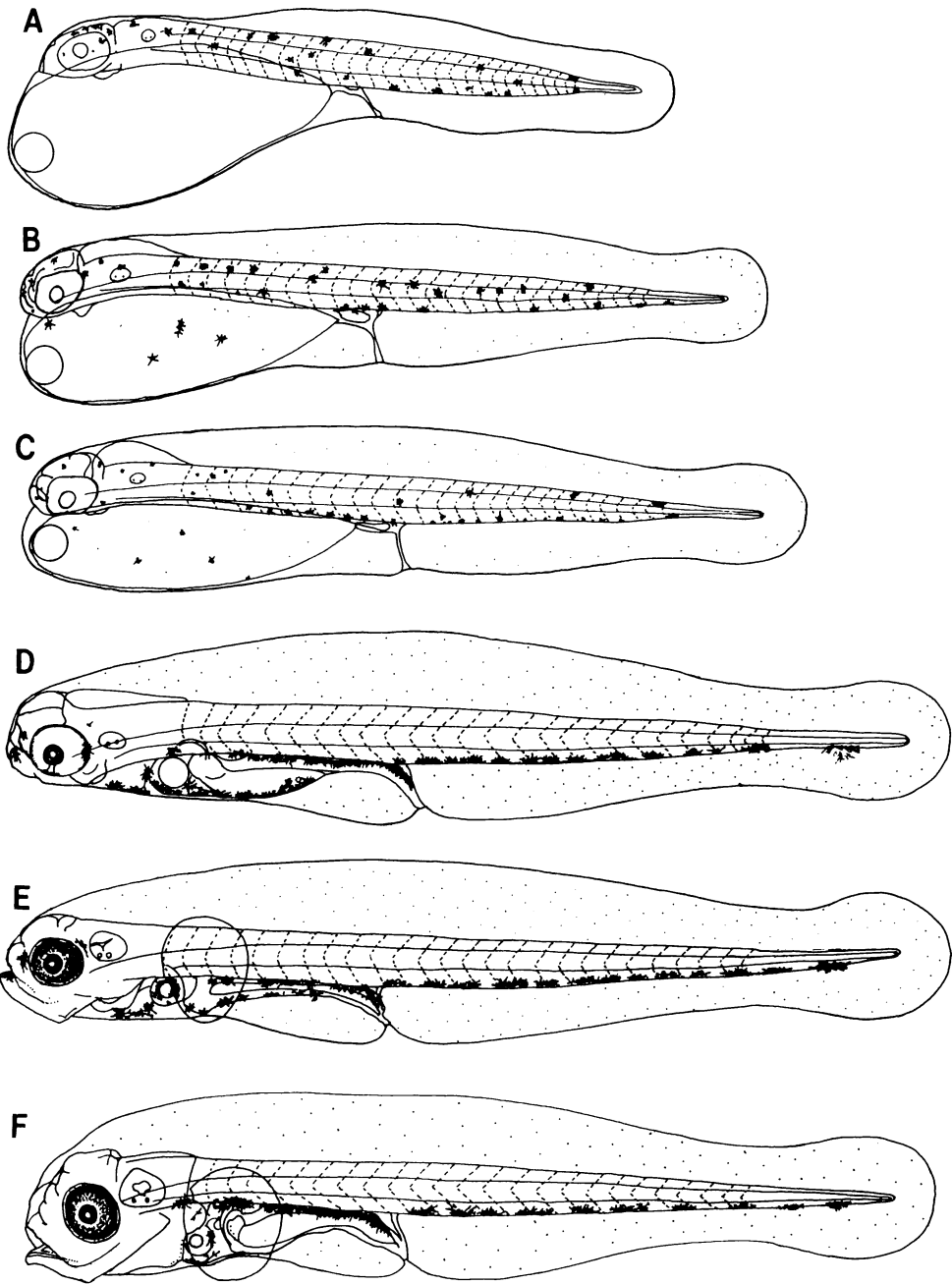


Fig. 3. Larval development in *Lutjanus stellatus* (lengths are total length). A) just-hatched larva; 2.52 mm; B) 6 h after hatching, 2.78 mm; C) 12 h after hatching, 2.90 mm; D) 30 h after hatching, 3.42 mm; E) 54 h after hatching, 3.46 mm; and F) 90 h after hatching, 3.40 mm.

