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# Karyotypes of Three Species of the Family Cottidae (Scorpaeniformes)

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**Abstract** Karyotypes and cellular DNA contents of three species of the family Cottidae viz. *Icelus cataphractus*, *Gymnocanthus intermedius* and *Alcichthys alcicornis* were analyzed. Structural modifications within the family were supposedly by Robertsonian translocations. The diploid chromosome numbers were determined to be 48 in *Alcichthys alcicornis*, 44 in *Gymnocanthus intermedius* and 40 in *Icelus cataphractus*. The DNA contents ranged from 1.46 to 1.50 pg/cell in the three species. The karyotype of *Icelus cataphractus* is unique in having the smallest chromosome number (2n=40) and 14 large-sized chromosomes. From the chromosome number and the existence of some large chromosome pairs, Robertsonian translocations seem to have occurred frequently in *Icelus cataphractus* and *Gymnocanthus intermedius*.

The family Cottidae with over 300 species is a large family within Scorpaeniformes, being very common in subboreal and temperate waters. The fishes of this group exhibit complex relationships because of the high interspecific variability in morphological characters (Taranetz, 1941, Yabe, 1985). Karyological analyses seem to be useful in clarifying systematic relationships. However, karyological studies have been attempted on no more than 20 species. The present study aims to clarify karyological aspects of three cottids, viz. *Icelus cataphractus* (Pavlenko), *Gymnocanthus intermedius* (Temminck et Schlegel) and *Alcichthys alcicornis* (Herzenstein). Details of their karyological features are described below.

## Materials and methods

The specimens used for this study were collected from Okirai Bay, Sanriku-cho, Iwate Prefecture. Data of materials are shown in Table 1.

For chromosomal studies, an in-vitro method (Ida et al., 1978) was applied. The gill tissues were removed from live fish and incubated in minimum essential medium (MEM) solution with 0.1 $\mu$g/m$l$ colchicine for 2 hours at room temperature. Gill tissues were then treated in a hypotonic solution for 90 minutes and fixed in Carnoy's solution for 1 hour. Classification of chromosomes followed that of Levan et al. (1964). Subtelocentrics and acrocentrics were treated as one-arm chromosomes. New arm number (NAN) terminology follows Arai and Nagaiwa (1976). The DNA value was determined by measuring erythrocyte nuclei stained with Feulgen reaction using an integrating microdensitometer, Nikon Vickers M-85A. Erythrocytes of the common carp, *Cyprinus carpio* Linnaeus, were used as the control.

## Results

The frequency distribution of chromosome numbers for each species is shown in Table 2. Photographs of mitotic metaphase chromosomes and their karyograms are shown in Figs. 1 and 2. Karyotypes with fundamental numbers (FN) and new arm numbers (NAN) are listed in Table 3. In *Icelus cataphractus*, chromosome spreads were available only from male material. No difference was found in male and female karyotypes of the other two species. Modal diploid chromosome numbers (2n) are 48 in *Alcichthys alcicornis*, 40 in *Icelus cataphractus* and 44 in *Gymnocanthus intermedius*. Details of the karyotype for each species are as follows.

*Icelus cataphractus* (Fig. 1A, C). The diploid chromosome set shows 12 metacentrics, 12 submetacentrics and 16 subtelocentrics or acrocentrics. 5 pairs of metacentric and 2 pairs of submetacentric chromosomes are more than twice the size of the rest.

*Gymnocanthus intermedius* (Fig. 1B, D). The diploid chromosome set shows 2 metacentrics, 4 submetacentrics and 38 subtelocentrics or acrocentrics. 1 pair of metacentric and 1 pair in submetacentric chromosomes are more than twice the size of the rest.

