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Karyotypes in Four Species of the Family Cottidae

Hitoshi Ida, Hiroaki Terashima
and Tatsuo Fujimi

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The fishes of the order Scorpaeniformes exhibit a large morphological variation, representing an interesting material for karyology in relation to morphology. Nearly fifty species of the order Scorpaeniformes have been studied on their karyology, out of which thirteen species belong to the family Cottidae. Most of these species were collected from western Pacific waters.

The present study aims to analyze the karyotypes of four species in the family Cottidae, out of which three species were from the coast of Washington State, eastern Pacific, and details of their karyological features are described below.

Materials and methods

The fish specimens used for this study were collected from the Yoshihama river, Sanriku-cho, Iwate Prefecture, and the waters around San Juan Island, Washington State, northern Pacific coast of U.S.A. The specimens from Sanriku-cho were treated at the School of Fisheires Sciences, Kitasato University. The rest of the specimens were treated at the Friday Harbor Laboratories, University of Washington. Data of the materials are shown in Table 1.

For chromosomal studies, the in vitro method (Ida et al., 1978) was applied. The gill tissues were dissected from the right side of live fish and were incubated in minium essential medium (MEM) solution with 0.1 μg/ml colchicine for 9 hours. They were then treated in a hypotonic solution for 1 hour and fixed in Carnoy's solution for at least 2 hours. The slide glasses that were used for chromosome spreading had been kept in 50% methanol at 5°C. Giemsa solution was employed for staining of the preparations. The classification of the chromosomes followed that of Levan et al. (1964). Metacentrics and submetacentrics were treated as two-arm chromosomes, and subtelocentrics and acrocentrics as one-arm chromosomes. The new arm number (NAN) used here is that of Arai and Nagaiwa (1976).

Counts for vertebrae were made on X-ray photographs and the observation of scales and teeth as based on the specimens stained with alcian blue and alizaline red S. The identification of species followed Hart (1973) and Matsubara (1955).

Results

The distributions of the chromosome counts obtained in the four species are given in Table 2. There was no difference in the karyotypes of the male and female of *Cottus pollux*. Chromosome spreads were available only for either the male or female in the other three species. The modal chromosome number is 48 in all the four species. The photographs of the mitotic metaphase chromosomes and their karyotypes are shown in the following two figures (Figs. 1, 2). The karyotypes with fundamental numbers (FN) and new arm numbers (NAN) are listed in Table 2.

Details of the karyotype of each species are as

Table 1. List of materials used for chromosome experiments and morphological observation.

Species (English name, Japanese name)	Date	Locality	Method	Sex	No. of specimens	SL (mm)
Cottus pollux	1986-9-27	Sanriku-cho	hand-net	male	7	83.1–115.3
(Kajika)	1986-9-27	Sanriku-cho	hand-net	female	13	77.2–108.6
Oligocottus maculosus	1987-5-21	San Juan Is.	SCUBA diving	female	1	44.7
(Tidepool sculpin, Buchikajika)	1987-5-25	San Juan Is.	SCUBA diving	?	1	38.4
Artedius fenestralis	1987-5-22	San Juan Is.	SCUBA diving	male	2	55.6–61.7
(Padded sculpin)	1987-5-22	San Juan Is.	SCUBA diving	male	1	61.7
Artedius lateralis	1987-5-22	San Juan Is.	SCUBA diving	male	1	57.2
(Smoothhead sculpin)	1987-5-24	San Juan Is.	SCUBA diving	?	1	35.8

