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Sexual Pattern of the Labrid Fishes Collected from Kuchinoerabu-jima, Kagoshima, Japan

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The sexual pattern of the nine species of labrid fishes collected at the Kuchinoerabu-jima, Kagoshima Prefecture, Japan is described. Histological evidence shows that all nine species are protogynous hermaphrodites. These are: *Anampses geographicus, Thalassoma quinquevittata, T. purpureum, Stethojulis trilineata, Halichoeres scapularis, Hologymnosus annulatus, Xyrichtys dea, X. geisha, and Epibulus insidiator.* Of these, *T. purpureum, H. annulatus* are diandric. Diandry can not be confirmed in the remaining species because sufficient sample sizes of each species could not be collected at this site.

INTRODUCTION

Patterns of the sexuality in labrid fishes have been the subject of considerable interest over the last forty years, beginning with Reinboth's (1957) examination of the Mediterranean species *Colis julis*. He found structual difference in the testis among the males born as such and those drived by protogynous sex change.

These two types of males are called primary and secondary males, respectively. Thus, the presence of primary and secondary males within a single species is defined as diandry. Monandry is defined as the absence of primary males within a species.

Labrid fishes, most of which are believed to be protogynous, present a good example of the wide diversity of sexual and color patterns that exist among reef fishes. They occur in both temperate and tropical waters but most labrids are found in warm shallow seas. Labrids tend to spawn the year round, although seasonality in water temperature limits this activity at higher latitudes. Thus, aspects of behaviour and life history characteristics can be easily observed in the field by a diver.

Since the pioneering work on protogyny in tropical and subtropical labrids by Reinboth (1957, 1962, 1970, 1973), there have been two major efforts towards systematically examining the sexual patterns of many labroid (Labridae and Scaridae) species. Choat (1969) studied a large number of western Pacific (Great Barrier Reef) wrasses and parrot fishes. Roede (1972) detailed sex and coloration patterns in seven Caribbean labroids, but did not distinguish between primary and secondary males. Nakazono (1979) investigated the relations between gonadal sex change and body color in five Japanese labrids.

Despite a number of studies on sexuality of fishes, the sexual patterns of only a relatively few species have been determined. A powerful tool for determination of sexual

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pattern is the histological examination of a series of gonads (Sadovy and Shapiro, 1987). Unfortunately, many studies have qualified sex change by describing changes in a species color pattern or body size alone. Histological examination of the species' gonads remain to be made.

There are over 200 species of labrids in the Indo-Pacific region, yet only a few easily studied species have been examined. Thus, our knowledge of sexual patterns in this spacious group is limited. The purpose of the current study was to reveal the sexual patterns of a cross section of labrid species from different genera in an attempt to understand the general features of sexual patterns within the family.

MATERIALS AND METHODS

Specimens of eighteen species were collected at Honmura bay and Nishiura bay on the island of Kuchinoerabu-jima in Kagoshima Prefecture, Japan. Collections were made between 1988–1993, mainly during the months of July, August, October, and November by staff members of Hiroshima University. Angling and gill netting were the methods used during these collections. The specimens collected by staff members of the Hiroshima University composed of 110 specimens, of 18 species. Because some specimens had been preserved in formalin for variable length of time prior to histological examination, and their condition was poor, only nine species could be examined histologically. These were: Anampses geographicus, Thalassoma quinquevittata, T. purpureum, Stethojulis trilineata, Halichoeres scapularis, Hologymnosus annulatus, Xyrichtys dea, X. geisha and Epibulus insidiator.

After measurements of standard length (SL), total length (TL) to the nearest mm and body weight (BW) to the nearest 0.01g, specimens were dissected afterward by abdominal incision, and the gonads carefully extracted weighed to the nearest 0.01g (GW). Then, sketches of the external morphology of the gonads were made.

