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Karyotypes of Five *Puntius* Species and One *Cyclocheilichthys* Species (Pisces, Cyprinidae) from Thailand

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Abstract Chromosomes of five species of *Puntius* and a species of *Cyclocheilichthys* from Thailand were observed. Karyotypes of six species are as follows. *Puntius daruphani*: 2n=50, NF=70; *Puntius stoliczkanus*: 2n=50, NF=94; *Puntius gonionotus*: 2n=50, NF=72; *Puntius altus*: 2n=50, NF=84; *Puntius sophoroides*: 2n=50, NF=54; *Cyclocheilichthys apogon*: 2n=50, NF=70. From viewpoint of karyology and morphology, classification of *Puntius* is discussed. The karyotype of *Cyclocheilichthys apogon* is the first report in *Cyclocheilichthys* species.

The classification of *Puntius* HAMILTON, 1822, is obscure. *Puntius* sensu lato comprises *Puntius* sensu stricto, *Capoeta* VALENCIENNES, 1842, and *Barbodes* BLEEKER, 1859, and these three genera are defined on the basis of the number of barbels, i. e., four barbels=*Barbodes*, two barbels=*Capoeta*, and no barbels=*Puntius*. Some workers adopt only *Puntius* sensu lato and criticize the classification of three genera, *Puntius*, *Capoeta*, and *Barbodes* (TAKI *et al.*, 1978). Others adopt these three genera, but their interrelationships have not been known (WU *et al.*, 1977).

On the other hand, karyological analysis of fishes has become important for fish systematics (ARAI, 1982; ARAI & AKAI, 1988). Then we carried out experiments for chromosome observation of five *Puntius* species and one *Cyclocheilichthys* species from Thailand. The results will be reported here.

Materials and Method

Two specimens of *Puntius daruphani* SMITH, 1934, 94.0 and 140.0 mm TL, five specimens of *P. gonionotus* (BLEEKER, 1850), 72.5 to 94.2 mm TL, six specimens of *P. altus* (GÜNTHER, 1868) 52.5 to 95.0 mm TL, two specimens of *P. sophoroides* (GÜNTHER, 1868), 43.5 and 87.9 mm TL, and two specimens of *Cyclocheilichthys apogon* (VALENCIENNES, 1842), 65.0 and 67.7 mm TL, were collected from Ayutthaya

Table 1. Morphological characters of material fishes.

Species	No. of fish	Standard length (mm)	Barbels	Lateral line scales	Branched dorsal fin rays	Branched anal fin rays	Vertebrae
Puntius daruphani	2	73.0–100.3	4	28	8	5	35
P. stoliczkanus	2	51.5– 53.5	0	22	8	5	30
P. gonionotus	5	58.0– 73.5	4	29–31	8	6	34
P. altus	6	41.7– 72.0	4	31–32	8	5	31–32
P. sophoroides	2	33.0– 66.1	2	25	8	5	30–31
Cyclocheilichthys apogon	2	51.3– 52.5	0	33	8	5	33

Province, Central Thailand. Two specimens of *Puntius stoliczkanus* (DAY, 1869), 66.6 and 67.0 mm TL, were caught in Mae Hong Son Province, North Thailand. Some morphological characters of material fishes are shown in Table 1.

Chromosome preparation was made from the head kidney following the method of OJIMA and KURISHITA (1980). The classification of chromosomes is adopted from LEVAN *et al.* (1964). Metacentrics and submetacentrics are described as two-arm chromosomes, and subtelocentrics and acrocentrics as one-arm chromosomes.

Results

Puntius daruphani (Fig. 1, A and C). As shown in Table 2, the diploid chromosome number is 50. The karyotype comprises 12 metacentric, 8 submetacentric, 6 subtelocentric, and 24 acrocentric chromosomes. The arm number is 70.

Puntius stoliczkanus (Fig. 1, B and D). The diploid chromosome number is 50. The karyotype comprises 22 metacentric, 22 submetacentric, 4 subtelocentric, and 2 acrocentric chromosomes. The arm number is 94.