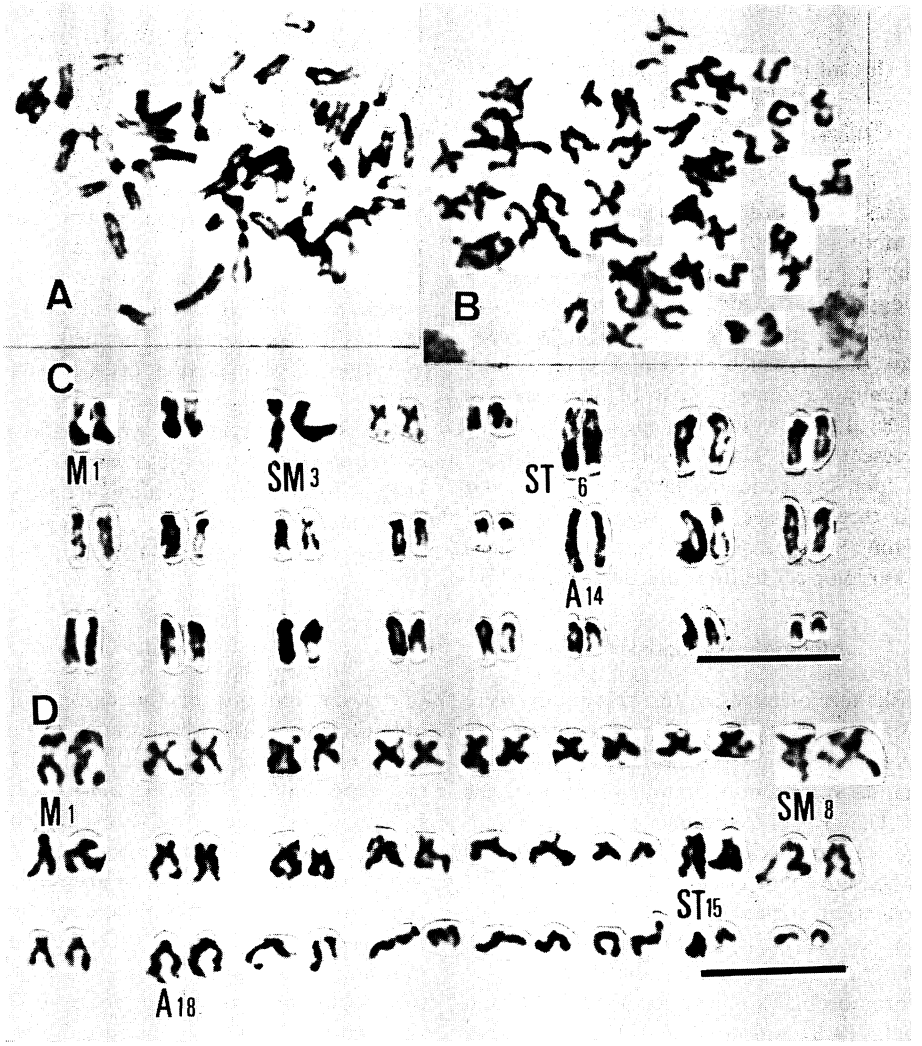

Fig. 1. Photographs of miotic metaphase chromosomes and their karoytypes. A and C, *Cottus pollux*; B and D, *Oligocottus maculosus*. Each scale indicates 10 μm.

Table 2. Frequency distributions of diploid chromosome numbers and karyotypes of four species

Species	No. of specimens	2n								Total
		43	44	45	46	47	48	49	50	
Cottus pollux	5				2	1	7	1		11
Oligocottus maculosus	2				1		8			9
Artedius fenestralis	3						4			4
Artedius lateralis	2						5			5