All gonads were examined histologically. At first, gonads were fixed in Bouin's solution for at least 24 hours. Then, gonads were dehydrated through a graded series of ethanol concentrations before being passed through ethanol- terpieol series or xylen. The gonads were embedded in Paraplast (E. Merck AG) and sectioned transversely with a microtome at a 5–10 micrometer thickness. Sections were stained with Mayers hematoxylin and eosin for observation (Humason, 1979).

Lager gonads were partitioned into anterioral, medial and posterioral portions, respectively. Smaller gonads were partitioned into two sections longitudinally on either side of the length of the gonad. Gonads were observed under a microscope (8-40x) to determine sex and phase.

RESULTS

The sexual pattern of all nine species examined was protogyny. Details for each species are as follow:

1. Anampses geographicus (Japanese name; Musibera)

This species is sexually dichromatic and maximum body size is 250 mm TL (Masuda

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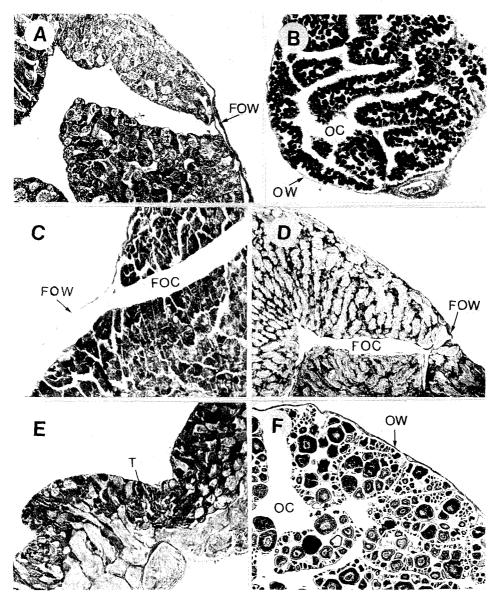


Fig. 1. Photomicrographs of labrid gonads. A: The secondary testis of *A. geographicus*, x 20. B: The ovary of *A. geographicus*, x 20. C: The secondary testis of *T. quinquevittata*, x 20. D: The secondary testis of *T. purpureum*, x 80. E: The primary testis of *T. purpureum*, x 20. F: The ovary of *T. purpureum*, x 8. FOC, former ovarian cavity. FOW, former ovarian wall. OC, ovarian cavity. OW, ovarian wall. T, testicular tissue.

Sex	GW(g)	BW(g)	TL(mm)	SL(mm)
SM	0.93	448.84	279	243
	#	360.26	257	231
SM	0.09	122.59	189	160
	#	15.69	100	87
SM	0.07	8.85	84	72

 Table 1. Length, weight and sexual pattern of Anampses geographicus.

 (#: gonad was not fixed and could not be subjected to histological examination, SM : secondary male)

et al., 1975). Five specimens were examined here, ranging from 72-243 mm SL. Three specimens were male and two undetermined, the latter because of the poor condition of the examined gonads (Table 1). All three males had secondary testes. The secondary structure of *A. geographicus* (Fig. 1-A) was fundamentally the same as that of many other labrids, having a large lumen enclosed by thin membrane corresponding to the former ovarian wall (Reinboth, 1962, 1970; Warner and Robertson, 1978). Ovary structure was similar to that of many other teleosts. Here, young oocytes filled the ovarian cavity (Fig. 1-B). Thus this species is protogynous.

2. *Thalassoma quinquevittata* (Japanese name; Hakobera)

This species reaches a maximum lengh of 150 mm TL (Masuda *et al.*, 1975). Three specimens were examined, ranging in size from 93–119 mm SL. Two specimens were males; the third could not be determined (Table 2). Both males had secondary testes with typical structure and a very thin former ovarian wall (Fig. 1-C). This species is protogynous.

