Spawning Behavior, Eggs, and Larvae of the Butterflyfish, Chaetodon nippon, in an Aquarium

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Abstract Spawning behavior and early life history of Chaetodon nippon Steindachner et Döderlein are described in detail from aquarium observations. The early life history of the present species is the first record among the family Chaetodontidae. Spawning of C. nippon occurs between multiple males and one female in the early hours of the night in March, August, September, and October at water temperatures of 23° C or higher. Fertilized eggs are buoyant, spherical, and colorless. They measure $0.70 \sim 0.74$ mm in diameter and contain a single oil globule. Hatching takes place 28 hrs. 10 min. after fertilization at water temperatures of $22.2 \sim 23.7^{\circ}$ C. Newly hatched larvae measure $1.45 \sim 1.53$ mm in total length and have a large ovoid yolk. An oil globule is situated at the rear of the yolk. Early larval stages of C. nippon are described for the first three days after hatching. The mode of reproduction and the characteristics of developing eggs and larval stages of this species are compared with those of allied fishes previously studied.

Both the angelfishes (family Pomacanthidae) and butterflyfishes (family Chaetodontidae) are well-known coral reef fishes. Among the former, the mode of reproduction has been reported on several species of the genus Centropyge by Lobel (1975, 1978) and by Moyer and Nakazono (1978) from underwater observations, and on two species of the genus Genicanthus by Suzuki et al. (1979) from aquarium observations. However, little or nothing is known of the spawning behavior and early life history of the butterflyfishes. In 1975, the reproduction of Chaetodon nippon occurred several times in the aquarium of the Marine Science Museum of Tokai University. Spawning behavior and characteristics of early larvae of C. nippon show unique and interesting differences in comparison with those of the angelfishes.

Materials and methods

Parental fish and their keeping. Chaetodon nippon is distributed from the Philippines and Taiwan (Shen and Liu, 1975) to the Pacific coast of central Japan, and inhabits shallow to rather deep coastal waters. Twenty-four adult fish were collected by SCUBA-divers from depths of 30~50 m near Ito City along the east coast of Izu Peninsula on February 14, 1974, and they were maintained in an

aquarium tank. The tank used for observations of spawning behavior of *C. nippon* is made of concrete with a glass window. It has a surface of 2.8 m², water depth of 1.3 m, and capacity of 3.6 m³. The tank is situated in a dimly lighted room and is illuminated from 8:00 to 18:00 hrs. usually by two 40-watt fluorescent lamps placed about 1 m above the surface of the water.

When spawning occurred, the fish were being maintained with 178~213 other small coastal fishes of 5~6 species: Sacura margaritacea (Hilgendorf), Pseudanthias elongatus (Franz), Franzia squamipinnis (Peters), Chaetodontoplus septentrionalis (Temminck et Schlegel), Cirrhilabrus temmincki Bleeker, and Sebastes joyneri Günther. All the fishes including C. nippon were fed on cut or chopped meat of horse mackerel, penaeid prawns, and clams once a day. The size of the adult C. nippon ranged from 119 to 161 mm in total length and from 44.1 to 126.0 g in body weight. Although the parental fish could not be divided into both sexes by their appearances prior to or after the spawning season, the males could be easily distinguished from the females by their courtship behavior, and the females generally had expanded abdomens during the spawning season. The aquarium water was circulated once an hour by a pump through a sand

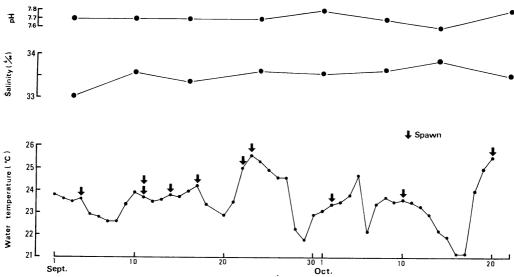


Fig. 1. Changes in water conditions during maintenance of parental individuals of *C. nippon* from September 1 to October 21, 1975. Spawning dates are shown as arrows.

filter. Changes in water temperature, pH, and salinity from the beginning of September to the end of October in 1975 are shown in Fig. 1. Reproduction of *C. nippon* occurred most frequently in that two-month period.

