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## **Determination of the Gynogenetic Reproduction Character of *Carassius gibelio* in Uluabat Lake**

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### **ABSTRACT**

The purpose of this study was to examine the status of ginogetic population Uluabat Lake *Carassius gibelio*. In this study, 466 individuals were caught per month between March 2006 and February 2007. It has been determined that 65% of the obtained samples are comprised of females, 34% is comprised of males and 1% is comprised of individuals, which are not mature sexually. Chromosome photographs were shoot by preparing chromosome preparations from the gills of 5 female individuals caught in the lake and in the chromosome remuneration based on these photographs it has been observed that 4 individuals has  $2n = 100$  chromosomes whereas one individual has  $3n = 156$  chromosomes. This result indicates that the *C. gibelio* population in Uluabat Lake is comprised of different colonies including diploid and triploid individuals.

**Key words:** Uluabat lake, *Carassius gibelio*, gynogenesis, chromosome number

### **INTRODUCTION**

Eggs and fries, which belong to 25 exotic species, have been introduced in Turkish waters for various purposes for more than fifty years. One of these exotic species is *C. gibelio* (Innal and Erkakan, 2006). This species is common in Western Asia, Siberia and the entire Europe (Kottelat, 1997). However, it is not one of natural species in European continent (Flajshans *et al.*, 2008). Only its occurrence in Northern Europe may be natural (Kottelat, 1997). *Carassius gibelio* entered into Turkey through River Maritza over Greece or Bulgaria or it was carried by people (Ozulug *et al.*, 2004). This species was observed in Thrace region in Turkey, it was reported from 46 fresh water systems in Thrace and Anatolia (Ozcan, 2007).

*Carassius gibelio* is a specie, which is omnivorous (Yilmaz *et al.*, 2007). They have sperm-based partogenetic (gynogenesis) reproduction. They establish populations with different genetic characteristics in different regions due to their gynogenetic reproduction capability (Jia *et al.*, 2008). They form diploid-polyploidy populations, in which most of the individuals are composed of female species, in natural environments as a result of gynogenetic reproduction (Kalous, 2002). It is thought to be this gynogenetic trait of *C. gibelio* that enables it to dominate marshy areas so rapidly and successfully. Thus, the majority of the population created by *C. gibelio*, which is infamous for stealing the sperms of the males belonging in other species, turns out to be females with  $3n$  chromosomes and so leads to a destructive competition for reproduction among fish. This being the case, it so appears that it is of vital importance to know reproductive ability of this

species and take necessary precautions accordingly in coping with it. In order to confirm that *C. gibelio* reproduces gynogenetically, it has been suggested that chromosome numbers of this fish be determined (Liasko *et al.*, 2010).

From Turkey, the population comprising triploid individuals as a consequence of the characteristic of gynogenetic reproduction has been notified to be observed in Kayaliköy Dam Lake and Porsuk Dam Lake (Kalous *et al.*, 2004; Emiroglu *et al.*, 2010). The structure of female weighted population which is an indicator of gynogenetic reproduction has been notified to be observed in Ömerli Dam Lake (Tarkan *et al.*, 2006) Bafra Fish Lake (Bostanci *et al.*, 2007) Topçam Dam Lake (Sasi, 2008) Buldan Dam Lake (Sari *et al.*, 2008).

By means of this study, the chromosome numbers of the *C. gibelio* species has been determined for the first time in Turkey and it has been set forth that it reproduces gynogenetically in Uluabat Lake.

## MATERIALS AND METHODS

Surface area of Uluabat Lake is 160 km<sup>2</sup>, its length in eastern-western direction is 25 km, its width in northern-southern direction is 10.5 and it is a shallow lake, which is situated in provincial boundaries of Bursa (Inan, 1999) (Fig. 1).

In this study, each month between March 2006 and February 2007, gill nets with various sizes (in mesh sizes of 18, 20, 25, 28, 32 and 40 mm) were used and 466 *C. gibelio* individuals were caught and their gender was determined by means of gonad examination. Five female individuals caught from the lake were carried to the laboratory by means of an oxygen bottle. The fish brought to the laboratory were kept in an aquarium for 7 days without feeding in order to let them get used to the laboratory environment. Then, they were intraperitoneally injected with 0.1% phytohemagglutinin (PHA) for every 100 g of body mass. Injected fishes were held in well aerated aquariums for 48 h without food. Fishes were anaesthetized with ether and were killed by cutting their gills with scalpel. The gills that were taken out were put into test tubes with 0.075 KCl and were kept in 30°C for 65 min. The solution was put into 2000 rpm centrifuge for 10 min and the supernatant was aspirated away. Seven milliliter of 3:1 methanol: GAA (Glacial Acetic Acid) was put in every tube for fixation and was waited 15 min in 4°C. Then it was put into 2000 rpm centrifuge at for 10 min and supernatant aspirated away, where in this step was repeated twice.

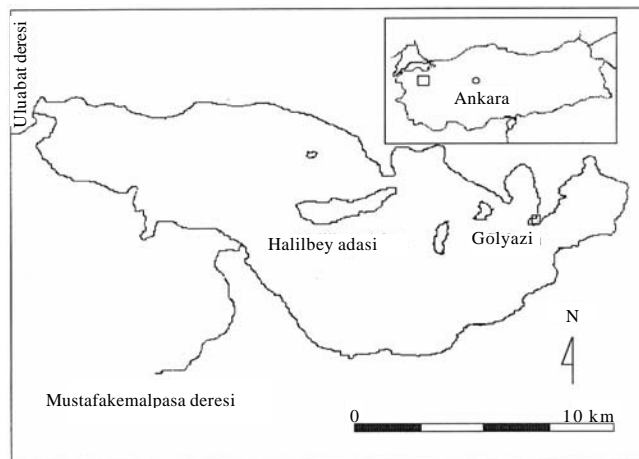


Fig. 1: Map of lake Uluabat

After 3rd centrifuge most of the supernatant was aspirated away and 2-3 drops of cell suspension that is left was stirred up well. 1-2 drops from 40-50 cm height on the slides of those were washed with 50% methanol and were dried. They were dried for 1 h at 60°C. Ninety five milliliter of 5 mL+pbs giemsa solution was taken and filled up to 100 mL and the slides were put in paint for 35 min at room temperature. The painted slides were shaken two times with acetone and were dipped into 1:1 Acetone: Xylol solution then were shaken 2 times with xylol and put into xylol for 5 min. After a while slides were fixed with enthellan. Pictures of chromosome were taken in 100x10 zoom.