Table 2. Length, weight and sexual pattern of *Thalassoma quinquevittata*(#: gonad was not fixed and could not be subjected to histological
examination, SM : secondary male)

Sex	GW(g)	BW(g)	TL(mm)	SL(mm)
SM	0.22	42.83	136	119
SM	0.18	37.03	131	113
	#	21.37	111	93

3. *Thalassoma purpureum* (Japanese name; Kinubera)

This species is sexually dichromatic and reaches maximum length of 300 mm TL (Masuda *et al.*, 1975). Twenty three specimens were examined, ranging in size from 85–219 mm SL. Ovaries were found in 18 specimens (85–219 mm SL), and testes in five (153–219 mm SL), of which three were secondary males, one a primary male, and one undetermined (Table 3). Secondary testes had typical secondary structures (Fig. 1-D). The primary testis, however, had neither the inner lumen nor the membrane surounding the testicular tissue typical of a secondary testis. Large vasa deferentia were observed in

	i: male)	iale, F: lemale, I		secondary male	(5M:
	Sex	GW(g)	BW(g)	TL(mm)	SL(mm)
	SM	0.32	264.82	250	219
	F	1.26	190.8	215	189
	F	0.62	293.06	253	219
	PM	0.27	161.18	212	186
	F	2.18	154.72	199	169
	\mathbf{F}	4.07	98.25	189	165
	F	0.64	72.74	172	147
	F	1.62	65.21	161	141
	F	1.71	52.12	149	131
1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	F	7.32	179.23	207	181
	F	5.2	244.73	247	206
	F	0.77	251.18	235	205
	F	2.6	175.82	207	185
	SM	0.78	186.82	221	192
	М	0.38	184.01	217	186
	\mathbf{F}	2.01	91.92	173	150
	SM	0.17	102.47	177	153
	\mathbf{F}	1.89	87.64	164	142
	F	4.7	81.05	160	140
	F	1.56	73.26	161	138
	F	0.47	63.54	153	128
1994 - 1997 1994 - 1997	F	0.34	51.86	146	125
	F	0.08	11.69	100	85

Table 3. Length, weight and sexual pattern of *Thalassoma purpureum*.

 (SM: secondary male, PM: primary male, F: female, M: male)

the central portion of the primary testis (Fig. 1-E). The ovary of *T. purpureum* showed a typical pattern, containing many oocytes whose development ranged from oogonial to perinucleolus stage (Fig. 1-F). This species is diandric and protogynous.

4. *Stethojulis trilineata* (Japanese name; Onibera)

This species is sexually dichromatic and reaches a maximum size of 150 mm TL (Masuda *et al.*, 1975). Seven specimens, ranging from 79–123 mm SL, were examined. Four specimens were female (all were smaller than 102 mm SL) and three specimens were male (Table 4). The males all had secondary testes, typical in structure, with thin former ovarian walls and former ovarian cavities (Fig. 2-A). This species is protogynous.

SL(mm)	TL(mm)	BW(g)	GW(g)	Sex
123	146	65.44	0.2	SM
120	148	60.11	0.15	SM
107	130	38.84	0.09	SM
102	125	35.98	1.22	\mathbf{F}
99	117	29.78	1.74	F
92	111	22.19	0.1	F
79	96	15.44	0.33	\mathbf{F}

 Table 4. Length, weight and sexual pattern of Stethojulis trilineata.

 (SM: secondary male, F: female)