Rearing of eggs and larvae. Just after reproduction was ascertained, spawned eggs were collected with a small net and transferred into small receptacles of 30 *I* capacity. The developing eggs and hatched larvae were reared in still water with weak aeration in the 30 *I* receptacles. The water was occasionally changed.

Results

Spawning behavior. Under usual conditions, the adult C. nippon swim separately or gather in loose aggregations in the aquarium. From 1 to 2 days prior to the spawning, the abdomens of gravid female expand remarkably. This expansion seems to reach an extreme $1\sim2$ hours before spawning occurs (Fig. 2). From 17:00 to 20:00 hrs., i.e., about one hour before spawning occurs, $5\sim6$ males begin to fight with each other. About $30\sim40$ minutes before spawning occurs, a gravid female, who has stayed close to a rock at the bottom, ascends rapidly alone towards the surface of the water. When the female reaches $10\sim20$ cm below the surface, she begins to swim in

circles. The largest male, who has stayed in mid-water, pursues the female and pecks her abdomen with his snout or nuzzles her with his forehead. Then the two fish swim together in circles repeatedly. During the mutual courtship, $2\sim3$ other males of C. nippon try repeatedly to approach the courting pair, but they are always driven away by the dominant male. In the early stages of courtship, the mutual activity is interrupted easily by casual incidents, and the courting fish part from each other and return to their previous sites.

As the spawning time draws near, the circular movement of the gravid female becomes more rapid and she never descends to the bottom. Finally, the dominant male pushes the female up towards the surface, and spawning occurs simultaneously just below the surface of the water. The spawning pair are side-by-side at the time of actual gamete release. Just after the mutual spawning occurs, $1 \sim 3$ other males, who have stayed at a distance of 10~30 cm behind the dominant male, rush rapidly towards the two fish and release sperm almost simultaneously. Furthermore, 1~2 smaller males, who have stayed at a distance of 1~1.5 m behind the dominant male, sometimes follow the spawning group and also shed sperm immediately after the

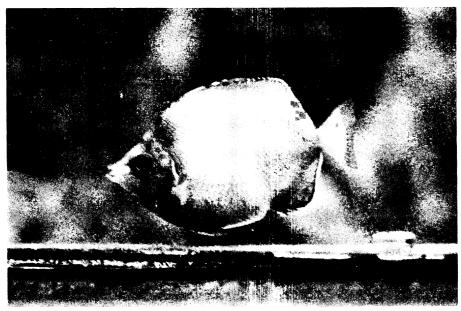


Fig. 2. Gravid female of C. nippon on the day of spawning.

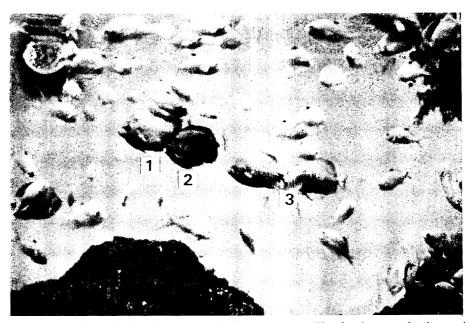


Fig. 3. Spawning behavior of *C. nippon* in the aquarium. The dominant male (2) nuzzles and pushes the belly of the gravid female (1) with his forehead. The other two males (3) follow the courting pair.

mutual spawning (Figs. 3, 4). The spawning of C. nippon takes place during $18:00\sim21:54\,\mathrm{hrs.}$ and occurs once a day except for a case on September, 11 (Table 1). After spawning, all the fish return separately to

their usual behavior. The spawning records of *C. nippon* in the aquarium are shown in Table 1.