Twelve hours after hatching (Fig. 3C), the melanophores on the head and yolk had not changed but those on the dorso-lateral surface had moved to the

ventral part of the body. The overall number of melanophores had slightly decreased.

Thirty hours after hatching (Fig. 3D), the mel-

anophores had become strongly dendritic, 16 or 18 melanophores being observed along the ventral and caudal edges of the body, with a further two or three beneath the eyes and seven or eight on the anterior part of the ventral membranous fin.

Fifty-four hours after hatching (Fig. 3E), the melanophores were observed along the ventral ridges of the body, those on the caudal part having increased in number and became larger at the previous stage. Numerous spotted melanophores had appeared on the posterior part of the digestive tract and in the eyes.

Ninety hours after hatching (Fig. 3F), the eyes had become blackish in color with the development of the melanophores. Elsewhere melanophores had become continuous, especially on the upper portion of the digestive tract.

**Morphometrics.** Morphometrics of just-hatched *L. stellatus* larvae, expressed as proportions of total length, are included in Table 1.

The pre-anal proportion of the total length decreased from 0.52 in just-hatched larvae to 0.41 in 80 hours-old larvae, the anal position having moved anteriorly from about the mid-point of the body.

### Discussion

**Spawning.** Reproductive behavior of *L. stellatus* in the tank was fundamentally similar to that described in previous reports of lutjanid spawning (Wiklund, 1969; Suzuki and Hioki, 1979). Several similar points were confirmed: 1) group spawner; 2) repeating spiral and ascending movements before spawning; 3) spawning time occurring during the crepuscular period to about 2 hours after sunset. However, some different behavior was also confirmed. In *L. kasmira*, initial spawning behavior was between pairs (Suzuki and Hioki, 1979) but in *L. stellatus*, a few males gathered around one female. Similar behavior in a natural habitat was observed in *L. synagris* by Wiklund (1969). This difference, pairing or one female with a few males, is difficult to explain, but different reproductive behavior might have caused resulted from different densities and sex ratios of mature adults. Furthermore, except during the spawning season, we could not usually distinguish between the sexes of *L. stellatus*. In the spawning season, however, it was easy to differentiate females from males on the basis of the body color pattern, a lighter-colored body and a swollen abdomen in the former (this was also described in *L.*

*synagris* by Wiklund [1969]). Nuptial coloration in lutjanids occurred before the onset of dusk. Thus spawning could be anticipated each night if a light-colored female was observed before dusk.

**Eggs and larvae.** Taxonomic information on eggs and just-hatched larvae of lutjanids has increased in the last ten years. Information on eggs and just-hatched larvae is available for *L. kasmira* from Japan (Suzuki and Hioki, 1979), *L. campechanus* from U.S.A. (Rabalais et al., 1980; Minton et al., 1983), *L. russelli* from China (Liu and Hu, 1980), *L. vitta* from China (Lu, 1981), *L. lutjanus* from China (Zhang et al., 1985), and *L. erythropterus* from China (Zhang et al., 1985). *L. griseus* larvae 2.8 mm and over in notochord length were described by Richards and Saksena (1980).

Table 2 shows important characteristics of eggs and larvae of eight species. *L. erythropterus* has a bigger egg (0.90–1.02 mm) than other lutjanids (0.70–0.85 mm). The lack of an oil globule in the former might be unique in *Lutjanus*.

Unfortunately, a basis for the identification of eggs and larvae of the above eight species cannot be provided because of the large number of species in the genus *Lutjanus* and the simplified figures and descriptions that are available. However, the pattern of melanophore pigmentation, especially melanophores along the ventral edges of the body, might prove to be a diagnostic feature applicable to *Lutjanus*.

Granules on the entire body surface were also observed in *L. lutjanus* (Zhang et al., 1985). In *L. stellatus*, this feature was visible in both fresh and fixed, 6 hour-old specimens. Because this seems to be a significant feature, it should be checked in the remaining species of *Lutjanus* and in other genera of Lutjanidae, and its functional status considered.