Puntius gonionotus (Fig. 2, A and C). The diploid chromosome number is 50. The karyotype of this species comprises 2 metacentric, 20 submetacentric, 4 subtelocentric, and 24 acrocentric chromosomes. The arm number is 72.

Puntius altus (Fig. 2, B and D). The diploid chromosome number is 50. The karyotype comprises 10 metacentric, 24 submetacentric, 4 subtelocentric, and 12

Table 2. Frequency distributions of diploid chromosome counts in six species of material fishes.

Species	2n									Total
	44	45	46	47	48	49	50	51	52	
Puntius daruphani	3		6		15	3	33		2	62
P. stoliczkanus			5		19		30	3	2	59
P. gonionotus	3		4		14		42		3	66
P. altus			3		12		35	3	1	54
P. sophoroides			3		6	1	24			34
Cyclocheilichthys apogon			4		6	4	29		2	45

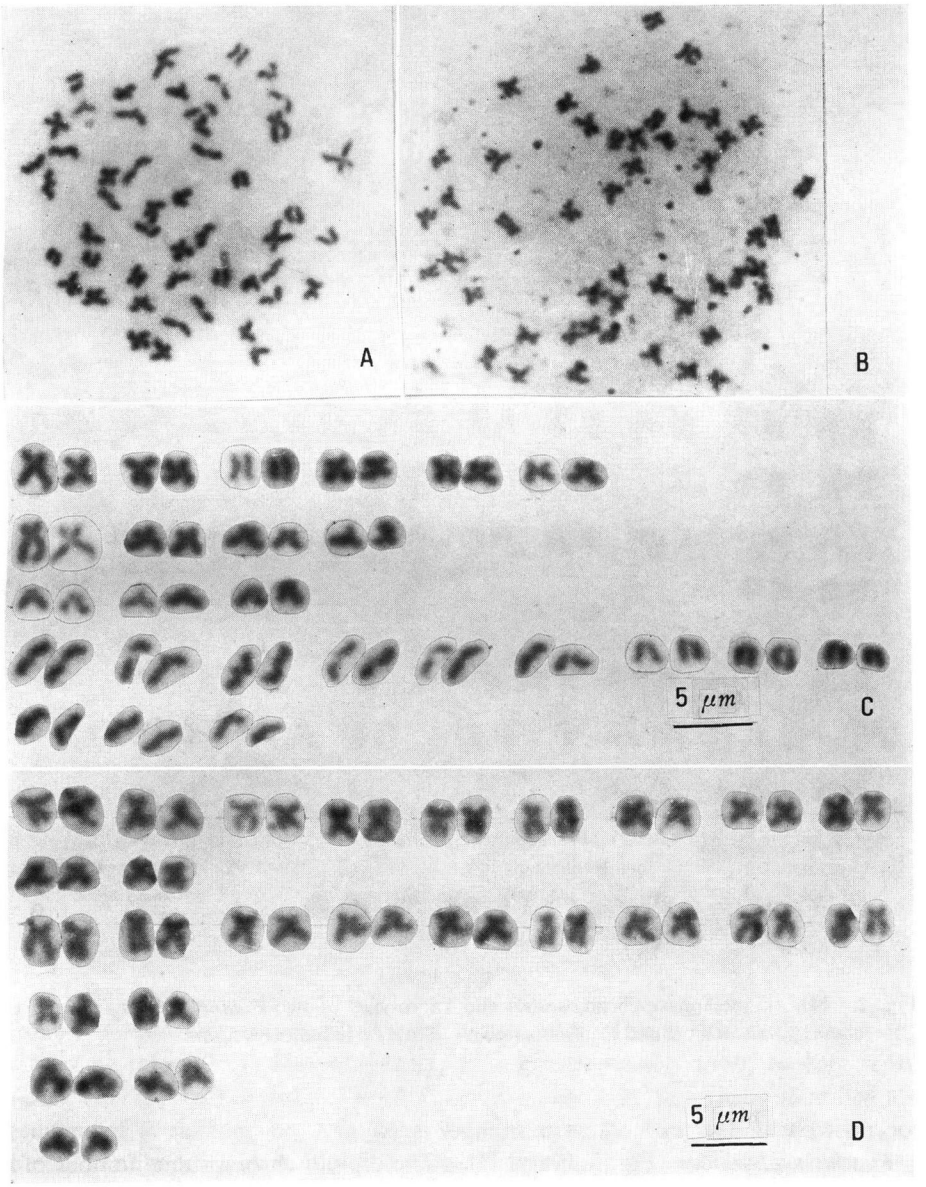

Fig. 1. Mitotic metaphase chromosomes and karyotypes of two *Puntius* species. A and C, *Puntius daruphani*; B and D, *Puntius stoliczkanus*. Each scale indicates 5 μm.

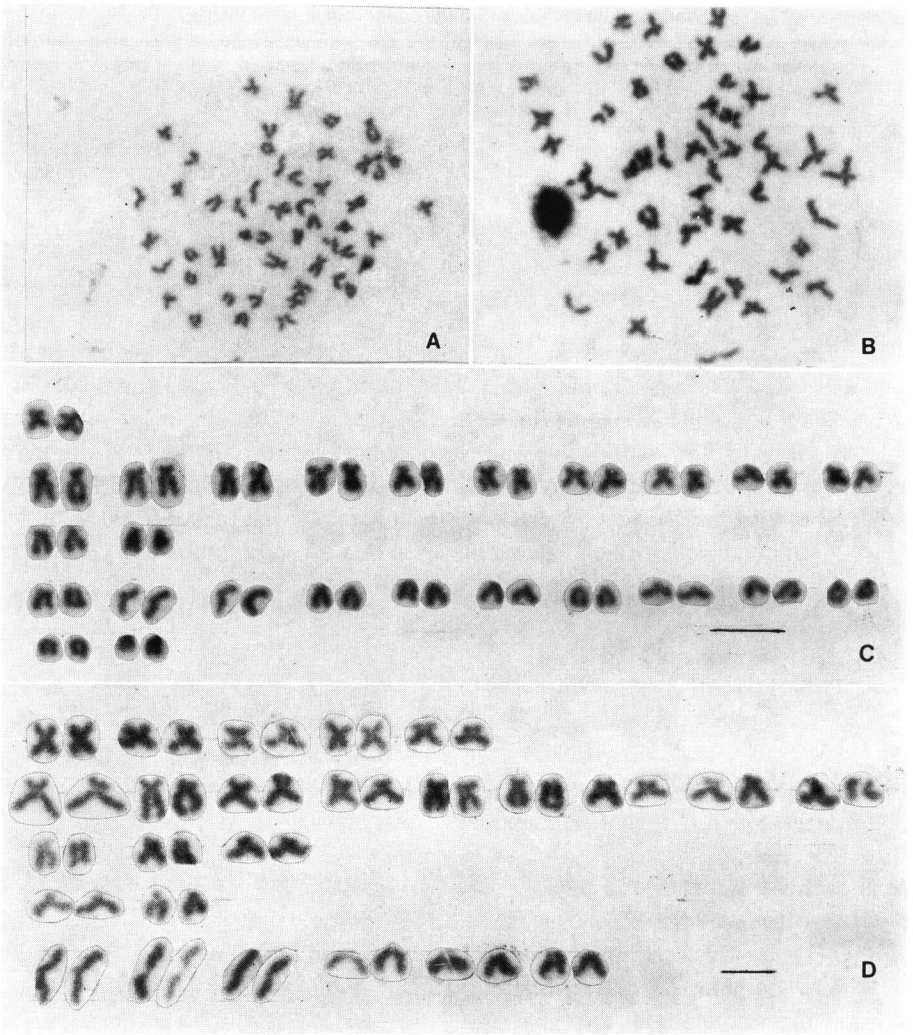

Fig. 2. Mitotic metaphase chromosomes and karyotypes of two *Puntius* species. A and C, *Puntius gonionotus*; B and D, *Puntius altus*. Each scale indicates 5 μm.

acrocentric chromosomes. The arm number is 84.