Early life history. The fertilized eggs are buoyant, spherical, and colorless. They

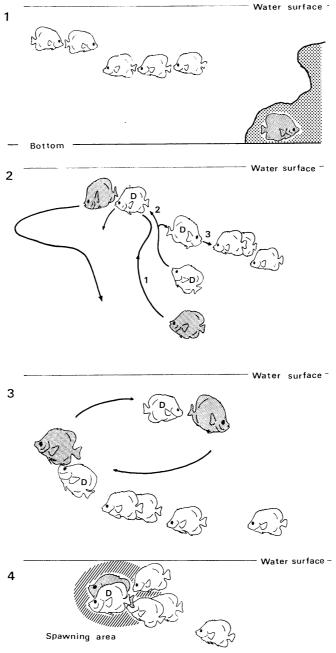


Fig. 4. Diagrammatic view of spawning behavior of *C. nippon* in the aquarium. 1. Before noon, males (white fish) swim separately or gather loosely together. A gravid female (shaded fish) stays at the bottom. 2. About 30 min. before spawning occurs, the female ascends to the surface of the water (1) and swims in circles. The dominant male (D) pursues the female and nuzzles her belly with his forehead (2). The other males try to approach the couple but they are driven out by the dominant male (3). 3. Just before spawning occurs, the female and the dominant male swim mutually in a circle with the male showing nuzzling and pushing behavior. The other males try to approach and follow them. 4. Mutual spawning occurs and the other males dash and shed sperm almost simultaneously.

Table 1. Record of spawning of C.

nippon in the aquarium

Date		Time of day	Number of adult fish	
			Present in aquarium	Fish spawned
1974 VIII	27	19:00	24	1 字 6 含含
1975 III	11	21:51	20	1 字 2 含含
	13	21:47	20	1 字 3 含含
	14	18:45	20	1 우 4 승승
IX	4	18:45	14	1 早 2 含含
	11	19:15	14	1 字 2 含含
	11	20:05	14	1 字 2 含含
	14	19:40	13	19 5 含含
	18	19:38	13	1 字 2 含含
	22	18:13	13	1 字 3 含含
	23	19:55	13	1 字 5 含含
X	2	19:11	12	1 字 2 含含
	10	18:08	12	1 字 5 含含
	10	19:20	12	1913

measure $0.70 \sim 0.74$ mm in diameter and contain a single oil globule measuring $0.18 \sim 0.19$ mm in diameter. The perivitelline space is narrow. No special structure is seen either at the surface of the egg-membrane or in the yolk. The developmental stages of the eggs and larvae are described from fertilized eggs which were taken on October 2, 1975.

The fertilized eggs reach the 2-cell stage in 55 min. (Fig. 5A), the morula stage in 4 hrs. 20 min. (Fig. 5B), the gastrula stage in 8 hrs. 10 min. after fertilization. At 11 hrs. 30 min. after fertilization, about 1/2 of the yolk is covered by the blastoderm and the embryonal body appears. At 12 hrs. 50 min., about 2/3 of the yolk is covered by the blastoderm and the embryonal body swells up from the surface of the yolk (Fig. 5C). At 16 hrs. 10 min., two myotomes, optic and Kupffer's vesicle can be seen. At 16 hrs. 50 min., the blastopore is closed. At 18 hrs. 30 min., 5 myotomes appear, punctate melanophore pigments can be seen along the dorsal side of the embryonal body, and numerous tubercles appear on the surface of the yolk (Fig. 5D). At 19 hrs. 10 min., $7 \sim 8$ myotomes, auditory vesicles and heart appear. At 23 hrs. 30 min., 15~16 myotomes can be seen, Kupffer's vesicle disappears, and lenses are formed in the eyes. Punctate melanophore pigments spread over the dorsal side of the embryo from the head

to the caudal part. At 26 hrs. $30 \, \text{min.}$, $24 \sim 25 \, \text{myotomes}$ can be seen. The heartbeat appears. Melanophore pigments increase in number and newly appear on the ventral side of the embryo (Fig. 5E). The embryo moves incessantly. At 28 hrs. $10 \, \text{min.}$, the first hatching takes place. About 95% of the larvae hatch out less than $1 \, \text{hr.} 40 \, \text{min.}$ after the first hatching, at water temperatures of $22.2 \sim 23.7 \, ^{\circ}\text{C.}$ No sterile eggs are found and hatching rate reaches about 95% or more.

The newly hatched larvae measure 1.43~ 1.53 mm in total length, and hold a large ovoid yolk of 0.95~0.98 mm in longest diameter. The front tip of the yolk extends beyond the snout of the larva. A single oil globule is situated at the tip of the yolk. The rear tip of the oil globule protrudes from the surface of the yolk. The number of myotomes is 12+12=24. The anus is situated at a point 2/3 of the total length from the head of the larva. Several melanophores lie on the snout, head, and lateral side of the body. Numerous tubercles are seen on the surface of the body (Fig. 5F). The newly hatched larvae float up-side-down just below the surface of the water in the receptacle.