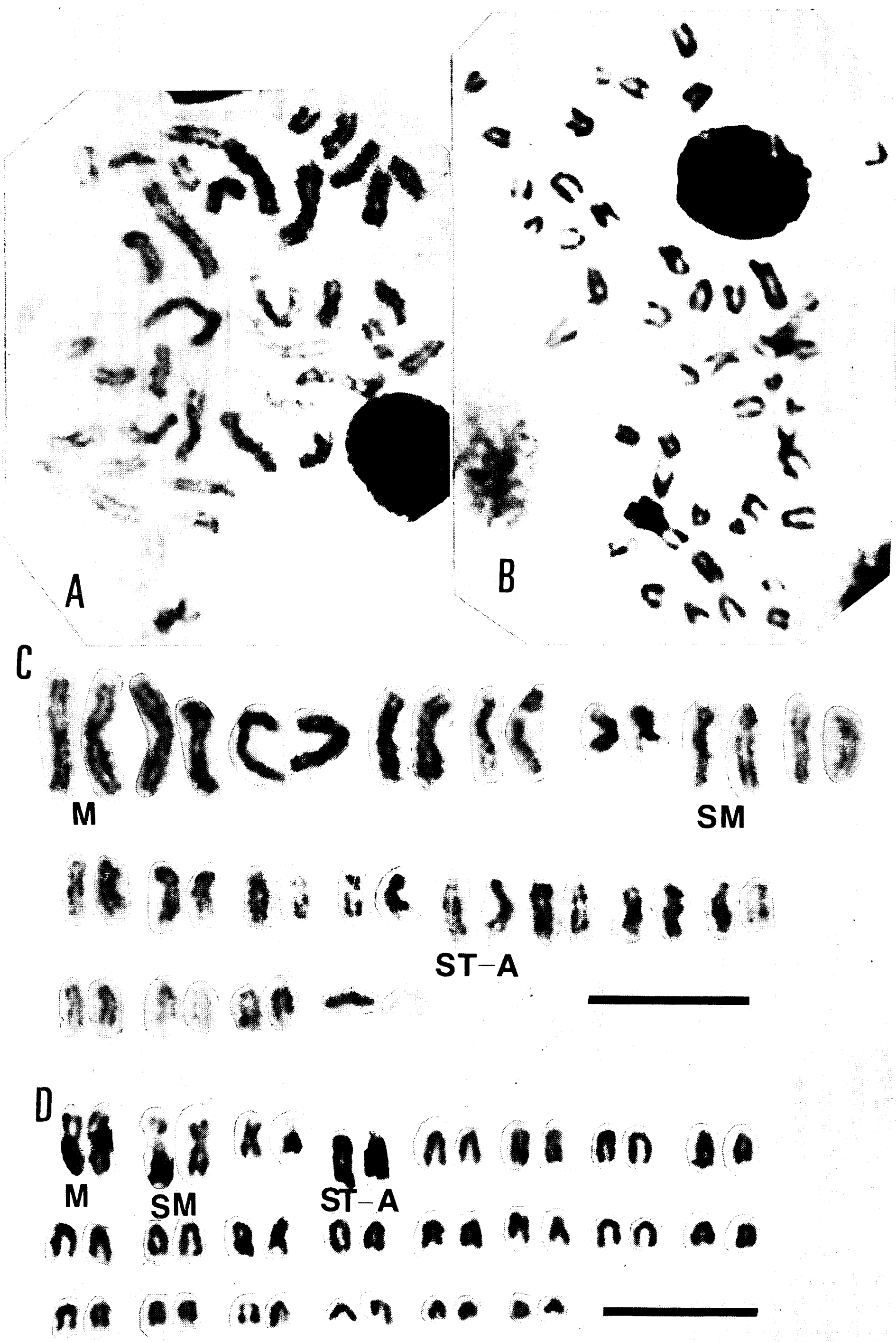


Fig. 1. Chromosome spreads and karyograms of two species of the family Cottidae. A and C, *Icelus cataphractus*; B and D, *Gymnocanthus intermedius*. Each scale indicates 10 $\mu$m.

