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A Chromosome Study on Thirteen Species of Japanese Gobiid Fishes

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Abstract Somatic chromosomes in 13 species of Japanese gobies were examined. Cells from gills and digestive tracts were used after treatment with colchicine. The preparations were made according to the air-drying method and stained with Giemsa.

Diploid chromosome numbers of the following 13 species were 40 to 48, and two groups were separated on the basis of chromosome morphology, i.e., (1) the group in which all chromosomes were subtelocentrics or acrocentrics: *Aboma lactipes*, $2n=40$, *Acanthogobius flavimanus*, $2n=44$, *Rhinogobius brunneus*, $2n=44$, *Rhinogobius flumineus*, $2n=44$, *Chasmichthys dolichognathus*, $2n=44$, *Mugilogobius abei*, $2n=46$, *Bathygobius fuscus*, $2n=48$, and (2) the group in which chromosomes consisted of metacentrics or submetacentrics and subtelocentrics or acrocentrics: *Pterogobius elapoides*, $2n=44$, *Luciogobius guttatus*, $2n=44$, *Tridentiger obscurus brevispinis*, $2n=44$, *Tridentiger trigonocephalus*, $2n=44$, *Chaenogobius annularis*, $2n=44$, *Periophthalmus cantonensis*, $2n=46$.

Gobiid fishes of more than 80 species have been reported from Japan (Tomiyama, 1936), and their comparative anatomy has been studied by many workers: the glossohyal bone (Takagi, 1950), the scale (Takagi, 1953), the sensory system organs on the head, vertical fins, the ventral fin (Takagi, 1963), the osteology of the head and the scapula (Prince Akihito, 1967, 1969, 1971), and sex characters (Egami, 1960; Arai, 1964). The classification of gobies, however, is not always in agreement among ichthyologists.

Recently, karyological approach in fish systematics has been considered to be useful: e.g., in trouts of western North America (Miller, 1972), in cyprinodontids, *Fundulus olivaceus* and *F. notatus* (Setzer, 1970), and the viviparous fish *Poeciliopsis* (Schultz, 1967). For the family Gobiidae, cytogenetic data are too meager to discuss their interrelationships, i.e., only 12 species have been reported in the world (Nogusa, 1960; Post, 1965; Chen and Ebeling, 1971). This study was performed to increase chromosome data of the family Gobiidae in Japan. Preliminary counts of diploid chromosome numbers in 13 species of the Gobiidae are reported here.

Materials and methods

The following is a list of 13 species studied, which are arranged in the order of Takagi's classification (1963). Number of specimens examined, their range of total length, and the sampling place are followed after each scientific name.

Gobiidae

Subfamily Periophthalminae

Periophthalmus cantonensis (Osbeck). 2, 73–79mm, from Gyotoku, river mouth of Edogawa River, Chiba Pref.

Subfamily Tridentigerinae

Tridentiger obscurus brevispinis Katsuyama, Arai, and Nakamura. 3, 65–70 mm, from Kihara, Lake Kasumigaura, Ibaraki Pref.

Tridentiger trigonocephalus (Gill). 3, 57–71 mm, from Kariyagasaki, Miura Pen., Kanagawa Pref.

Subfamily Rhinogobiinae

Rhinogobius brunneus (Temminck and Schlegel). 6, 76–91 mm, from Ōkawa River, Itō City, Shizuoka Pref. and 6, 33–44 mm, from Imperial Palace, Akasaka, Tokyo.

Rhinogobius flumineus (Mizuno). 6, 57–62mm, from Akuta River, Osaka.

Chasmichthys dolichognathus (Hilgendorf). 2, 55–56 mm, from Arasaki, Miura Pen., Kanagawa Pref.

Mugilogobius abei (Jordan and Snyder). 2, 47–52 mm, from Hioi-machi, Wake-gun, Okayama Pref.

Aboma lactipes (Hilgendorf). 6, 55–76 mm, from Lake Kasumigaura.

