

**CHROMOSOMES OF THE PHILLIPINE SNAKE-HEAD
GUDGEON, *OPHIELEOTRIS APOROS* (ELEOTRIDAE)
FROM LAKE TAAL, LUZON ISLAND**

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ABSTRACT

Metaphase chromosomes analyzed from the anterior kidneys of *Ophieleotris aporos* obtained from Lake Taal in Luzon Island, Philippines revealed that the diploid chromosome number was $2n=46$. The fundamental number (FN) is 48 (2 submetacentric and 44 acrocentric chromosomes) was known. There was no distinguishable heteromorphic pair of chromosomes in the Giemsa-banded metaphase spreads. This is an initial report on the chromosome set of *O. aporos* in this country.

Key words : chromosomes, Philippines, Lake Taal, snake-head gudgeon, *Ophieleotris aporos*

INTRODUCTION

Family Eleotridae, which includes gudgeons and sleepers, is a large group of fishes with more than 35 genera and 150 species. Members of this family thrive in marine, brackish, or freshwater areas and are mostly found in tropical and sub-tropical regions (Allen 1982; Allen & Jenkins 1999). Gudgeons are a diverse group of teleosts and are widespread in shallow marine fresh to brackish waters of tropical to subtropical zones, particularly throughout the Indo-Pacific regions (Weber & de Beaufort 1922; 1953; Sterba 1985).

The snakehead gudgeon, *Ophieleotris aporos* is found in several regions of the world and is widely distributed in the Philippines (Conlu 1986). Synonyms of *O. aporos* are as follows: (a) *Ophiocara aporos*; (b) *Eleotris aporos* (Bleeker 1854); (c) *Eleotris tumifrons*; and (d) *Eleotris ophicephalus* (Valenciennes 1837). Although *O. aporos* is considered a food fish and an aquarium pet, it has not been investigated in the Philippines in terms of its population biology and basic chromosomal characters. Similar with other eleotridine gobies in South East Asia (SEA) (Masagca 2000; Masagca & Sumantadinata 1994), there is little information on the existing stocks or races of this fish under study.

O. aporos is known as "aporos sleeper" (from India), "snake head gudgeon" (from Fiji, Papua New Guinea and Australia) and "ornate eleotrid sleeper" in other parts of the world. In the Philippines, *O. aporos* is commonly known as "bakulihan", "bangayngay", "dalagan" or "dalak" (Conlu 1986). The early works of Villadolid (1937) and Herre (1927a,b; 1953a,b,c,d) reported the presence of *O. aporos*, while Pagulayan *et al.* (1997) recently updated the information on the present diversity of littoral fishes in Lake Taal that included *O. aporos*.

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Cytogenetics research of fishes has been gaining importance due to its contribution to aquaculture (Bye & Ponniah 1983; Thorgaard & Allen 1987; Tave 1993) and mutagenesis development (Tave 1993), in genetic conservation (Avisé 1994, 1998; Avisé & Hamrick 1998) and systematics/cytotaxonomy (Chiarelli & Capana 1973; Thompson 1979; Stevenson 1981; and Bertollo *et al.* 1986; and Rishi 1989).

This paper reports the results of a chromosomal investigation of *O. aporos* obtained from Lake Taal in Luzon Island, Philippines. Specifically, this paper provides benchmark information on the somatic chromosome number, fundamental number and karyotypic formula of the fish under study.

MATERIALS AND METHODS

Collection sites

Fish samples of *O. aporos* were obtained from Lake Taal (formerly Bombon Lake), Luzon Island, Philippines (14:00N, 121:19E; 2.5 m above sea level). This is an important freshwater lake situated 72 km south of Manila (capital city of the Philippines). In particular, the collection sites are in the Volcano Island or "Pulo" I and II of Lake Taal. This lake is a caldera, the third largest lake in the country whose most violent eruptions date back 5000 to 7000 years ago. The lake has a surface area of about 26,368 ha, perimeter of 120 km, average depth of 60.1 m and with a maximum depth of 198 m (Castillo & Gonzales 1975). There are 37 tributary streams and rivers that drain its 656.5 sq. km watershed.

Identification of fish specimens

O. aporos samples were identified according to the descriptions using Fish Base of the International Center for Aquatic and Living Resources (ICLARM), Food and Agricultural Organization (FAO) Identification Sheets and several taxonomic works such as Smith (1965), Sterba (1985), Mohsin and Ambak (1983), Conlu (1986), Nelson (1994) and Larson & Murdy (2001).

