

Multiple Sex Chromosomes in the Redfin Velvetfish, *Hypodytes rubripinnis*

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Multiple sex chromosomes have been found in 11 out of approximately 1,600 fish species studied for their karyotypes. Ueno and Miller (1971) first discovered this type of sex chromosome system in a cyprinodontid fish. They applied the theory developed by Matthey (1965) who observed the specific karyotype in a rodent species. Since then, fish researchers have considered that sex is controlled by the $X_1 X_1 X_2 X_2-X_1 X_2 Y$ type of sex chromosome system, when they observed consistent presence of a large, unpaired chromosome in males instead of two small chromosomes as in females. Ichthyologists have also employed the supporting evidence that specific trivalent formation and successful segregation of the chromosome sets occur during male meiosis. In the course of chromosome studies in marine Teleostei, we discovered multiple sex chromosomes in the redfin velvetfish, *Hypodytes rubripinnis* (Temminck et Schlegel) (Scorpaeniformes, Congiopodidae).

Materials and methods

The locality and number (in parentheses) of specimens examined were: Amatsu-kominato, Chiba Prefecture (2 females and 2 males); Shimizu, Shizuoka Prefecture (1 female and 2 males); Wakayama Pre-

fecture (2 females and 3 males); Obama, Fukui Prefecture (2 females and 2 males); and Iwaya, Awajishima, Hyogo Prefecture (1 female and 2 males). Sex determination was based on differences in length of the second and third dorsal spines. The difference was apparent between sexes (those of the male were 1.5 times longer than those of the female) even in specimens lacking developed gonads.

Live fish received an intraperitoneal injection of approximately 0.5 ml of 100 μ g/ml colcemid solution per gram of body weight. After 2 $\frac{1}{2}$ -3 hours, kidney and testis tissues were removed. Chromosome preparations were made by the conventional air-drying method and stained with Giemsa. After microscopic observation, slides were C-banded by the BSG method (Sumner, 1972). Silver staining of NORs followed Howell and Black (1980). Chromosome terminology according to centromeric position followed the criteria established by Levan et al. (1964).

Results and discussion

Female karyotype. Chromosome counts on 105 well-spread metaphase plates obtained from 8 specimens showed a sharp mode of $2n=48$, indicating the normal diploid number for females (Fig. 1A). These chromosomes were classified into 2 metacentric and 46 acrocentric chromosomes (Fig. 2A). The two smallest acrocentric chromosomes often had satellites. The acrocentric chromosomes showed a gradual decrease in size. The size of the metacentric chromosomes was close to the medium-sized acrocentric chromosomes. C-bands appeared on the centromeric regions of about a half number of the chromosomes and on the satellites (Fig. 2B). Silver-

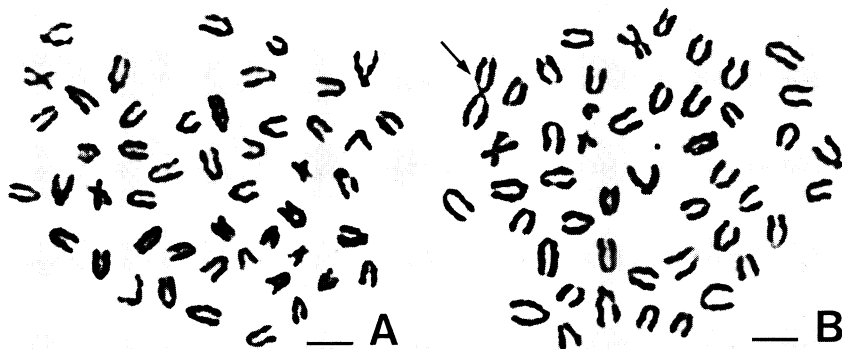


Fig. 1. Somatic chromosome complements of the redfin velvetfish (A: female $2n=48$, B: male $2n=47$). Arrow indicates the neo-Y chromosome. Each scale indicates 3 μ m.