Acanthogobius flavimanus (Temminck and Schlegel). 6, 72–122 mm, from Hanaizumi River, Oiso, Kanagawa Pref. and Kariyagasaki, Miura Pen., Kanagawa Pref.

Pterogobius elapoides (Günther). 2, 94–95 mm, from Niemon I., Awa, Chiba Pref.

Chaenogobius annularis Gill [=*C. urotaenia* (Hilgendorf)]. 2, 56–86 mm, from Hayakawa River, Odawara City, Kanagawa Pref.

Luciogobius guttatus Gill. 2, 58–60 mm, Arasaki, Miura Pen., Kanagawa Pref.

Subfamily Gobiinae

Bathygobius fuscus (Rüppell). 2, 63–67 mm, from Arasaki, Miura Pen., Kanagawa Pref.

Several characteristics of these species are shown in Table 1.

Collected fish were kept in a well aerated aquarium of 58×40×35 cm in size, and marine species were fed with the carpet shell, *Palourdes japonaises*, and aquatic oligochaetes of the family Tubificidae were given to freshwater species.

Each fish was injected with 0.03 to 0.15 ml of 0.1% colchicine solution, and kept for 3 to 4.5 hours at room temperature after the injection. The injected fish were killed by decapitation. The gill and the digestive tract were removed and minced with scissors, treated with 0.9% sodium citrate hypotonic solution for 20 minutes at room temperature, and then, fixed with methyl alcohol and acetic acid (3:1, v/v). Free cells were separated by centrifuging at 900rpm for 5 minutes. Chromosome slides were made according to the air-drying method and stained with Giemsa solution. Chromosomes were counted using microphotos.

Classification of chromosomes is referred to Levan et al. (1964). In the present report, metacentric and submetacentric chromosomes are described as biarms, and subtelocentric and acrocentric chromosomes described as monoarms.

All specimens used for the experiments are deposited in the fish collection of National Science Museum, Tokyo.

Results and discussion

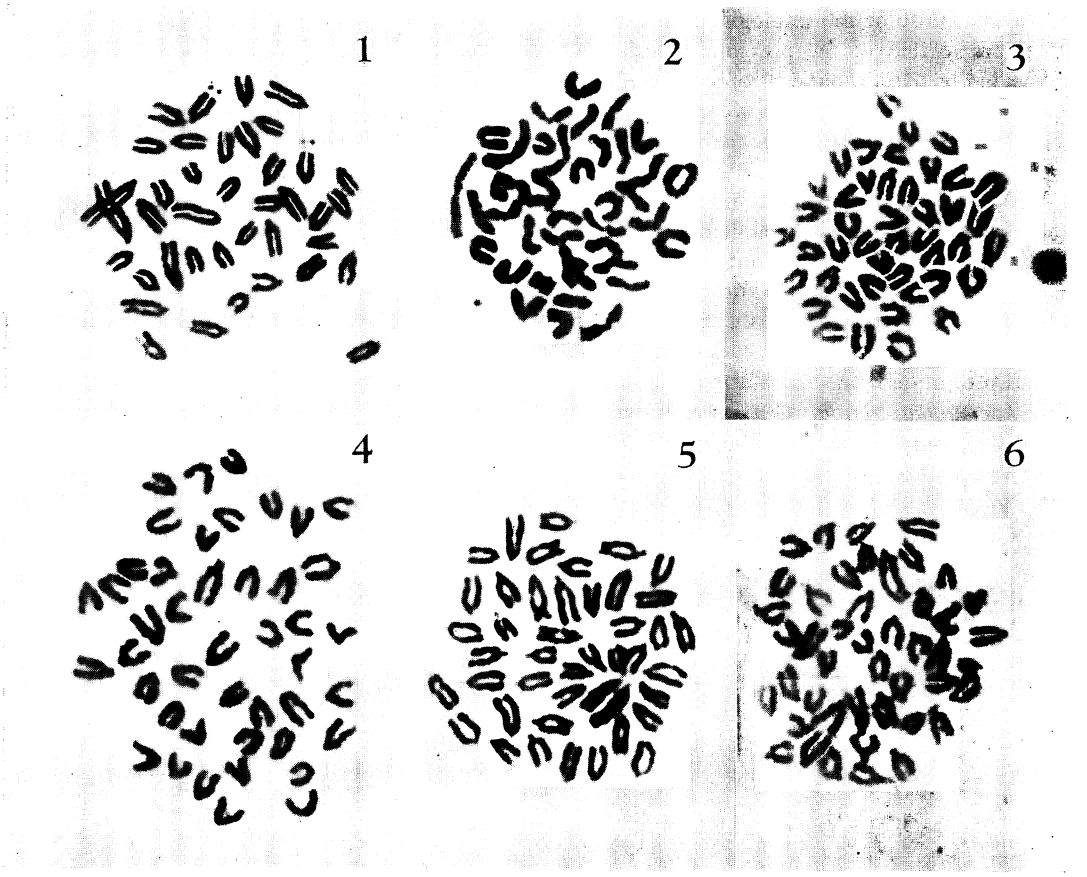
(1) *Aboma lactipes* (Hilgendorf)
"Ashishiro-haze"