Puntius sophoroides (Fig. 3, B and D). The diploid chromosome number of this species is 50. The karyotype comprises 2 metacentric, 2 submetacentric, and 46 acrocentric chromosomes. The arm number is 54.

Cyclocheilichthys apogon (Fig. 3, A and C). The diploid chromosome number is 50 (Table 2). The karyotype comprises 12 metacentric, 8 submetacentric, 6 subtelocentric, and 24 acrocentric chromosomes. The arm number is 70.

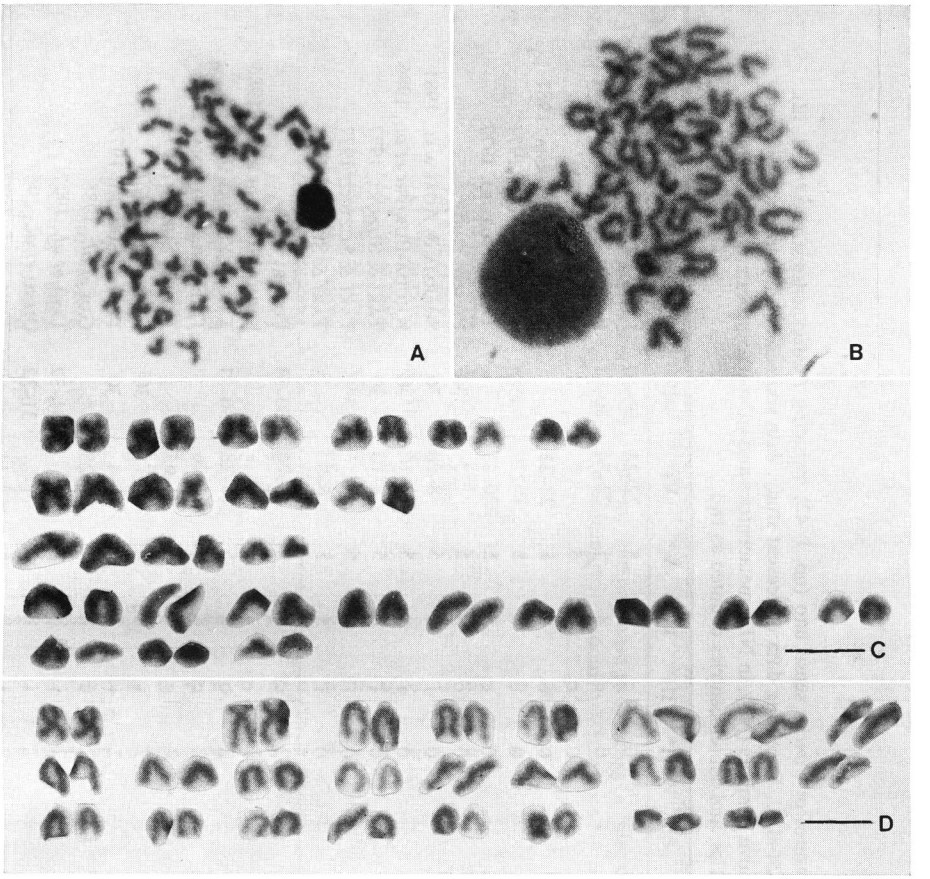

Fig. 3. Mitotic metaphase chromosomes and karyotypes of a *Puntius* species and a *Cyclocheilichthys* species. A and C, *Cyclocheilichthys apogon*; B and D, *Puntius sophoroides*. Each scale indicates 5 μm.

