

Karyotype Analysis in *Orthrias angorae* (Steindachner, 1897)

Taylan Özgür KAYA*

Süleyman GÜL*

Gökhan NUR*

* Department of Biology, Faculty of Science & Arts, Kafkas University, Kars-TURKEY

Yayın Kodu: 2005/41-A

Summary

Karyotype analysis was performed in *Orthrias angorae* (Steindachner, 1897) (Fam: *Balitoridae*) by investigating the number and structures of their chromosomes. The fish used in this study were caught with fishing nets from the Kura-Aras river basin and taken to the laboratory. Fishes were injected intraperitoneally (i.p.) with doses of 0.01 ml/g body weight of 0.6% solution of colchicine and left for 190 minutes before sacrifice. It was determined that *O. angorae* had $2n=50$ chromosomes by metaphase investigation. Their karyotypes were determined as being composed of 7 metacentric, 7 submetacentric and 11 acrocentric chromosome pairs with NF:78. We were unable to identify any sex-related chromosomes in this species.

Keywords: *Balitoridae*, *Orthrias angorae*, Karyotype, Kura-Aras river basin.

Çöpçü Balığı (*Orthrias angorae*, Steindachner 1897)'nda Karyotip Analizi

Özet

Orthrias angorae (Steindachner, 1897) (Fam: *Balitoridae*)'nin kromozomlarının sayısı ve yapıları incelenerek, karyotip analizi yapılmıştır. Bu çalışmada kullanılan balıklar Kura-Aras havzasından ağlarla yakalanarak laboratuvara getirilmiştir. Her bir gram vücut ağırlığı için 0.01 ml, %0.6'lık kolsişin solusyonu balıkların karın boşluğuna enjekte edilmiş ve balık kesilmeden önce 190 dakika beklenilmiştir.

Metafaz incelemeleri ile *O. angorae*'nin $2n=50$ kromozoma sahip olduğu belirlenmiştir. Karyotiplerinin; 7 metasentrik, 7 submetasentrik ve 11 akrosentrik kromozom çiftinden (NF=78) oluştuğu tespit edilmiştir. Bu türde cinsiyete bağlı herhangi bir kromozom tespit edilememiştir.

Anahtar sözcükler: *Balitoridae*, Çöpçü Balığı, Karyotip, Kura-Aras havzası.

İletişim (Correspondence)

Biyolog Taylan Özgür KAYA

Tel: +90 474 2120201 / 3085

e-mail: taylankaya391@myinet.com

INTRODUCTION

Balitoridae is represented by 36 geni and 500 species worldwide. Distribution area is Eurasia. Mouth inferior, with at least 3 pairs of barbels. Pelvic fins separate or fused under belly. In subfamily *Balitorinae*, only paired fins serve as adhesive organs about 35 cm maximum length¹.

Chromosomal analysis is important for fish breeding from the viewpoint of genetic control, the rapid production of inbred lines, taxonomy and evolutionary studies. Genetic divergence of populations and their local adaptation are a potential resource for breeding programs in aquaculture and for fishery management².

Standart karyotypes (chromosome and chromosome arm number) have been reported approximately 10% of the more than 20.000 species of fishes. The application of chromosome banding methodologies to fish chromosomes has been minimal. The main difficulty in working with fish chromosomes is to obtain high quality metaphase spreads. A few studies have used fish standart karyotypes to examine taxonomic or systematic problems^{3,9}.

The aim of this study was to investigate the chromosomes and karyotype of *O. angrae* from the Kura-Aras river basin in Turkey.

MATERIALS and METHODS

Ten females and ten males of *O. angrae* were caught in the Kura-Aras river basin [(lat 42° 15'E, long 41° 10'N), (lat 42° 30'E, long 40° 40'N), (lat 43° 10'E, long 41° 15'N), (lat 43° 15'E, long 41° 10'N)] in Eastern Turkey. The fishes were transported live to the laboratory, and kept in a well-aerated aquarium at 20-25°C before analysis.

Fishes were injected intraperitoneally with doses of 0.01 ml/g body weight of 0.6% solution of colchicine and left for 190 minutes before sacrifice. The gill filament tissues were removed and placed in hypotonic fetal calf serum (fetal calf serum diluted with distilled water, 1:7), for 45 min 4-12. They were then fixed in fresh and cold Carnoy solution (3:1) for 40 min. Staining was performed with 10% Giemsa in Sorenson buffer solution for 10 min.

Observations and microphotographs were made with a Nikon light microscope. Chromosomes were classified on the basis of the arm-length ratio⁸.

RESULTS

Relatively small and high number chromosomes were observed in *O. angrae*. In 78 metaphases from the gill epithelial cells of fifteen *O. angrae* specimens, the diploid number was found to be $2n=50$ (Figs. 1,2). Different chromosome numbers in a total of 12 metaphase cells were recorded ranging from 48 to 52 (Table 1).