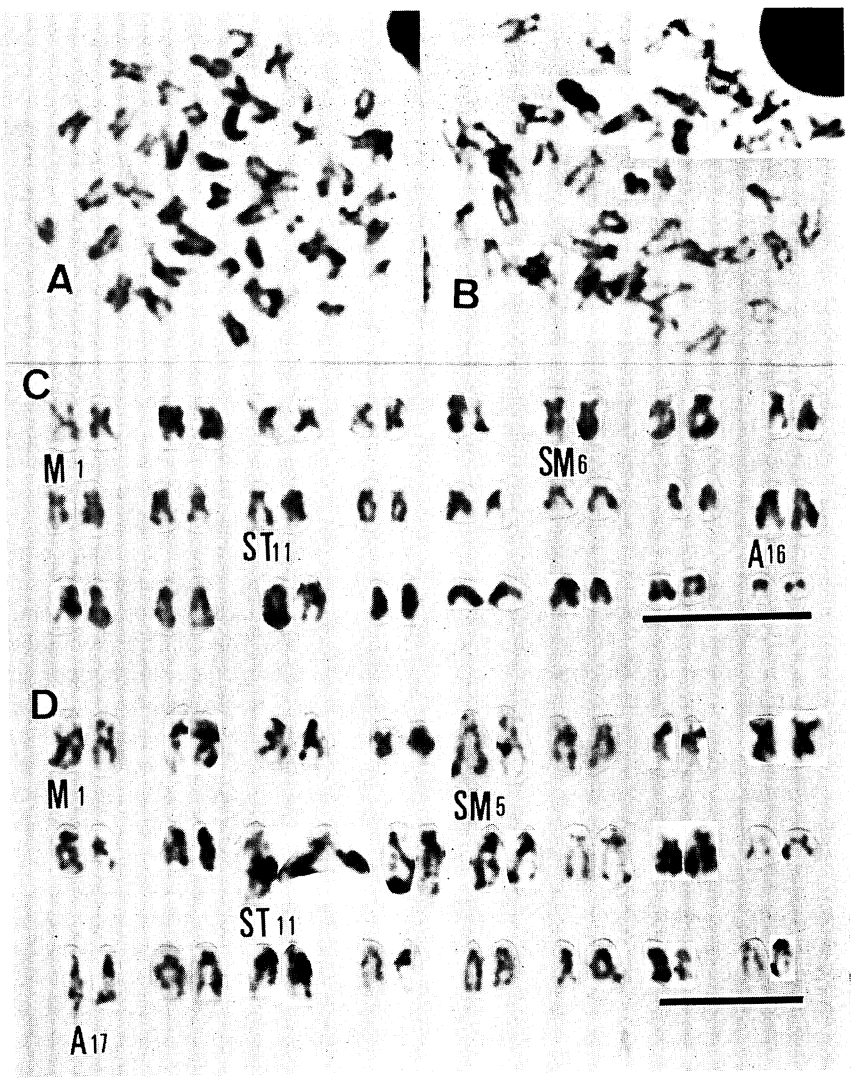

Fig. 2. Photographs of miotic metaphase chromosomes and their karyotypes. A and C, *Artedius fenestralis*. B and D, *A. lateralis*. Each scale indicates 10 μm.

of the family Cottidae.

2n	Meta-centric	Submeta-centric	Subtelo-centric	Acro-centric	FN	NAN
48	4	6	16	22	58	48
48	14	14	6	14	76	48
48	10	10	10	18	68	48
48	8	12	12	16	68	48

follows.

Cottus pollux Günther (Fig. 1A, C): The karyotype of *Cottus pollux* consists of 2 pairs of metacentric, 3 pairs of submetacentric, 8 pairs of subtelocentric, and 11 pairs of acrocentric chromosomes. The sizes of the 3rd submetacentric and the 6th subtelocentric chromosomes are larger than the rest. The sizes of the rest decrease gradually.

Oligocottus maculosus Girard (Fig. 1B, D): The karyotype of *Oligocottus maculosus* consists of 7 pairs of metacentric, 7 pairs of submetacentric, 3 pairs of subtelocentric and 7 pairs of acrocentric chromosomes. The sizes of the 1st metacentric and 8th submetacentric chromosomes are larger than the rest. The 14th submetacentric and 24th acrocentric chromosomes are apparently smaller than the rest.

Artedius fenestralis Jordan et Gilbert (Fig. 2A, C): The karyotype of *Artedius fenestralis* consists of 5 pairs of metacentric, 5 pairs of submetacentric, 5 pairs of subtelocentric and 9 pairs of acrocentric chromosomes. The 15th subtelocentric and 24th acrocentric chromosomes are smaller than the rest. The sizes of the rest decrease gradually.

Artedius lateralis (Girard) (Fig. 2B, D): The karyotype of *Artedius lateralis* consists of 4 pairs of metacentric, 6 pairs of submetacentric, 6 pairs of subtelocentric, and 8 pairs of acrocentric chromosomes. The sizes of the 5th submetacentric and two pairs of subtelocentric (11th and 12th) chromosomes are larger than the rest. The sizes of the rest decrease gradually.

Morphological characters, such as fin ray counts and vertebral composition, are shown in Table 3. Scales are absent in *C. pollux* and *O. maculosus*. The scales of *A. fenestralis* and *A. lateralis* are arranged in many rows on most part of the trunk. The palatine teeth are absent in *C. pollux*, but present in the other three species. The prevomerine teeth are present in all of the four species. The gill membranes of *O. maculosus*, *A. fenestralis* and *A. lateralis* are not united with the isthmus but are united with it in *C. pollux*. The vertebrae are composed of 11 abdominal and 24 caudal vertebrae in *C. pollux*, 12 and 22 in *O. maculosus*, and 10 and 23 in *A. fenestralis* and *A. lateralis*.