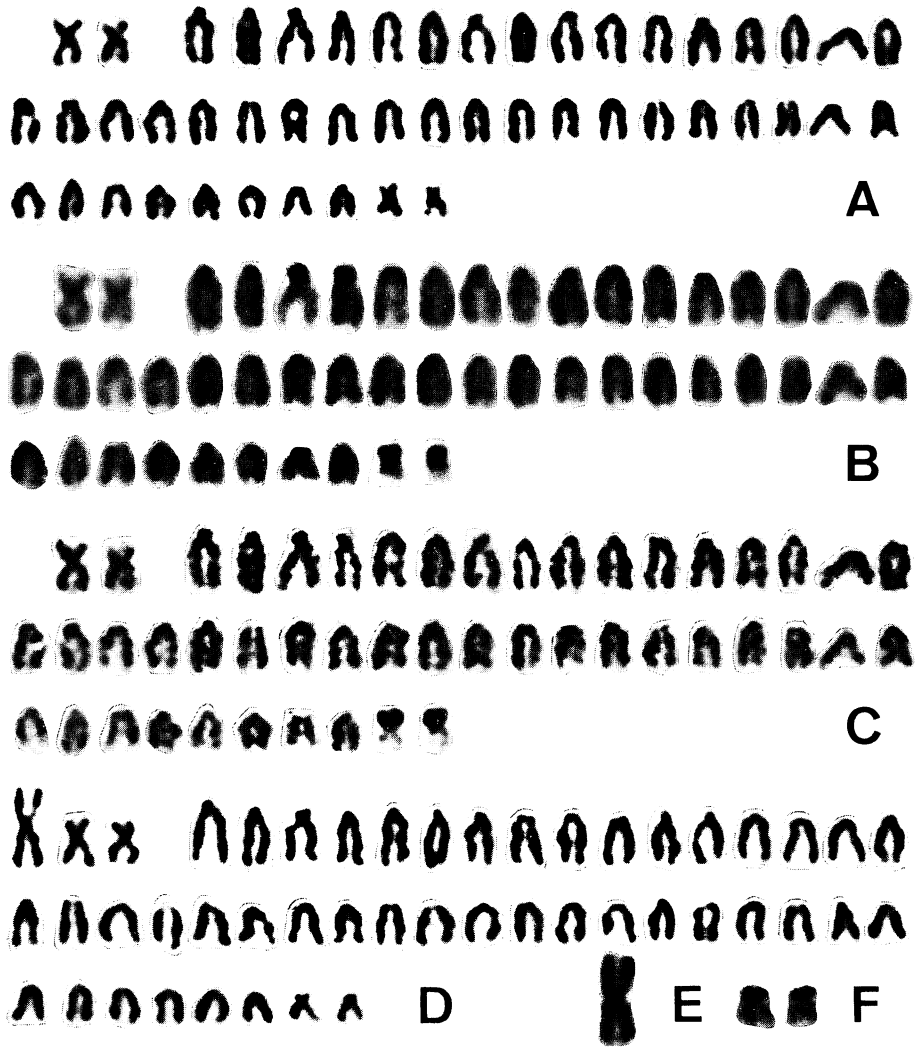


Fig. 2. Giemsa-stained karyotypes (A: female, D: male), C-banding patterns (B: female, E: male) and silver-stained NORs (C: female, F: male) in the redfin velvetfish.

stained NORs were found on the satellite regions (Fig. 2C). The arm number (NF) was 50.

Male karyotype. One hundred and twenty well-spread metaphase plates obtained from 11 specimens showed a sharp modal count of 47, indicating that the male diploid chromosome number is $2n = 47$ (Fig. 1B). The complement consisted of 3 metacentric and 44 acrocentric chromosomes, including 2 satellite-bearing chromosomes (Fig. 2D). The NF was 50 as in females. One of the metacentric chromosomes was unpaired and extremely large. It was approximately twice as long as the medium-sized

acrocentric chromosomes, with neither of its two arms C-band stained (Fig. 2E). This chromosome was considered to be neo-Y, which we assume to have arisen from the Robertsonian fusion of the primitive Y chromosome and one autosome. Banding patterns of the remaining chromosomes were the same as in the females (Fig. 2F).