Karyotype analysis

Each *O. aporos* sample was given intramuscular injection of colchicine (Sigma) at 0.05% in 0.8% NaCl, 5-6 hours (h) prior to sacrifice by hypothermia. For smaller specimens, colchicine treatment was done by allowing *O. aporos* swim in 0.05% of the drug for 5-6 h in well-aerated aquaria. After colchicine treatment, the specimens were sacrificed by decapitation or hypothermia (in cracked ice for 3 to 5 minutes). The kidneys were dissected out, carefully cleared of blood vessels and placed in a petri dish containing 0.5 - 0.6% sodium citrate for hypotonization. Kidney tissues were minced with fine surgical scissors, centrifuged for 4-5 minutes at 1500-3000 rpm and fixed with Carnoy's solution. The chromosome preparation followed the techniques as modified from the works of Gold (1974), Kligerman & Bloom (1977), Rivlin *et al.* (1985),

Reddy & John (1987) and as used by Masagca (2000) and Masagca & Sumantadinata (1994). Both conventional and differential staining techniques were tried as follows: 1) 4-5% Giemsa at pH 6.8; 2) G-banding by ASG (Acetic-Saline - Giemsa) technique; and 3) C - banding by BSG (Barium hydroxide-saline-Giemsa) (Denton & Howell 1969; Denton 1973).

A total of 22 fish samples were used for direct chromosome preparation from the kidney metaphase cells. A total of 703 cells prepared from the anterior kidney were examined for this investigation.

RESULTS AND DISCUSSION

Chromosomes were counted in well spread metaphase cells. As shown in Figure 1, out of the total 703 metaphase cells examined from 2 collection sites in Lake Taal (Pulo I and Pulo II), 435 cells (61.9%) have the characteristic diploid chromosome number of 46; 160 cells (22.8%) have 44 chromosomes; 44 cells (6.3%) have 42; 23 cells (3.3%) have 43; 22 cells (3.1%) have 47 and 8 cells (1.1%) have 48.

Out of the 267 metaphase cells analyzed from samples of *O. aporos* collected from the waters off Pulo I, 192 cells (71.91%) have the characteristic count of $2n=46$; 51 cells (19.5%) have a count of $2n=44$; 9 (3.4%) cells with a count of $2n=42$; 7 (2.6%) cells have $2n=45$; 3 (1.12%) each with $2n=47$ and 48 counts and 2 (0.75%) cells have a count of 43.

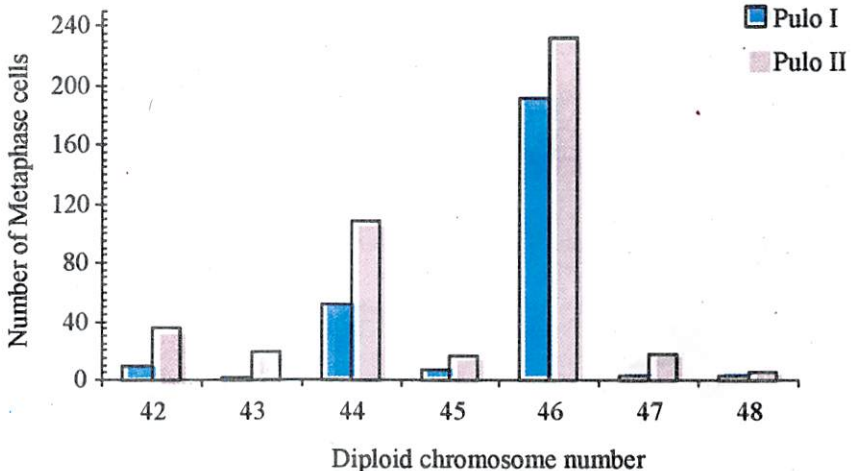


Figure 1. Chromosome counts of *O. aporos* from different locations in Pulo I and II, Lake Taal, Luzon Island, Philippines.

Chromosome counts made on 436 metaphase spreads from Pulo II fish samples revealed that 243 (55.73%) have the count of $2n=46$; 109 (25%) metaphase cells have chromosome number of 44; 35 (8.03%) have $n=42$; 20 (4.6%) metaphase cells have 43; 18 (4.13%) metaphase cells with $2n=47$; 16 (3.7%) cells with $2n=45$ and 5 (1.15%) metaphase cells with the count of 48.

The karyotype of *O. aporos* consists of 2 submetacentric chromosomes and 44 acrocentric chromosomes. Similar pairs of chromosome were arranged in decreasing order of sizes. There were no morphological differences that were observed in the karyotypes prepared.