Aboma lactipes had $2n=40$ chromosomes, and all chromosomes were monoarms (Figs. 1 and 2). As shown in Fig. 1, a pair of chromosomes with two satellite-chromosomes at both ends of long arms are found. As far as authors are

Table 1. Diploid chromosome numbers and some characters of 13 species of Japanese gobiid fishes.

Species	2*n*	Chromosome forms*	VN	D.	A.
Subfamily Rhinogobiinae					
Aboma lactipes	40	mono	31～32	VIII-I, 10	I, 9～10
Acanthogobius flavimanus	44	mono	33～34	VIII-I, 13	I, 11～12
*Rhinogobius brunneus*** (from Itō)	44	mono	26	VI-I, 8	I, 8
(from Tokyo)	44	mono	26	VI-I, 8～9	I, 8～9
Rhinogobius flumineus	44	mono	27～28	VI-I, 7～8	I, 6～7
Chasmichthys dolichognathus	44	mono	32	VI-I, 10	I, 9
Pterogobius elapoides	44	mono+bi	34	VI-I, 21	I, 19～20
Luciogobius guttatus	44	mono+bi	36	I, 10～11	I, 11～13
Chaenogobius annularis	44	mono+bi	33	VI-I, 10～11	I, 10～11
Mugilogobius abei	46	mono	25	VI-I, 8	I, 8
Subfamily Tridentigerinae					
Tridentiger obscurus brevispinis	44	mono+bi	26	VI-I, 9～11	I, 10
Tridentiger trigonocephalus	44	mono+bi	26	VI-I, 11～12	I, 10
Subfamily Periophthalminae					
Periophthalmus cantonensis	46	mono+bi	26	XII-I, 11	I, 11
Subfamily Gobiinae					
Bathygobius fuscus	48	mono	27	VI-I, 9	I, 8

* bi, biarm chromosome=metacentric+submetacentric; mono, monoarm=subtelocentric+acrocentric.
** As for two forms of *R. brunneus*, see text.



Figs. 1-6. Somatic metaphase figures of Japanese gobies. ×1740.
Figs. 1 and 2. *Aboma lactipes*: Fig. 3. *Acanthogobius flavimanus*: Fig. 4. *Rhinogobius brunneus* (from Itō City): Fig. 5. *Rhinogobius brunneus* (from Tokyo): Fig. 6. *Rhinogobius flumineus*.

aware, this type of chromosomes has not been reported among fishes. It might be the characteristic chromosomes of *A. lactipes*. The diploid chromosome number of the species is the fewest in the Gobiidae.