O. angrae has the largest metacentric chromosome pair (Fig. 2).

Cells lacking normal values ($2n=48-52$) were probably caused by losses during preparation or additions from nearby cells. The karyotype consisted of 7 pairs of metacentric and 7 pairs of submetacentric and 11 pairs of acrocentric chromosomes (Fig. 3). The number of chromosome arms were therefore determined to be $NF=78$.

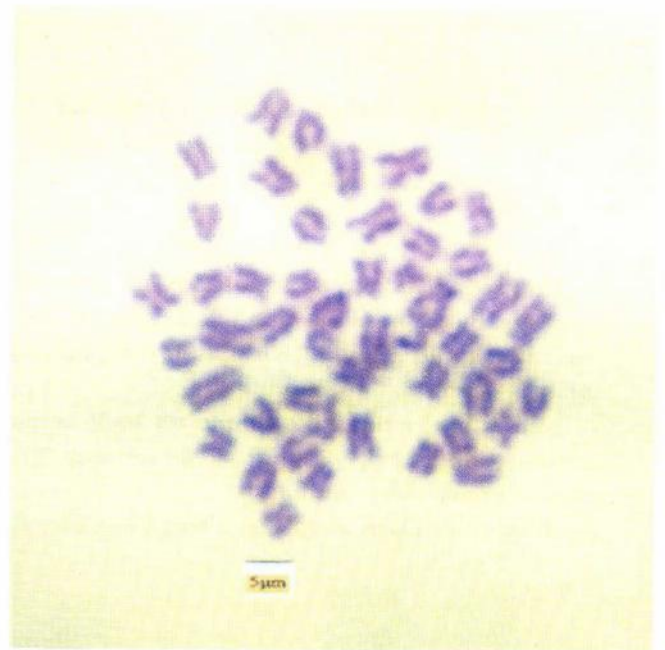


Figure 1. Metaphase spreads ($2n=50$) from gill epithelial tissue of *O. angrae* from the Kura-Aras river basin (Turkey). X 1.600. Bar, 5 μ m.
Şekil 1. Kura-Aras havzasından *O. angrae*'nin solungaç epitel dokusundan elde edilen metafaz örneği ($2n=50$). X 1.600. Bar, 5 μ m.

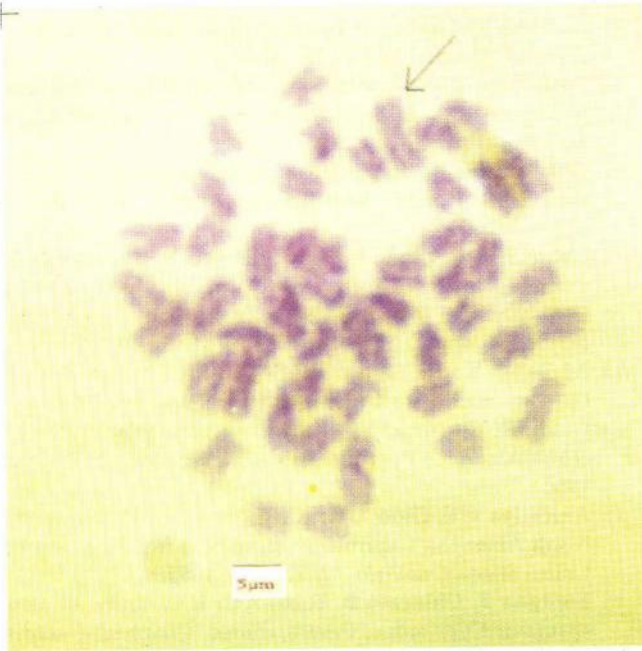


Figure 2. Metaphase spreads ($2n=50$) from gill epithelial tissue of *O. angorae* from the Kura-Aras river basin (Turkey). Largest metacentric chromosome pair, indicated by arrow. X 1.600. Bar, $5\mu\text{m}$.

Şekil 2. Kura-Aras havzasından *O. angorae*'nin solungaç epitel dokusundan elde edilen metafaz örneği. Büyük metasentrik kromozom ok ile gösterilmiştir. X 1.600. Bar, $5\mu\text{m}$.

Table 1. Chromosome complement of *O. angorae*.
Table 1. *O. angorae*'nin kromozomlarının sayısal dağılımı.