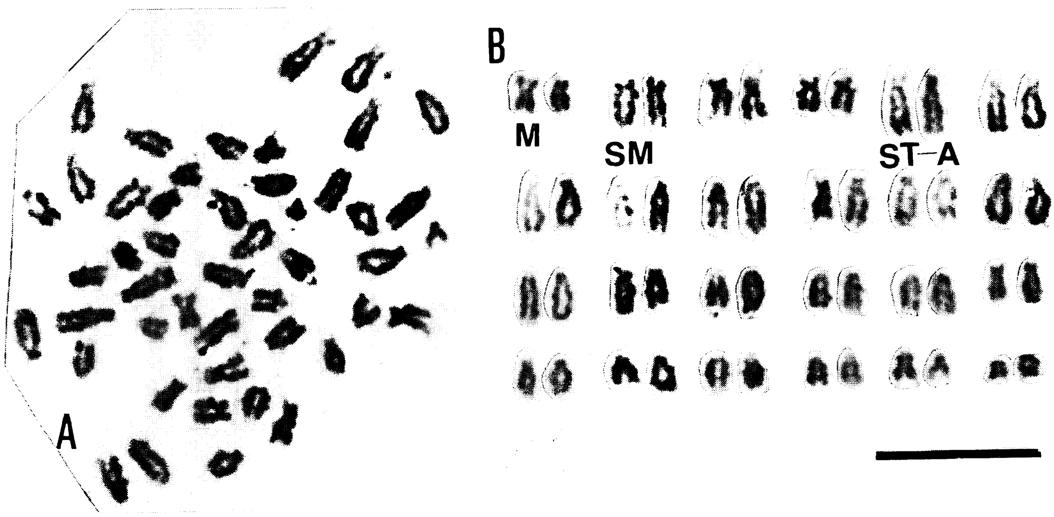


Fig. 2. Chromosome spread and karyogram of *Alcichthys alcicornis*. The scale indicates 10 μm.

*Alcichthys alcicornis* (Fig. 2A, B). The diploid chromosome set shows 2 metacentrics, 6 submetacentrics and 40 subtelocentrics or acrocentrics. The size of the chromosomes decreases gradually. No marker chromosome of conspicuous size was observed.

The DNA contents of these three species are very similar, ranging from 1.46 to 1.50, as shown in Table 4.

## Discussion

The karyotypes of the three species studied have show rather wide variation. As can be seen from Table 3, their fundamental number ranges from 50 to 64. Judging from both the chromosome number and the presence of some large chromosomes, Robertsonian translocations seem to have frequently occurred in *Icelus cataphractus* and *Gymnocanthus intermedius*. The karyotype of *Icelus cataphractus* was the first analyzed for the genus *Icelus*. Arai and Fujiki (1978) briefly reviewed karyotypes of fishes in the order Scorpaeniformes. On the basis of 18 species, they considered the new arm number for the order to be 48. Ida and Yunokawa (1980) examined 5 species in the family Platycephalidae and also found the new arm number to be 48. Ida et al. (1982, 1989) later examined 8 species of *Sebastes* (Scorpaenidae) and 4 species in the family Cottidae, finding the new arm number to be 48 in all the species. Thus most species in the order Scorpaeniformes have a new arm number of 48. From a systematical consideration, it may be expected that *Icelus cataphractus* would be the same or similar new arm number to the above, but it can be estimated that the former has, in fact, a much higher new arm number (54?). It is apparent, therefore, that the new arm number of the family Cottidae needs further investigation. *Icelus cataphractus*, which is the deepest dwelling of the three species considered here, is characterized by the smallest chromosome number (2n=40) and the presence of 14 large chromosomes. This suggests that the karyotype is very specialized. The diploid and new arm numbers of *Gymnocanthus intermedius* are 44 and 48, respectively. Thus, the four, large, two-arm chromosomes in the karyotype of this species seem to have originated from centric fusion. The diploid and new arm numbers of *Alcichthys alcicornis* are both 48. Judging from the diploid chromosome number and their size, bi-arm chromosomes in this species seem to have originated by pericentric inversion.

The DNA contents in these three species ranged from 1.46 to 1.50 pg/cell and showed almost the same values. Thode et al. (1985) analyzed cytogenetic aspects of two species of scorpion fishes and mentioned that the most evolved karyotype of the two had probably been derived by numerous rearrangement involving great genome losses. They referred to unequal crossing-over and exchange, as illustrated by

Ohno (1974), as the mechanism for reduction of genome size. In the present case, on the other hand, DNA values were very similar whereas karyotypes showed remarkable variation among the three species. It may therefore be said that any structural modification in the karyotypes arose without definite change of DNA contents. Bickham and Baker (1979) proposed a canalization model of chromosomal evolution from a karyological study of turtles and bats. They grouped systematic divergences into three stages. Stage I was characterized by radical diversification at the familial level. Such diversification was accompanied by non-Robertsonian translocations such as inversions. At stages II and III, karyological stability within the family resulted from Robertsonian translocations. As an additional comment, if a pericentric inversion was accompanied by unequal crossing-over, it would be expected to result in sudden changes in both genome size and gene arrangement. On the other hand, Robertsonian translocation and pericentric inversion without the unequal crossing-over would not result in extreme changes in genome size. From osteological features, Berg (1940) proposed the family Icelidae, comprising three genera, *Icelus*, *Ereunias* and *Marukawichthys*. However, Yabe (1981) reassessed Berg's

Table 1. List of materials.