(2) *Acanthogobius flavimanus* (Temminck and Schlegel) "Ma-haze"

The diploid chromosome number was 44, and all chromosomes were monoarms (Fig. 3). There was found no difference in chromosome number and morphology between specimens from Hanaizumi River and from Kariyagasaki.

(3) *Rhinogobius brunneus* (Temminck and Schlegel) "Yoshinobori"

Itō and Mizuno (1972) devided this species into three types, i.e., Ō-mon Type, Kokushoku Type, and Large Kokushoku Type.

In the present study, two forms of *R. brunneus* were examined. The form from the upper stream of Ō-kawa River, Itō City, which lived with "Amago", the land-locked form of *Oncorhynchus rhodurus*, seems to be similar to Large Kokushoku Type. The form from the moat of the Imperial Palace resembles to Ō-mon Type, but differs from Ō-mon Type by absence of dark spots on pectoral fin base. Chromosomes of the gill and the digestive tract were examined.

Both forms had the same diploid chromosome number of 44, which were all monoarms (Figs. 4 and 5). This species showed no noticeable karyotype variation among preparations, individuals or tissues. The result agrees well with that of Nogusa (1950).

(4) *Rhinogobius flumineus* (Mizuno) "Kawa-yoshinobori"

This species is morphologically very similar to *R. brunneus* (Mizuno, 1960a, 1960b; Nakamura, 1963).

The chromosome morphology of this species resembles to that of the previous species. As shown in Fig. 6, the diploid chromosome number was 44, and all chromosomes were monoarms.

(5) *Chasmichthys dolichognathus* (Hilgendorf) "Ago-haze"

The diploid chromosome number was 44, and all chromosomes were monoarms (Fig. 7).

(6) *Pterogobius elapoides* (Günther) "Kinubari"

The diploid chromosome number was 44. Differing from the previously described five species, this species had two types of chromosomes, i.e., monoarms and biarms (Fig. 8).

(7) *Luciogobius guttatus* Gill "Mimizu-haze"

The diploid chromosome number was 44, which consisted of monoarms and biarms (Fig. 9).

(8) *Chaenogobius annularis* Gill "Ukigori"

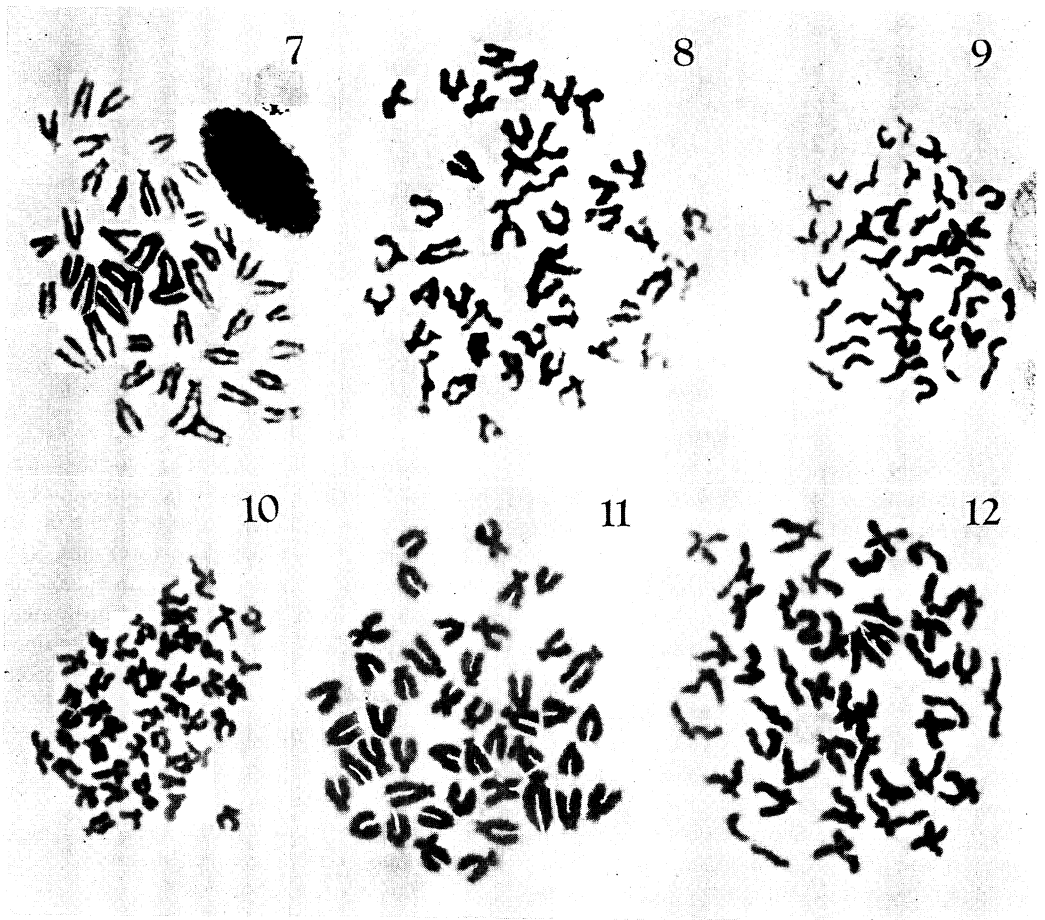
Following Takagi (1966), *Chaenogobius urotaenia* is considered as a synonym of *C. annularis* in this report.