Number of fish	chromosome number				total metap hases	chromosome number			
	48	49	50	52		m	sm	a	NF
1	1		5		6	14	14	2	78
2			6	2	8				
3		1	7		8				
4			3		3				
5			7		7				
6		1	5		6				
7			6		6				
8			4		4				
9			4		4				
10	1		3		4				
11			5	1	6				
12			3		3				
13			5		5				
14			3		3				
15			5		5				
Totals	2	2	71	3	78				

DISCUSSION

Karyotypes are prepared from metaphases with well spread chromosomes. The major difficulty encountered is the morphological variation existing even between homologous chromosomes in the same nucleus^{7,13}. Sometimes it could happen that some chromosomes are more contracted than others, so chromosome measurements are very difficult, and especially in fish, which have very small chromosomes compared to those of mammals⁹. Another problem is that fish karyotypes are not identical as in human or in other animal species. Therefore, for fish, we cannot have a standart karyotype because differences not only exist between species, but polymorphism often occurs within one fish species¹³. Several incomplete metaphases were encountered in the preparation that may probably have been resulted from hypotonic overtreatment⁶.

The majority of authors classify uni-armed and bi-armed chromosomes according to the guidelines of Levan and et al⁸. In which differences in the number of chromosome arms have been reported for the same species. This is

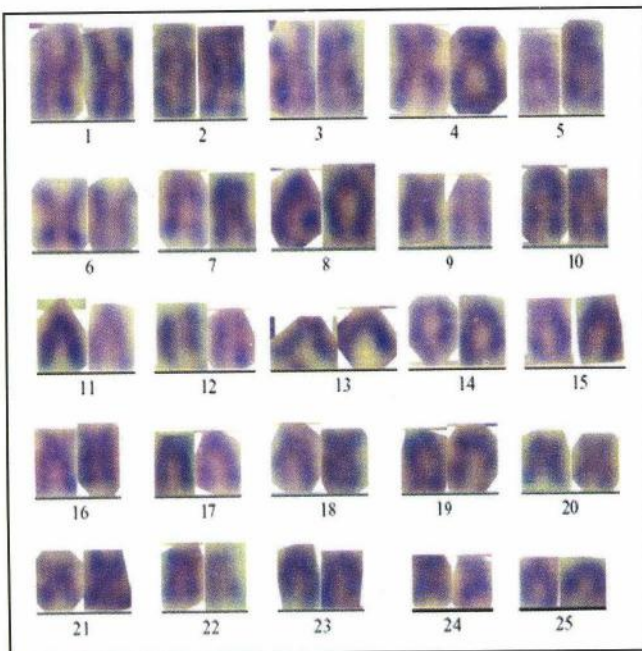


Figure 3. Karyotypes of a metaphase from *O. angorae*.
Şekil 3. *O. angorae*'nin karyotipi.

usually result of a difference in the scoring of subtelocentric chromosomes by different authors³.

The majority of cyprinid species have $2n=50$ chromosomes¹³. Gross karyotypic change in North American cyprinids appears to have been minimal: Over 90% of all species assayed (including all *Notropis* species examined) possess diploid chromosome numbers of 50 (range=48-52) and estimated diploid chromosome arm numbers between 92 and 100 (range=80-100)^{14,15}. Karyotypes have been described for specimens of *R. aula*, *S. erythrophthalmus* and *R. rubilio* all possessing $2n=50$ chromosome^{12,16} while *Cyprinus carpio* has $2n=98-100$ ⁹ and the polyploid *Barbus species* from Southern Africa have $2n=148$ or 150 chromosomes¹⁰. A number of 48 chromosomes were found in a few species, such as *Chalcalburnus mossulensis*¹¹ and *Ctenopharyngodon idella*¹⁷. Heteromorphic sex chromosomes have been identified in *Coregonus sardinella*, *Oncorhynchus mykiss*, *Oncorhynchus nerka* and *Salvelinus namaycush*². There was an XY/XX system in *S. namaycush* and *O. mykiss*, and an XYY system in *C. sardinella*². The formation of heteromorphic sex chromosomes often involves heterochromatin addition, as in other animals, and this appears to be the case in *S. namaycush* and *O. mykiss*^{2,25}.

There was no evidence of sexual dimorphism of the chromosomes in *O. angrae*. Similar results were also observed in most fish species^{3,7,9,11-13,18,26}.