## RESULTS

It has been determined that among the *C. gibelio* individuals caught from Uluabat Lake, approximately 65% is female, 34% is male and the remaining approximately 1% is comprised by the individuals which have not yet reached to gonadic maturity. The proportion of males to females has been found to be 0.52/1 and it has been observed that the female individuals are more dominant compared to the male individuals. The photographs of the preparations prepared from the gill tissues of 5 female individuals brought to the laboratory for their chromosome numbers were taken (Fig. 2 and 3). Based on the chromosome preparations, diploid individuals with  $2n = 100$  chromosomes and triploid individuals with  $3n = 156$  chromosomes formed by gynogenetic reproduction have been determined.

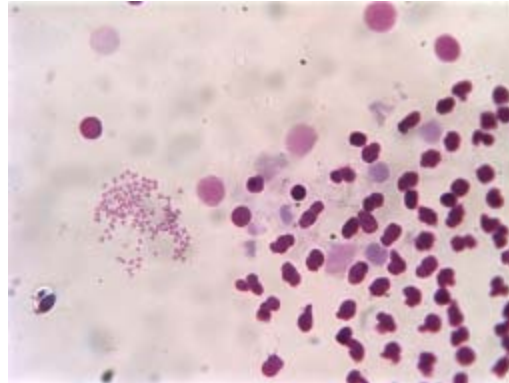


Fig. 2: Individual with  $2n = 100$  chromosomes

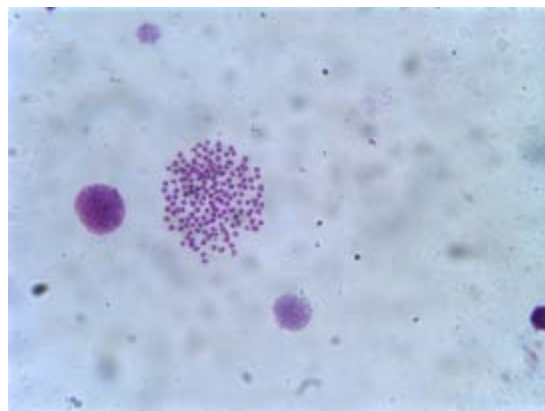


Fig. 3: Individual with  $3n = 156$  chromosomes

## DISCUSSION

Because of the advantages particularly provided in cultural fishing, the populations comprised of triploid individuals are preferred. Together with the developing technology, the studies for suppression of the chromosomes related with gender formation and for creating individuals with more or larger chromosomes have been very successful. Triploid fish manufacturing can be used as a biotechnological means for realization of some certain management purposes besides the studies for increasing productivity.

The first source of inspiration of these methods is the species which can achieve this task naturally in the wild. Particularly, *C. gibelio* has a unique reproduction skill which is performed without being based on a male, namely the reproduction by gynogenesis. In this manner, they form colonies with different chromosome numbers in the same environment. Since the triploid individuals have a higher power of reproduction and adaptation compared to the other diploid fish, they become the dominant species in their surrounding of invasion.

It was reported that the individuals which arise when *C. gibelio* eggs are stimulated with *Cyprinus carpio* consist of 98% female and 2% male and 15% of the bisexual generation which arises as a result of crossbreeding of male and female individuals is male individuals (Fan and Shen, 1990). Pipoyan and Rukhkyan (1995) reported that *Carassius auratus gibelio* gynogenetically reproduces and Paschos *et al.* (2004) reported that the female individuals in Pamvotis Lake in Greece which gynogenetically reproduce and have 150 chromosomes are sperm parasites and they gynogenetically reproduce with the sperms of other *Cyprinidae* species and therefore 97.7% of the population consists of female individuals. The fundamental chromosome number of the *C. gibelio* species is  $2n = 100$ . However, *C. gibelio* can have  $3n = 156$  chromosomes by means of gynogenesis (Liasko *et al.*, 2010).

There are various studies which demonstrate the *C. gibelio* species reproduces by gynogenesis, which has invaded many fresh water system (Kalous *et al.*, 2004; Emiroglu *et al.*, 2010; Tarkan *et al.*, 2006; Bostanci *et al.*, 2007; Sasi, 2008; Sari *et al.*, 2008). However, for the first time, it has been determined based on the chromosome numbers that *C. gibelio* species reproduces gynogenetically and create individuals with  $3n = 156$  chromosomes. It is observed that each individual has  $3n = 156$  chromosomes while 4 individuals whose chromosome preparations are prepared have  $2n = 100$  chromosomes.

## CONCLUSION

The European and Turkish fresh water systems, which are severely invaded by *C. gibelio* are losing their productivity and water quality under the effect of this invader species. The most significant factor to cope with these invader species and to protect the fresh water system is to know the characteristics of the species you cope with. This study has determined the chromosome numbers of the *C. gibelio* population in Uluabat Lake and the respective gynogenetical reproduction type. This should be taken into consideration in the future studies for protection of the Uluabat Lake.

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