The diploid chromosome number was 44, most of which were biarms (Fig. 10). Although Nogusa (1957) reported one pair of small-sized chromosomes, such chromosomes were not found in the present material.

(9) *Mugilogobius abei* (Jordan and Snyder) "Abe-haze"

The diploid chromosome number was 46, and all chromosomes were monoarms (Fig. 13). The chromosome number agrees with that of Nogusa (1957), but one pair of small-sized chromosomes described in his report were not found.

(10) *Tridentiger obscurus brevispinis* Katsuyama, Arai, and Nakamura "Numa-chichibu"

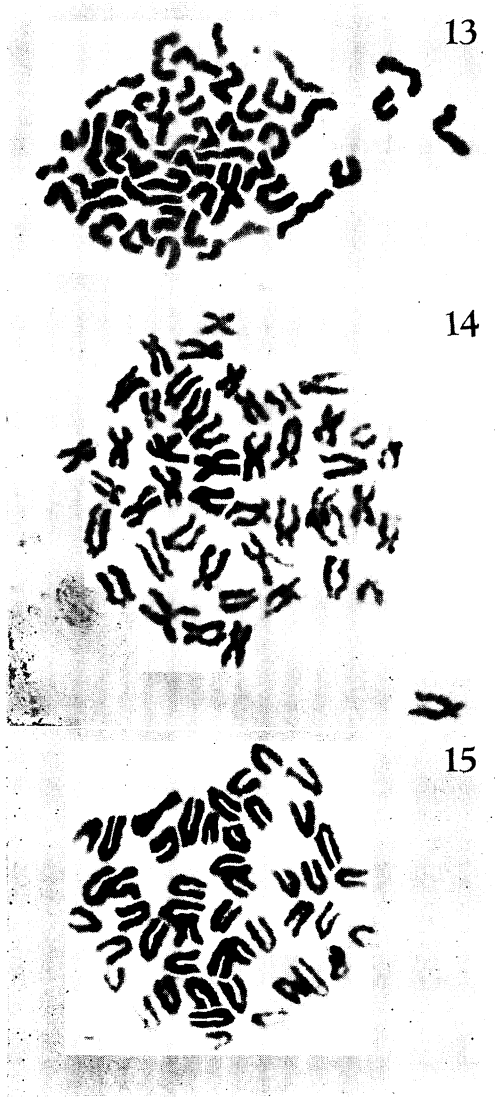


Figs. 7–12. Somatic metaphase figures of Japanese gobies. ×1800.
Fig. 7. *Chasmichthys dolichognathus*: Fig. 8. *Pterogobius elapoides*: Fig. 9. *Luciogobius guttatus*: Fig. 10. *Chaenogobius annularis*: Fig. 11. *Tridentiger obscurus brevispinis*: Fig. 12. *Tridentiger trigonocephalus*.