REFERENCES

- 1 **www.fishbase.net**, 2005.
- 2 **Philips R, Rab P**: Chromosome evolution in the Salmonidae (Pisces): An update. *Biol Rev*, 76: 1-25, 2001.
- 3 **Bolla S**: Cytogenetic studies in Atlantic salmon and rainbow trout embryos. *Hereditas*, 106: 11-17, 1987.
- 4 **Cestari MM, Pedro M, Galetti J**: Chromosome studies of *Serrasalmus spilopleura* (Characidae, Serrasalminidae) from the Parana- Paraguay rivers: Evolutionary and cytotaxonomic considerations. *Copeia*, 1: 108-112, 1992.
- 5 **Amemiya CT, Gold JR**: Cytogenetic studies in North American minnows (Cyprinidae). *Hereditas*, 112:231-237, 1992.
- 6 **Nanda I, Schartl M, Feichtinger W, Schlupp I, Parzefall J, Schmid M**: Chromosomal evidence for laboratory synthesis of a triploid hybrid between the gynogenetic teleost *Poecilia formosa* and its host species. *J Fish Biol*, 47: 619-623, 1995.
- 7 **Vitturi R, Catalano E, Colombera D**: Chromosome analysis of *Bothus podas* (Pisces, Pleuronectiformes) from the Mediterranean Sea. *J Fish Biol*, 43: 221-227, 1993.
- 8 **Levan A, Fredga K, Sandberg AA**: Nomenclature for centromeric position on chromosomes. *Hereditas*, 52: 201-202, 1964.
- 9 **Al Sabti K**: The karyotypes of *Cyprinus carpio* and *Leuciscus cephalus*. *Cytobios*, 47: 19-25, 1985.
- 10 **Oellerman LK, Skelton PH**: Hexaploidy in yellowfish species (*Barbus*, Pisces, Cyprinidae) from southern Africa. *J Fish Biol*, 37: 105-115, 1990.
- 11 **Gül S, Çolak A, Sezgin İ**: Gümüş Balığı'nda (*Chalcalburnus mossulensis* Heckel, 1843) Karyotip Analizi. *Turk J Biol*, 24: 657-662, 2000.
- 12 **Cataudella S, Sola L, Accame Muratori R, Capanna E**: The chromosomes of 11 species of Cyprinidae and one Cobitidae from Italy, with some remarks on the problem of polyploidy in the Cypriniformes. *Genetica*, 47 (3): 161-171, 1977.
- 13 **Al Sabti K**: Handbook of Genotoxic Effects and Fish Chromosomes. J Stephan Institute, Ljubljana, 97, 1991.
- 14 **Gold JR**: Chromosomal change and rectangular evolution in North American cyprinid fishes. *Genet Res*, 35: 157-164, 1980.
- 15 **Amemiya CT, Gold JR**: Karyology of twelve species of North American Cyprinidae (minnows) from the Southern United States. *Cytologia*, 52: 715-719, 1987.
- 16 **Fontana F, Chiarelli B, Rossi AC**: Il cariotipo di alcune specie di Cyprinidae, Centrarchidae, Characidae studiate mediante coltura in vitro. *Carilologia*, 23: 549-564, 1970.
- 17 **Demirok NK, Ünlü E**: Karyotypes of cyprinid fish *Capoeta trutta* and *Capoeta capoeta umbla* (Cyprinidae) from the Tigris River. *Turk J Zool*, 25: 389-393, 2001.
- 18 **Elo K, Vuorinen JA, Niemela E**: Genetic resources of Atlantic salmon (*Salmo salar* L.) in Teno and Naatamö Rivers, northernmost Europe. *Hereditas*, 120: 19-28, 1994.
- 19 **Nishikawa S, Amaoka K, Karasawa T**: On the Chromosomes of two species of eels (*Anguilla*) Chrom Inf Serv. No:12, Shiminoseki University of Fisheries. Shiminoseki, 27-28, 1971.
- 20 **Nishikawa S, Amaoka K, Karasawa T**: A Preliminary study on the chromosomes of *Cichlasoma citrinella* (Cichlidae; Pisces), Chrom Inf Serv. No:14 Shiminoseki University of Fisheries Shiminoseki, 32-33, 1973.
- 21 **Yamazaki FA**: Chromosome study of Ayu, Salmonoid Fish. *Bull Japan Soci Sci- Fish*, Vol: 137 No:8, 707-710, 1971.
- 22 **Lloyd MA, Thorgaard GH**: Restriction endonuclease banding of rainbow trout chromosomes. *Chromosoma*, 96: 171-177, 1988.
- 23 **Hartley SE**: C, Q and restriction enzyme banding of the chromosomes in brook trout (*Salvelinus fontinalis*) and Arctic charr (*Salvelinus alpinus*). *Hereditas*, 114: 253-261, 1991.
- 24 **Jankun M, Rab P, Vuorinen J**: A karyotype study of vendace, *Coregonus albata* (Pisces, Coregoninae). *Hereditas*, 115: 291-294, 1991.
- 25 **Baker CJ**: A method for display of chromosomes of plaice *Pleuronectes platessa* and other marinefishes. *Copeia*, 2: 365-368, 1972.
- 26 **Çolak A, Sezgin İ, Süngü S**: Sazangiller (Cyprinidae) familyasına ait beni balığında *Cyprinion macrosomum* (Heckel, 1843) kromozomal araştırmalar. *Doğa Bilim Dergisi*, A2 9, 2: 193-195, 1985.