Discussion

Table 3 shows karyotypes of *Puntius* sensu lato. From this table following points may be inferred: 1) Karyologically, *P. stigma* is similar to *P. sophore* sensu RISHI *et al.* (1977) (no. 3 in Table 3) and *P. chrysopterus*. 2) As regards identification of specimens of *P. sophore*, no. 3 in Table 3 may not be the same species with no. 14 in Table 3. 3) The karyotype of no. 14 is similar to that of *P. sophoroides* or *P. chola*. 4) *P. sophoroides*, *P. chola*, and *P. tetrarupagus* are similar to each other. 5) SMITH (1945) synonymized *P. javanicus* with *P. gonionotus*, and karyologically *P. javanicus* is also very similar to *P. gonionotus*. 6) *Puntius* sensu stricto comprises two types of karyotypes. In one type, 2n is 48 and NF_2 is 52 to 54 (no. 1–4), while in the other type 2n is 50 and NF_2 is more than 82 (no. 5–12). This fact suggests that karyological

Table 3. Karyotypes and morphological features of *Puntius* sensu lato (no. 1–42) including *Puntius* sensu stricto (no. 1–12), *Capoeta* (no. 13–23), and *Barbodes* (no. 24–42). Excepting data of present study, data sources of karyology are different from those of morphology. As regards arm numbers (NF), in NF_1, metacentrics and submetacentrics are counted as two, while in NF_2, metacentrics, submetacentrics, and subtelocentrics counted as two.

No.	Species	2n	NF_1	NF_2	I*	II*	III*	IV*	V*	VI*	VII*	Literature on karyotypes
1	*Puntius stigma*	48	54	54	–	0	c	8	5	22–24	30–31	KHUDA-BUKHSH & BARAT, 1987
2	*P. stigma*	48		52	–	0	c	8	5	22–24	30–31	RISHI, 1973
3	*P. sophore*	48	54	54	–	0	c	8	5			RISHI *et al.*, 1977
4	*P. chrysopterus*	48	52	54	–	0	c	8	5	23–25		TRIPATHI & SHARMA, 1987
5	*P. filamentosus*	50		84	–	0	c	8	5	21	31	TAKI & SUZUKI, 1977
6	*P. nigrofasciatus*	50		100	+	0	c	8	5	20–21		TAKI & SUZUKI, 1977
7	*P. stoliczkanus*	50	94	98	+	0	c	8	5	22	30	Present study
8	*P. conchonius*	48	78	88	+	0	i	8	5	8–10	30	SHARMA & AGARWAL, 1981
9	*P. conchonius*	50	90	92	+	0	i	8	5	8–10	30	KHUDA-BUKHSH *et al.*, 1986
10	*P. conchonius*	50		94	+	0	i	8	5	8–10	30	TAKI & SUZUKI, 1977
11	*P. cumingi*	50		94	+	0	i	8	5	4		TAKI & SUZUKI, 1977
12	*P. ticto*	50		100	+	0	i	8	5	6–8		TAKI & SUZUKI, 1977
13	*P. sophoroides*	50	54	54	–	2	c	8	5	24–26	30–31	Present study
14	*P. sophore*	50	56	56		?						KHUDA-BUKHSH *et al.*, 1986
15	*P. chola*	50		56	–	2	c	8	5	26–28	30–31	TAKI & SUZUKI, 1977
16	*P. tetrarupagus*	50	52	58	–	2	c	8	5	24–26		TRIPATHI & SHARMA, 1987
17	*P. arulius*	50		82	–	2	c	8	5	21–23		TAKI & SUZUKI, 1977
18	*P. oligolepis*	50		88	–	2	i	8	5	6–7	30	TAKI *et al.*, 1977
19	*P. titteya*	50		98	–	2	i	7	5	3–4	30	TAKI & SUZUKI, 1977
20	*P. semifasciolata*	50	76	90	+	2	c	8	5	23–25		GUI *et al.*, 1986
21	*P. tetrazona*	50		84	+	2	i	8	5	7	31–32	TAKI *et al.*, 1977
22	*P. tetrazona*	50	84	90	+	2	i	8	5	7	31–32	OHNO *et al.*, 1967
23	*P. partipentazona*	50		90	+	2	i	8	5	10–11		TAKI *et al.*, 1977