The cytogenetic data on *O. aporos* indicates that $2n=46$ falls within the range of diploid chromosomes of most eleotrids. The tentative Nombre Fundamental or NF was found to be 48 because there were 2 bi-armed chromosomes (SM) and 44 acrocentric chromosomes (A) in the karyotype.

The fundamental number of 48 for *O. aporos* is lower than the previously studied eleotrid, *O. marmoratus* (NF=50) from Indonesia (Masagca 2000; Masagca & Sumantadinata 1994) as well as those from Thailand (Arai and Fujiki 1979) and from India (Manna 1989). In comparison with a goby (*Eleotris picta*) from Mexico, Del Carmen-Maldonado *et al.* (1985) obtained an NF of 90, which means that most of the chromosomes are bi-armed.

In the Gobiidae family, most of the genera have chromosome number of $2n=44$ to $2n=48$, such as in *Bathygobius fuscus* ($2n=48$) and *Chaetogobius annularis* ($2n=44$). The chromosomes of gobies and eleotrids show variability from $2n=43$ to 62 (Masagca 2000), with most of the chromosome numbers are $2n=44$, 46 and 48. In another study, the karyotype of *G. microdon* has a diploid number of $2n=56$ and an NF of 66 with a chromosome formula of $4M+6SM+46ST, A$.

The diploid chromosome number of *O. aporos* is also similar to the findings of Arai & Fujiki (1979) for *Eleotris acanthopomus*. Other eleotridine species having the same chromosome number are *Chaenogobius striatus*, *Boleophthalmus dussumieri* and *B. boddaerti* (Arai & Sawada 1974). In another eleotrid, *Eleotris picta* has 52 uniarmed chromosomes (Uribe-Alcocer & Díaz-Jaimes 1996; 2000).

Variability in chromosomal counts would lead to certain generalization of the possibility of changes in chromosomal number due to fusions, translocations and other mechanisms. However, this is not conclusive in the present study since there is a need for further chromosomal banding studies and constancy of variation in the counts. In some studies, like the paedomorphic goby, *Aphia minuta* (Gobiidae) wherein the diploid complement ranged from 44 to 41 due to Robertsonian fusions (NF=44). Another case of Robertsonian fusion was known in the gobiid, *Gobius paganellus* (Giles *et al.* 1985). Data on spermatogenesis suggest that structural heterozygotes are fertile and that these chromosomal changes are not involved in speciation process (Caputo *et al.* 1999).

In this study, characteristic counts of 41, 42, 43, 44 and counts higher than 46 in the test animal (*O. aporos*) were observed. Variations in chromosome number maybe attributed to several factors: (1) handling techniques; (2) chemically induced; and (3) inherent genetic characteristic of the test fishes. Handling techniques would explain the variability in chromosome counts. However, in the case of the eleotrid *O. aporos*, variability in chromosome number seems to be similar with other eleotrids, e.g. *P. glehni* having pericentric inversion (Manna & Prasad 1977).

Considering that the karyotype of this eleotridine species, *O. aporos* was not previously described, the cytogenetic analysis by the conventional techniques, do not permit a comparative study with the pattern of the species in the SEA region. The karyotypes of other gobioids such as *G. giuris* previously described in Japan, India, the Philippines (Masagca & Ordonez 2003) and elsewhere could permit the researchers to have further comparison using the conventionally stained chromosomes and in the future the banded chromosomes from fully elongated chromosomes.

The chromosome number of *O. aporos*, which is $2n=46$ is common to the Order Perciformes. This number is also known in *Selene setapinnis* (family Carangidae) as described by Netto *et al.* (1998). In fishes, 48 rod-like chromosomes have been considered to be the modal number as shown in the works of Nogusa (1960) and Roberts (1967). It was Manna (1989) who advocated that 48 chromosomes of mixed morphology and only rods were the modal ones from which the evolution of different karyotypes can be envisaged.

CONCLUSIONS

This study concluded that the Philippine snakehead gudgeon, *O. aporos* has a diploid chromosomal formula of $2n=46$. The fundamental number (FN) is 48 (2 submetacentric and 44 acrocentric chromosomes) was known. There was no distinguishable heteromorphic pair of chromosomes in the Giemsa-banded metaphase spreads. Characteristic chromosome counts ranged from 41 to 44 and some counts where higher than 46. This is an initial report on the chromosome set of *O. aporos* in the Philippines.

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