Meiotic chromosomes. There were 22 autosome bivalents and one trivalent comprising sex chromosomes in all of the numerous nuclei at diakinesis and first meiotic metaphase in the males observed (Fig. 3). Chiasmata were present at approximately half of



Fig. 3. Meiotic chromosome complements from spermatocytes showing 22 bivalents and 1 trivalent. Arrows and arrowheads indicate the sex trivalents (A–C) and the chiasmatic associations (B, C), respectively. Each scale indicates 3 μ m.

the autosomal synapses, while a chiasma often appeared at one of the two associations of the trivalent (Figs. 3B, C). It is highly possible that the

sex chromosomes conjugate terminally between the primitive Y and X (X_1) while chiasmatically between the autosomal arm of the neo-Y and its corresponding element (X_2).

Arai and Katsuyama (1974) have already reported that the karyotype of this species is $2n=48$, consisting of 4 meta- and submetacentrics, and 44 subtelo- and acrocentrics (the former includes the satellited chromosomes). However, in the present study multiple sex chromosomes were observed in specimens obtained from the same locality. There would be no males in the material they used. All of the redfin velvetfish specimens, obtained from several localities in this study, had sex chromosomes of the $X_1 X_1 X_2 X_2-X_1 X_2 Y$ type. This indicates that the presence of sex chromosomes in this species is not limited to a local population.

Sex chromosomes of this type have been reported in various fish lineages: a cyprinodontid (Uyeno and Miller, 1971), a goodeid (Uyeno and Miller, 1972), an aluterid (Murofushi et al., 1980), an erithrinid (Bertollo et al., 1983), two callionymids (Murofushi et al., 1983), a gobiid (Pezold, 1984), an ophichthid (Murofushi and Yosida, 1984), a berycid (Ojima and Kikuno, 1986), a monodactylid (Suzuki et al., 1988) and a cobitid fish (Saitoh, 1989). The widely dispersed presence of multiple sex chromosomes in various fish groups means that neo-Y formation is a completely incidental event. During meiosis, a time lag of chromosome condensation and movement towards the poles often exists between autosomes and sex chromosomes (White, 1973). Thus the probability of translocation between a sex chromosome and an autosome is much lower than between autosomes. Further, the formation of neo-Y is possible only when there is no disorder in sex determination. The rare occurrence of multiple sex chromosomes is probably the result of these restrictions. The study of multiple sex chromosomes will be useful not only for understanding the mechanisms of sex differentiation in fish but also for analyses of evolutionary relationships.

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ハオコゼにみられた複合性染色体

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ハオコゼ *Hypodytes rubripinnis* の雌の核型は、2 個の中部着糸型と 46 個の端部着糸型 (2 個の付随染色体を含む) の染色体から構成され、 $2n=48$ であった。一方、雄の核型は 3 個の中部着糸型と 44 個の端部着糸型 (2 個の付随染色体を含む) の染色体で成り立ち、 $2n=47$ であった。雄の 1 個の中部着糸型染色体は対をなさず大形で、これは Y 染色体と 1 個の常染色体とが融合して生じた neo-Y と考えられた。精母細胞のデアキネシス期と第 1 減数分裂中期の像には、22 個の 2 価染色体と $X_1Y X_2$ 結合によると思われる 1 個の 3 価染色体が認められた。3 価染色体の 2 箇所の結合部のうち、一方はキアズマ型、他方は末端型を示し、前者は neo-Y の形成にあずかった常染色体と、それと相同の染色体 (X_2) との結合、後者は Y と $X(X_1)$ の結合によるものと考えられた。この $X_1X_2X_2X_2X_1X_2Y$ 型複合性染色体は、日本の 5 地域 (関東と関西、太平洋側と日本海側の地域を含む) より得られたすべての個体に所有されていたことから、本種に普遍的なものと考えられた。

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