At this stage, unique species-diagnostic features of the larva cannot be confirmed. Further information is needed on melanophore pigmentation patterns and changes in growth or yolk shape.

Table 1. Changes in body proportions (relative to total length) during the first 80 hours of development of *Lutjanus stellatus* (proportion at hatching given first)

	Larvae
Head length	0.44–0.56
Body depth	0.72–0.66
Pre-anal length	0.52–0.41
Eye diameter	0.20–0.24

Table 2. Important characteristics of eggs and larvae of eight lutjanid fishes

	<i>L. campechanus</i>	<i>L. erythropterus</i>	<i>L. griseus</i>	<i>L. kasmira</i>	<i>L. russelli</i>	<i>L. lutjanus</i>	<i>L. stellatus</i>	<i>L. vitta</i>
Egg size	0.77–0.85	0.90–1.02	—	0.78–0.85	0.70–0.78	0.75–0.79	0.80–0.85	0.78–0.84
Single oil globule	present	none	—	present	present	present	present	present
Oil globule size	0.15–0.19	—	—	0.13–0.14	0.15–0.16	0.16–0.17	0.16–0.17	0.16–0.18
Fertilized eggs	buoyant	buoyant	—	buoyant	buoyant	buoyant	buoyant	buoyant
Total length of just-hatched larvae	2.2* (probably live)	1.36 (fixed)	—	1.83 (live)	2.0–2.2 (live)	1.29 (fixed)	2.48–2.56 (live)	1.45–1.50 (fixed)
Morphology in yolk	strongly elongated	elliptic near rounded	—	elliptic near rounded	elliptic near rounded	strongly elongated	elliptic near rounded	elliptic
Melanophore pattern in just-hatched larvae	— (not adequate in description)	3–4 pigments in predorsal membrane and upper part	—	upper and posterior part	pigmentation observed in whole body	not clear pigmentation observed	upper and lateral part	upper and lateral part
Melanophores along ventral edges of body	present	present	present (2.8 mm in NL)	present	— (not adequate in description)	present slightly	present	present
References	Rabalais et al. (1980); Minton et al. (1983)	Zhang et al. (1985)	Richards and Saksena (1980)	Suzuki and Hioki (1979)	Liu and Hu (1980)	Zhang et al. (1985)	this study	Lu (1981)

\* Size is measured in standard length, probably notochord length (NL)



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フェダイ科魚類フェダイ *Lutjanus stellatus* の飼育下における産卵行動とその卵内発生及び孵化仔魚

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高知県幡多郡柏島周辺近海で採集後飼育されたフェダイ科魚類フェダイ (*Lutjanus stellatus*) の飼育下における産卵行動とその卵内発生及び孵化後 3.5 日目までの仔魚を記載した。

受精卵は、1984 年 5 月 13 日から 6 月 14 日まで毎夜観察され、また 5 月 28 日から 6 月 14 日までの間で延べ 37 回の産卵行動を観察した。親魚は、まず水槽内の採光窓の下の一ヶ所に群れ (100 尾以上) を作った後、腹部が膨出し体表全体が白変した一尾の雌と、数尾の雄による求愛によって産卵行動を始めた。それらの雄は雌の腹部を交互につつき、その雌を上方に向けて下から押し上げた。更に多数の雄がそれに追尾し、水表面まぎわまで螺旋状に一瞬のうちに上昇した後、放精・放卵に至った。

本種の受精卵は、油球 1 個を有する卵径 0.80-0.85 mm の球形分離浮性卵で、水温  $24.0 \pm 0.5^{\circ}\text{C}$  では受精後 30 時間後に孵化を開始し、その後 3 時間で 80% の卵が孵化を終えた。孵化直後の仔魚は、平均全長 2.52 mm、卵黄は長卵形で大きく、その先端は吻端より前方に突出していた。油球はほぼ卵黄先端にあり、卵黄表面から突出していなかった。孵化後 3.5 日の仔魚は全長 3.40 mm で、卵黄をほぼ吸収して口と肛門が開いた。

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