Larvae 12 hours after hatching, measuring $1.63 \sim 1.83$ mm in total length, have 11+14=25 myotomes. The yolk sac somewhat degenerates in size. The front tip of the yolk is situated at an equal position to the snout. The caudal section of the body is elongated and the anus is situated a little behind the midpoint of the body. Branch-like melanophore pigments appear on the surface of the frontal part of the yolk and on the surface of the oil globule. Punctate xanthophore pigments appear above the eyes and lie along the body axis from behind the eyes to the ventro-caudal section of the body (Fig. 5G).

Larvae 24 hours after hatching, measuring $2.30 \sim 2.52$ mm in total length, have 11+14=25 myotomes. Xanthophores increase in number and spread over from the ventral side to the dorsal side of the body. Melanophores also increase in number and in size, and extend along each myotome (Fig. 5H).

Larvae 48 hours after hatching, measuring $2.46 \sim 2.68$ mm in total length, have 11+10=21

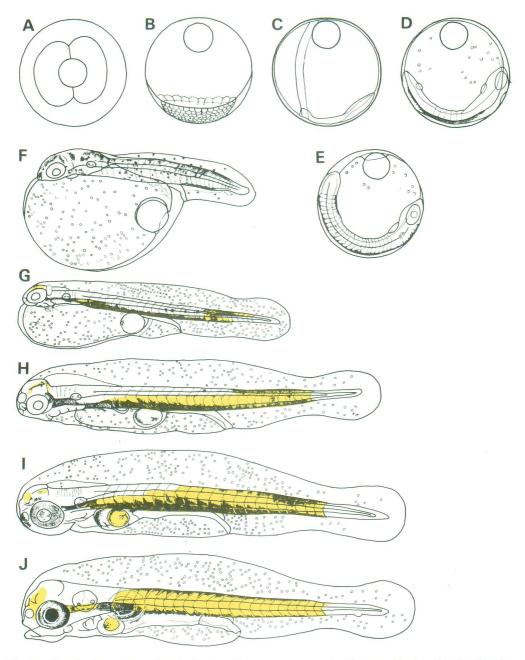


Fig. 5. Developing eggs and early larvae of C. nippon. A: 2-cell stage, 55 min. after fertilization. B: Morula stage, 4 hrs. 20 min. C: About 2/3 of the yolk is covered by blastoderm, 12 hrs. 50 min. D: 5-myotome stage, 18 hrs. 30 min. E: 24-25 myotome stage, 26 hrs. 30 min. F: Newly hatched larva, 28 hrs. 10 min. after fertilization. G: Larva, 12 hrs. after hatching. H: Larva, 24 hrs. I: Larva, 48 hrs. J: Larva, 72 hrs. Xanthophores are shown in yellow.

myotomes. The mouth has just formed, but is not open yet. Punctate xanthophores are newly seen on the surface of the oil globule. Other melanophore and xanthophore pigments increase in number and in size from the previous stage. They spread over the lateral side of the body (Fig. 51).

Larvae 72 hours after hatching, measuring $2.46 \sim 2.48$ mm in total length, i.e., the same size as the previous stage, have 11+10=21 myotomes. The mouth and anus are open. Vermiculation of the digestive organ can be seen. Melanophores decrease in number on the ventral and dorso-caudal section of the body, but are newly seen along the dorsal side of the central section of the body. Xanthophores spread remarkably over and cover almost all of the myotomes. Pectoral fins are formed and the larvae begin to swim actively (Fig. 5J).

Rearing of larval fish. Larvae of 3 days of age were fed on the rotifer, Brachionus plicatilis, larvae of the oyster, Crassostrea gigas, and larvae of the sea urchin, Temnopleurus reevesi. Although a few larval oysters were observed in the digestive organ of some larvae of C. nippon, the larval fish decreased gradually in number and the last one died at 8 days after hatching, with no remarkable growth in size nor change in shape from 3 days after hatching.