| Species | Data | Locality | Sampling method | No. of specimens | SL (mm) | Sex |
|---|---|---|---|---|---|---|
| *Icelus cataphractus* | 1987–2–21 | off Sanriku | trawl | 1 | 188 | male |
| *Gymnocanthus intermedius* | 1986–4–19 | Okirai Bay | set net | 1 | 182 | male |
| | 1988–5–21 | Okirai Bay | set net | 1 | 176 | female |
| *Alcichthys alcicornis* | 1986–4–19 | Okirai Bay | set net | 1 | 182 | male |
| | 1986–5–18 | Okirai Bay | set net | 1 | 184 | male |
| | 1986–6–28 | Okirai Bay | set net | 1 | 173 | male |
| | 1988–4– 9 | Okirai Bay | set net | 1 | 165 | female |

Table 2. Distribution of chromosome counts for three species of the family Cottidae.

| Species | Chromosome count | | | | | | | | | | | No. of cells observed |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | 39 | 40 | 41 | 42 | 43 | 44 | 45 | 46 | 47 | 48 | 49 | |
| *Icelus cataphractus* | 4 | 12 | 2 | | | | | | | | | 18 |
| *Gymnocanthus intermedius* | | | | | 2 | 10 | | | | | | 12 |
| *Alcichthys alcicornis* | | | | | | | | 4 | 2 | 26 | | 32 |

Table 3. Karyotypes of three species of the family Cottidae.

| Species | 2n | Metacentric | Submetacentric | Subtelo or acrocentric | FN | NAN |
|---|---|---|---|---|---|---|
| *Icelus cataphractus* | 40 | 12 | 12 | 16 | 64 | ? |
| *Gymnocanthus intermedius* | 44 | 2 | 4 | 38 | 50 | 48 |
| *Alcichthys alcicornis* | 48 | 2 | 6 | 40 | 56 | 48 |

Table 4. DNA content (pg/cell) of three species of the family Cottidae. *Control.

| Species | Cells observed | Arbitrary DNA unit | Standard error | Standard deviation | Absolute DNA pg/cell |
|---|---|---|---|---|---|
| *Icelus cataphractus* | 100 | 11.81 | 0.03 | 0.35 | 1.50 |
| *Cyprinus carpio** | 100 | 27.05 | 0.10 | 0.71 | 3.40 |
| *Gymnocanthus intermedius* | 100 | 11.30 | 0.07 | 0.66 | 1.47 |
| *Cyprinus carpio** | 100 | 26.17 | 0.09 | 0.95 | 3.40 |
| *Alcichthys alcicornis* | 100 | 13.36 | 0.06 | 0.34 | 1.46 |
| *Cyprinus carpio** | 100 | 30.47 | 0.02 | 0.83 | 3.40 |

Icelidae and concluded that *Icelus* was a member of the family Cottidae, later (Yabe, 1985) locating it centrally in his cladogram of 44 cottid genera. From karyological features, it is considered that the karyotype of *Icelus cataphractus* may have resulted from Robertsonian fusion. Accordingly, the uniqueness of the karyotype in this species would influence only to a small degree the genome size and gene arrangement. Thus the karyotype of *Icelus cataphractus* does not contradict the systematic allocation of *Icelus* given by Yabe (1981). Arai and Fujiki (1978) reported the karyotypes of three cottid species, viz. *Pseudoblennius cottoides* (Richardson), *P. marmoratus* (Döderlein) and *Ocynectes maschalis* Jordan et Starks. They noted that both chromosome number and new arm number was 46 in *Ocynectes maschalis*. From a consideration of the relatively constant cellular DNA value, large variation in chromosome size in each species, and relatively complicated shape of chromosomes in each species, it may be concluded that simple inversions without unequal crossing-over and centric fusions are very common among fishes of the family Cottidae.

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## 本邦産カジカ科3種の核型

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本邦産カジカ科3種（コオリカジカ *Icelus cataphractus*, アイカジカ *Gymnocanthus intermedius*, ニジカジカ *Alcichthys alcicornis*）の核型を air-drying 法により分析した．3種の染色体数・FN 値は，*I. cataphractus* で 2n=40 FN=64, *G. intermedius* で 2n=44 FN=50, *A. alcicornis* で 2n=48 FN=60 であった．*G. intermedius* と *A. alcicornis* の NAN 値は 48 であったが，*I. cataphractus* には 14 個の大型染色体が見られ，大型染色体×2+通常の染色体数は，カサゴ目魚類の基本的な染色体数と考えられる 48 を上回っていた．*I. cataphractus* と *G. intermedius* には，大型の2腕性染色体が複数個見られ，ロバートソン型の動原体融合が生じていると考えられる．

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