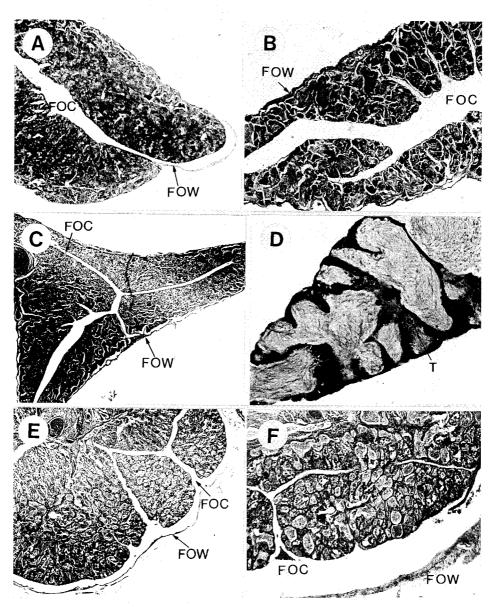


Fig. 2. Photomicrographs of labrid gonads. A: The secondary testis of *S. trilineata*, x 8. B: The secondary testis of *H. scapularis*, x 20. C: The secondary testis of *H. annulatus*, x 8. D: The primary testis of *H. annulatus*, x 40. E: The secondary testis of *X. dea*, x 8. F: The secondary testis of *X. geisha*, x 20. FOC, former ovarian cavity. FOW, former ovarian wall. OC, ovarian cavity. OW, ovarian wall. T, testicular tissue.

5. *Halichoeres scapularis* (Japanese name; Seitenbera)

This species is rare in Japanese waters, reaching a lengh of 150 mm TL (Masuda *et al.*, 1975). Four specimens, ranging from 83-137 mm SL, were examined. Two specimens were female, one male and the last undetermined (Table 5). The male had a typical secondary testis with a large lumen and former ovarian wall, and a former ovarian cavity (Fig. 2-B). This species is protogynous.

SL(mm)	TL(mm)	BW(g)	GW(g)	Sex
137	165	77.09	0.34	SM
123	146	57.93	1.1	F
113	135	34.19	#	
83	101	14.15	0.37	F

Table 5. Length, weight and sexual pattern of Halichoeres scapularis.

 (#: gonad was not fixed and could not be subjected to histological examination, SM: secondary male, F: female)

6. Hologymnosus annulatus (Japanese name; Namerabera)

This species displays changes in color pattern with growth, reaching a maximum size of 400 mm TL (Masuda *et al.*, 1975). Thirteen specimens, ranging from 131–277 mm SL, were examined. Eight specimens were female (192–275 mm SL), three male and two not determined (Table 6). One male had a typical secondary testis. The other male had a primary testis, in which large vasa deferentia were seen in the upper left-hand portion (Fig. 2-C). This testis lacked an inner lumen (Fig. 2-D). This species is diandric and protogynous.

Table 6. Length, weight and sexual pattern of *Hologymnosus annulatus*.
(#: gonad was not fixed and could not be subjected to histological examination, SM: secondary male, PM: primary male, F: female, M: male but not defined as primary or secondary male, U: undetermined)

SL(mm)	TL(mm)	BW(g)	GW(g)	Sex
277	320	326.65	0.84	SM
275	315	290.05	9.05	F
229	261	154.58	1.03	U
272	305	264.64	3.1	F
251	283	218.15	3.42	\mathbf{F}
231	258	181.31	2.95	F
254	291	221.12	4.41	\mathbf{F}
229	256	137.46	1.04	F
251	284	121.79	0.29	PM
222	251	147.6	0.89	\mathbf{F}
192	211	77.98	0.24	F
189	209	79.01	#	
131	151	23.89	0.03	М

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7. Xyrichtys dea (Japanese name; Tensu)

This species reaches 300 mm TL (Masuda *et al.*, 1975). Three specimens, ranging from 143–267 mm SL, were examined. Two specimens were male (Table 7) and had typical secondary testes with a large lumen and former ovarian wall, and a former ovarian cavity; large vasa deferentia were also seen in the central portion (Fig. 2-E). Sex of the third specimen could not be defined. This species is protogynous.

8. Xyrichtys geisha (Japanese name; Kurobuchitensu)

Only one specimen could be examined, a male of 148 mm SL. The testis was secondary (Table 8), with a typical secondary structure including a thick former ovarian wall (Fig. 2-F). This species is protogynous.

9. *Epibulus insidiator* (Japanese name; Gichibera)

This species is polychromatic and maximum body size is 350 mm TL (Masuda *et al.*, 1975). A single specimen was examined, a male of 225 mm SL. The testis was secondary (Table 9), with a brown body and a large lumen (Fig. 3). This species is protogynous.

(SM	secondary mal	e, U: undetermi	ned)	
SL(mm)	TL(mm)	BW(g)	GW(g)	

Table 7. Length, weight and sexual pattern of *Xyrichtys dea*.