Discussion

The karyotypes of the four species of the family Cottidae show a rather wide variation. Both the chromosome number and new arm number are 48 in all the four species. Robertsonian translocation of chromosomes was not found among these species. As can be seen from Table 2, the smallest fundamental number is 58 found in *Cottus pollux*. Abe (1976) reported the karyotype of *Cottus pollux* as having 2 metacentric, 8 submetacentric and 38 subtelocentric or acrocentric chromosomes. There is no difference in the fundamental number of *C. pollux* between the present result and his report. Arai and Fujiki (1978) briefly reviewed the karyotypes of eight species of the genus *Cottus*, viz. *C. pollux*, *C. hangiongensis*, *C. nozawae*, *C. reini*, *C. kazika*, *C. gobio* and *C. poecilopus*. Further, Ojima (1983) reviewed the karyotypes of five species of the genus *Cottus*, viz. *C. gobio*, *C. kajika*, *C. poecilopus*, *C. pollux*, and *C. reini*. Among these species, two different chromosome numbers are reported in *C. gobio*. Starmach (1967) reported that its diploid chromosome number was 52 while Post (1965) reported that it was 48. It is not clear whether the difference found between these two reports reflects the variation within a species or the difference of the species of their materials. The chromosome numbers of the genus *Cottus* range from

Table 3. Some selected morphological characters of the material fish. VN, vertebral number (abdominal+caudal).

Species	n	Dorsal fin	Anal fin	VN	SL/BD	SL/HL	Scale	Teeth on palatine	Gill membranes and isthmus
Cottus pollux	20	VIII, 18–19	13–15	11+24	4.4	3.9	−	−	united
Oligocottus maculosus	2	VIII, 17	12–14	12+22	4.2	4.2	−	+	not united
Artedius fenestralis	3	VIII or IX, 16	12–14	10+23	4.3	2.8	+	+	not united
Artedius lateralis	2	VIII or IX, 17	12–14	10+23	5.0	2.7	+	+	not united

40 to 52. In these species, the smallest chromosome number is 40 as represented by *Cottus kazika* which is restricted to western Japan. Another extreme is represented by *C. gobio* which is restricted to the rivers around the Baltic sea. Thus fishes of the genus *Cottus* show rather wide variations in karyological characters. This fact seems to be related to their restricted distribution.

From the osteological viewpoint, Yabe (1985) studied the phylogeny of the superfamily Cottoidea, and placed the genus *Cottus* as the most generalized one among the three genera, viz. *Cottus*, *Artedius* and *Oligocottus*, by the existence of the palatocranial articulation. Further, he placed the genus *Oligocottus* as the most specialized one among the three genera mentioned above by the absence of scales. On the other hand, according to Yabe (1985), the condition of the gill membranes and the denticulate palatine are regarded as important characters for the phylogenetic classification. But as can be seen in Table 3, *Cottus pollux* shows a more specialized (the fusion of gill membranes with the isthmus) or degenerated (absence of palatine teeth) state than the other three species, viz. *A. lateralis*, *A. fenestralis* and *O. maculosus*. From the karyological viewpoint, there is a slight difference observed in fundamental numbers among the four species. And the number of metacentric chromosomes is in the order of *C. pollux*, *A. lateralis*, *A. fenestralis*, and *O. maculosus*. Generally, the increase of bi-armed chromosomes, especially by the pericentric inversion, is regarded as an advanced or specialized state (Arai and Nagaiwa, 1976). Thus, in these species, the karyological specialization seems to be in accordance with the phylogeny suggested by Yabe (1985). But *C. pollux* shows more specialized morphological characters than the other species as mentioned above. And most chromosomes have a short arm in these species, suggesting the occurrence of a frequent pericentric inversion among the group. The differences in the relative length of chromosomal arms in subtelocentric and submetacentric chromosomes are very gradual in these species and the observed differences in fundamental number among these four species do not seem to be so critical. Thus, a more detailed comparison of the chromosome structure seems to be needed for the analysis of the systematic relation in the family Cottidae.

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(School of Fishery Sciences, Kitasato University, Sanriku-cho, Kesen-gun, Iwate Pref. 022–01, Japan)

カジカ科4種の核型

井田　齊・寺島裕晃・藤見辰男

本邦産カジカ科魚類1種及び合衆国産カジカ科魚類3種の核型を air-drying 法により分析した．染色体数，new arm number はいずれも 48 であった．核型はカ

ジカ *Cottus pollux* で中部着糸型 (M)=4, 次中部着糸型 (SM)=6, 次端部着糸型 (ST)=16, 端部着糸型 (A)=22; *Oligocottus maculosus* で, M=14, SM=14, ST=6, A=14; *Artedius fenestralis* で, M=10, SM=10, ST=10, A=18; *A. lateralis* で, M=8, SM=12, ST=12, *A*=16 であった. 4 種の FN 値 (Fundamental number) はカジカ 58, *Artedius* 属の 2 種 68, *Oligocottus maculosus* 76, の順に増加していた. 上記 4 種には, 2 腕性染色体が多く, 挟動原体逆位が多く生じていると考えられる.

(022-01 岩手県気仙郡三陸町 北里大学水産学部)