24	*P. gonionotus*	50	72	76	+	4	c	8	6	29–31	34	Present study
25	*P. javanicus***	50	70	78	+	4	c	8	6			KHUDA-BUKHSH, 1975
26	*P. daruphani*	50	70	76	+	4	c	8	5	28	35	Present study
27	*P. lacustris*	50	80		+	4	c	8	5	45–49		ZAN *et al.*, 1984
28	*P. daliensis*	50	82		+	4	c	7–8	5	34–39		ZAN *et al.*, 1984
29	*P. fasciatus*	50		82	+	4	c	8	5	27–30		TAKI *et al.*, 1977
30	*P. fasciatus*	50	82	86	+	4	c	8	5	27–30		OHNO *et al.*, 1967
31	*P. melanampyx*	50	74	88	—	4	c	8	5	20		KHUDA-BUKHSH *et al.*, 1986
32	*P. altus*	50	84	88	+	4	c	8	5	31–32	31–32	Present study
33	*P. everetti*	50		86	+	4	c	8	5	22–25		TAKI *et al.*, 1977
34	*P. lateristriga*	50		88	+	4	c	8	5	23		TAKI *et al.*, 1977
35	*P. sarana*	50	76	88	+	4	c	8	5	31–34		RISHI, 1981
36	*P. schwanenfeldii*	50		84	+	4	c	8	5	35–36	32	TAKI *et al.*, 1977
37	*P. binotatus*	50		92	+	4	c	8	5	24		TAKI *et al.*, 1977
38	*P. orphoides*	50		92	+	4	c	8	5	31–34	35–36	TAKI *et al.*, 1977
39	*P. pentazona*	50		98	+	4	c	8	5	22–25		TAKI *et al.*, 1977
40	*P. caldwelli*	100	150	176	—	4	c	9	5	21–25	36–38	GUI *et al.*, 1985
41	*P. denticulatus denticulatus*	100	150	176	+	4	c	9	5	26–30	37–38	GUI *et al.*, 1985
42	*P. sinensis*	100	150	176	+	4	c	9	5	28–32		GUI *et al.*, 1985

* I. Serration of the longest simple dorsal ray: +, serrated; —, smooth. II. Number of barbels. III. Lateral line: c, complete; i, incomplete. IV. Number of branched dorsal fin rays. V. Number of branched anal fin rays. VI. Number of pored scales. VII. Number of vertebrae.

** *Puntius javanicus* was erroneously reported as *P. japonicus* (pers. comm. by KHUDA-BUKHSH, 1989).

difference between two types is so large that *Puntius* sensu stricto are not a monophyletic group. 7) *Capoeta* also comprises two types of karyotypes, i.e., a group with NF_2=54 to 58 (no. 13–16) and the other type with NF_2=82 to 98 (no. 17–23). Karyological difference between two types in *Capoeta* is so large that *Capoeta* are not a monophyletic group. 8) On the basis of three features such as karyotypes, the body color (e.g., a black spot near the caudal fin base), and non-serration of the longest simple dorsal fin ray, species of no. 1 to 4 are most similar to species of no. 13 to 16 in species listed in Table 3. Therefore, these species (no. 1–4 and no. 13–16) may form a monophyletic group. 9) No. 40 to 42 in Table 3 are characteristic in having 2n=100 and 9 branched dorsal fin rays. These three species belong to the subgenus *Spinibarbus* in the genus *Barbodes* (WU *et al.*, 1977), but recently *Spinibarbus* was ranked up from subgenus into genus (ZHENG *et al.*, 1989).

As classification of *Puntius* is very difficult, identification of *Puntius* species should be more careful, and materials used for experiments should be deposited for checking of their scientific names and the study on interrelationships between karyotypes and morphology. In conclusion, classification of *Puntius* must be revised.

As regards chromosomes of *Cyclocheilichthys*, the karyotype of *Cyclocheilichthys apogon* is the first report in *Cyclocheilichthys* species.

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