SL(mm)	TL(mm)	BW(g)	GW(g)	Sex
267	300	550.96	0.6	SM
149	171	69.72	0.27	SM
143	165	69.01	0.15	U

 Table 8. Length, weight and sexual pattern of Xyrichtys geisha.

 (SM: secondary male)

SL(mm)	TL(mm)	BW(g)	GW(g)	Sex
148	173	81.67	0.37	SM

Table 9. Length, weight and sexual pattern of *Epibulus insidiator*.(SM: secondary male)

SL(mm)	TL(mm)	BW(g)	GW(g)	Sex
225	291	357.63	0.47	SM

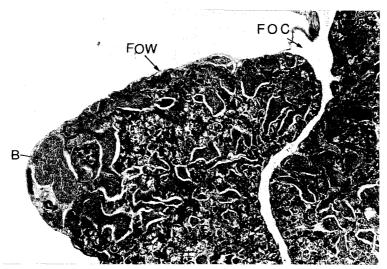


Fig. 3. Photomicrographs of the secondary testis of *E. insidiator*, x 20. FOC, former ovarian cavity. FOW, former ovarian wall. B, brown body.

DISCUSSION

Histological examination of gonads remains a powerful method for determining whether the fish is a hermaphrodite or not (Sadovy and Shapiro, 1987). During protogynous sex change, ovarian tissues degenerate leaving only the ovarian wall, while testicular tissues develop from the former ovarian lamellae. Thus, secondary testes transformed from ovaries possess a structure corresponding to a former ovarian wall and an ovarian lumen (Reinboth, 1962). This structure is not seen in primary testes, which arise during ontogenic development. The secondary testes examined here were fundamentally similar to those of other sex-changing labrid species previously examined (Reinboth, 1962,1970; Warner and Robertson, 1978). The presence of brown bodies, as observed in the testicular lobes of *Xyrichtys geisha* (Fig. 3), can not be adopted as an evidence for sex change, however (Sadovy and Shapiro, 1987).

The nine labrid species examined here are all protogynous hermaphrodites (Table 10). Two of these, *Hologymnosus annulatus* and *Thalassoma purpureum*, are diandric as well. The remaining seven species could not be identified as either diandric or monandric. To reveal that, histological examination of gonads from smaller sized specimens is needed.

Of the tropical labrid species examined thus far, all are protogynous hermaphrodite with secondary males drived from functional females. This trait does not extend throughout the family, however (Thresher, 1984). Two species of temperate wrasse, *Oxyjulis califonica* (Diener, 1976) and *Crenilabrus melops* (Dipper and Pullin, 1979) are not hermaphroditic.

The results of this study indicate that most labrid species are probably hermaphroditic. Additional data will likely confirm this prediction. For example, 14 of 15

Species	Sexua	al pattern
	Monandry	Diandry
Anampses geographicus	yes	?
Thalassoma quinquevittata	yes	?
T. purpureum	50°	yes
Stethojulis trilineata	yes	?
Halichoeres scapularis	yes	?
Hologymnosus annulatus		yes
Xyrichtys dea	yes	?
X. geisha	yes	?
Epibulus insidiaitor	yes	?

Table 10. Summary of the sexual patterns of nine species of wrasses

specimens of Anampses caeruleopunctatus collected at Kuchinoerabu-jima, were female, and their body sizes ranged from 52–237 mm TL; the sex of the 15th specimen (52 mm TL) could not be determined (Leem *et al.*, unpublished data). Masuda *et al.* (1975) reported a maximum body size for this species of 300 mm TL. Given this disparity in size range, protogynous hermaphroditism is indicated for this species too, but can only be confirmed by histological examination of gonads from larger specimens (Sadovy and Shapiro, 1987). Histological examination of the gonads from a wide body size range in other species is necessary to determine the level of prevalence of protogynous hermaphroditism in this highly diverse family of fishes.

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