This subspecies differs from *T. obscurus obscurus* by its long first dorsal fin and the smaller number of gill-rakers. *T. o. brevispinis* lives in freshwater (Katsuyama et al., 1972).

The diploid chromosome number was 44, which contained monoarms and biarms (Fig. 11). The chromosome number accords with that of *Tridentiger obscurus* reported by Nogusa (1950), though two pairs of small-sized chromosomes were not found.

(11) *Tridentiger trigonocephalus* (Gill)



Figs. 13-15. Somatic metaphase figures of Japanese gobies. ×1610.
Fig. 13. *Mugilogobius abei*: Fig. 14. *Periophthalmus cantonensis*: Fig. 15. *Bathygobius fuscus*.

"Shima-haze"

The diploid chromosome number was 44, which consisted of monoarms and biarms (Fig. 12).

(12) *Periophthalmus cantonensis* (Osbeck)
"Tobi-haze"

The diploid chromosome number was 46, which consisted of biarms and monoarms (Fig. 14). This species had the same chromosome number with *Mugilogobius abei*, but differed in chromosome morphology. The chromosome number of *P. cantonensis* agrees with that reported by Nogusa (1957), but three pairs of remarkably small-sized chromosomes were not found.

As for the mudskipper, it was reported that two species of American mudskippers, *Gillichthys seta* and *G. mirabilis* had 44 chromosomes, respectively (Chen and Ebeling, 1971).

(13) *Bathygobius fuscus* (Rüppell)
"Kumo-haze"

The diploid chromosome number was 48, and all chromosomes were monoarms (Fig. 15). This number, $2n=48$, was first reported among Japanese gobies. Forty-eight chromosomes were reported in the bumblebee fish, *Brachygobius nunus* (Post, 1965).

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日本産ハゼ科魚類13種の核型について

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日本産ハゼ科魚類13種類の核型をコルヒチン処理した魚の鰓と消化管で観察した．体細胞の染色体数は $2n=40$, $2n=44$, $2n=46$, $2n=48$ の4型で，各染色体の形態については，(1) すべての染色体が subtelocentrics もしくは acrocentrics のものと，(2) metacentrics, submetacentrics および subtelocentrics もしくは acrocentrics からなるものとが観察された．

(1) のグループに属するものは，アシシロハゼ *Aboma lactipes* (Hilgendorf), $2n=40$, マハゼ *Acanthogobius flavimanus* (Temminck et Schlegel), $2n=44$, ヨシノボリ *Rhinogobius brunneus* (Temminck et Schlegel), $2n=44$, カワヨシノボリ *Rhinogobius flumineus* (Mizuno), $2n=44$, アゴハゼ *Chasmichthys dolichognathus* (Hilgendorf), $2n=44$, アベハゼ *Mugilogobius abei* (Jordan et Snyder), $2n=46$, クモハゼ *Bathygobius fuscus* (Rüppell), $2n=48$ の7種類で，(2) のグループに属するハゼは，キヌバリ *Pterogobius elapoides* (Günther), $2n=44$, ミミズハゼ *Luciogobius guttatus* Gill, $2n=44$, ヌマチチブ *Tridentiger obscurus brevispinis* Katsuyama, Arai et Nakamura, $2n=44$, シマハゼ *Tridentiger trigonocephalus* (Gill), $2n=44$, ウキゴリ *Chaenogobius annularis* Gill, $2n=44$, トビハゼ *Periophthalmus cantonensis* (Osbeck), $2n=46$ の6種類であった．

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