Discussion

Reproduction of C. nippon, at least under aquarium rearing conditions, involved one female and one to several males. In comparison with our observations, Lobel (1975, 1978), Moyer and Nakazono (1978), and Suzuki et al. (1979) reported "haremic" spawning in several angelfishes both from field and aquarium observations. The mode of reproduction in C. nippon is undoubtedly different from those of the angelfishes studied. Lobel (1978) briefly described the spawning behavior of three Hawaiian butterflyfishes, Chaetodon fremblii, C. unimaculatus, and C. multicinctus, under natural conditions. According to his observations, the reproduction of these butterflyfishes occurred between one male and one female, forming a courting pair. Although there is a difference in number of the mates, the courtship behavior of C. nippon bears resemblance to those of the Hawaiian butterflyfishes mentioned above. In the sea, reproduction of angelfishes and butterflyfishes seems to occur immediately at sunset and to be controlled by lunar cycles (Lobel, 1978, and others). However, reproduction of C. nippon always took place during early hours of the night without any suggestion of a lunar Aquarium rearing conditions may delay the spawning time and cycle of the butterflyfish. Although the spawning season of C. nippon is unknown, it is presumed to be from mid-summer to the end of fall when water temperatures range from about 23° to 27°C along the Pacific coast of central Japan. This range of coastal water temperatures fits in well with that in the aquarium when the spawning of C. nippon occurred.

Characteristics of the fertilized and developing eggs and early larvae of C. nippon bear a resemblance to those of angelfishes previously studied. But the larvae of the present species have a rather short body and yolk, whereas those of the angelfishes have a longer body and yolk (Fujita and Mito, 1960; Suzuki et al., 1979). An unique larval characteristic of C. nippon is the appearance of numerous xanthophores at $12 \sim 72$ hours after hatching. These pigments increase in number and in size accompanying larval development in this species, while no xanthophore has been observed in early larval stages until 72 hours after hatching in Genicanthus lamarck and G. semifasciatus (see Suzuki et al., 1979), and a few xanthophores appear as a passing phenomenon at 22 hours after hatching in Chaetodontoplus septentrionalis (see Fujita and Mito, 1960). It is well-known that the butterflyfishes have a distinctive tholichthys larval stage, however, the present authors could not find any indication nor rudiment of such a bony plate on the head in the early larval stages. The mode of reproduction and the larval shape of C. nippon are almost identical to those of the allied C. modestus observed by the present authors (in press).

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水槽内で観察されたシラコダイの産卵習性と卵および 仔魚

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水族館で $6\sim20$ 为月間飼育されたシラコダイで観察された産卵生態と初期生活史について報告する。産卵は 3 月, 8 月, 9 月及び 10 月に観察され、産卵時刻は 18 時 08 分 ~21 時 51 分、産卵時水温は 23°C またはそれ以上であった。本種の産卵は通常雌 1 尾と複数の雄とによってなされ、既知のキンチャクダイ類の産卵がハーレム内の雌雄 1 対でなされるのとは明らかに相違する。

受精卵は卵径 0.70~0.74 mm, 油球 1 個を有する無 色透明の球形分離浮性卵で、卵膜及び卵黄表面に特殊 な構造は見出せず, 卵膜腔は狭い. 水温 22.2~23.7°C で受精後28時間10分に孵化が始まる。孵化直後の仔 魚は全長 1.45~1.53 mm, 長卵形の卵黄 (長径 0.95~ 0.98 mm) の先端は頭部より前方へ突出する. 卵黄後 端に1個の油球 (径 0.18 mm) を有し、油球の後端は 卵黄表面より突出する. 孵化直後の仔魚の体形は既知 のキンチャクダイ類3種のそれに比べて短く全体に丸 味を帯び、近縁のゲンロクダイ (印刷中) のそれに似 る. 孵化 12 時間後に黄色素胞が出現し、孵化 72 時間 後まで仔魚の成長に伴って顕著に発達する.これはキ ンチャクダイでは孵化 22 時間後に一時的に黄色素胞 が発現し、タテジマヤッコでは孵化 72 時間後まで黄 色素胞が見出されなかったのとの顕著な相違点であ る.

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