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RESEARCH ARTICLE

Constitutive heterochromatin in *Acanthobrama marmid* and *Cyprinion macrostomus* (Osteichthyes, Cyprinidae)

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Summary

Constitutive heterochromatin were studied in two Cyprinids *Acanthobrama marmid* and *Cyprinion macrostomus* in Turkey. The C-band patterns of the metaphase chromosomes of these species were reported. Diploid chromosome number was 2n=50 in all specimens. The C-bands were seen in the pericentromeric regions of all chromosomes in both species. In addition C-bands were observed on the short arms of two pairs of submetacentric chromosomes of *A. marmid* and on the short arms of two pairs of submeta-subtelocentric chromosomes of *C. macrostomus*. There were similarities between C-band blocks and NOR regions. C-band patterns were also similar in both species.

Keywords: *C-banding, constitutive heterochromatin, Acanthobrama marmid, Cyprinion macrostomus, Cyprinidae*

Acanthobrama marmid ve *Cyprinion macrostomus* (Osteichthyes, Cyprinidae)'un Konstitütif Heterokromatini

Özet

Türkiye'de yaşayan iki Cyprinid *Acanthobrama marmid* ve *Cyprinion macrostomus* üzerinde konstitütif heterokromatini araştırıldı. Bu türlerin metafaz kromozomlarından C-band kalıpları rapor edildi. Bütün örneklerde diploid kromozom sayısı 2n=50 bulundu. Her iki türde de bütün kromozomların perisentromerik bölgelerinde C-band görüldü. Ayrıca *A. marmid*'de iki çift submetasentrik kromozomun kısa kollarında ve *C. macrostomus*'ta iki çift submeta-subtelosentrik kromozomun kısa kollarında C-band gözlendi. C-band blokları ve NOR bölgeleri arasında benzerlikler bulundu. Her iki türün C-band kalıpları arasında da benzerlikler mevcuttu.

Anahtar sözcükler: *C-bandlama, konstitütif heterokromatin, Acanthobrama marmid, Cyprinion macrostomus, Cyprinidae*

INTRODUCTION

Acanthobrama marmid (Heckel, 1843) and *Cyprinion macrostomus* (Heckel, 1843) were distributed eastern Anatolia, Middle East and Arabian Peninsula. One hundred sixteen species and subspecies of Cyprinidae have been reported from Turkey by Kuru [1]. However, cytogenetic studies, concerning fish in this country are not sufficient. There are a few karyological studies and yet studies about [2-6], C-banding is really limited [7]. Karyotypes and nucleolus organizer region (NOR) characteristics of *C. macrostomus* and *A. marmid* were studied by different researchers [4,6,7]. C-banding, used to establish heterochromatin regions on the chromosomes is frequently used in plants and animals specially in mammals and fish and thus is useful for examining intra and interspesific chromosomal differences between closely related species [8-10]. C-band patterns are all of similar in pericentromeric heterochromatin among plants and animals. However, there is great variation in the distribution of heterochromatin on chromosomal arms. C-bands are found at or around pericentric region and

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frequently of the telomeric region and frequently at the telomeres. C-bands may also be found within chromosomal arms and occasionally the short arms of acrocentric chromosomes may be entirely heterochromatin. Investigations about heterochromatin differentiation and C-band patterns of fish chromosomes are far behind the studies of all other vertebrates due to the common reason that relates to the general difficulty is working with fish chromosomes. There may be differences in heterochromatin regions within chromosomal arms among related genera. Thus in this paper firstly, we made an attempt to establish the distribution of heterochromatin and secondly, thus chromosomal evolution and cytotaxonomy of two closely related species *A. marmid* and *C. macrostomus*.

MATERIAL and METHODS

Adult males and females of *A. marmid* (n=8) and *C. macrostomus* (n=7) were collected from Sultansuyu River in Malatya (38° 26' N, 38° 08' E), Turkey at 2006-2007. Chromosome preparations were made from head kidney according to the method of Collares-Pereira with slight modifications [11]. The C-bands were stained by the techniques of Sumner [12]. The preparations observed and photographed digitally at Leica DMLB research microscope. The nomenclature of Levan et al. was used to describe to chromosome morphology [13]. Specimens analyzed were deposited as vouchers in the Cytogenetics Laboratory of Department of Biology, Faculty of Science and Arts, University of Ahi Evran, 40200, Kirsehir, Turkey, M. Gaffaroglu (M.G. 30, 31).

RESULTS

The diploid chromosome number in both species is 2n=50. There were eight pairs of metacentric, thirteen pairs of submetacentric, and four pairs of subtelocentric to acrocentric chromosomes in the karyotypes of *A. marmid* and three pairs of metacentric, twelve pairs of submetacentric and ten pairs of subtelocentric to acrocentric chromosomes in the karyotypes of *C. macrostomus*. No heteromorphic sex chromosomes were detected in both species. C-band analysis revealed that constitutive heterochromatin was located at pericentromeric regions of all chromosomes in both species. Beside this, C-bands were observed on the short arms of one or two pairs of submetacentric chromosomes in *A. marmid* and on the short arms of one or two pairs of submeta-subtelocentric chromosomes of *C. macrostomus*. Heterochromatin blocks on the arms were bigger and more prominent. There were differences in the distribution of C-bands within the two species. That was to say those C-band patterns within species were heteromorphic. In other words there were C-bands on one pairs of chromosomes of some specimens and on one pairs of chromosomes on the others. Pericentric C-band regions were small and weakly dyed. In fact weak colour of heterochromatin region was characteristic for both species. C-band patterns were similar in the two species. That was the locality and the amounts of heterochromatin in two species were almost similar. On the other hand there were similarities between NOR location and C-band patterns.

DISCUSSION

C-bands indicate the presence of constitutive heterochromatin associated with ribosomal genes. Similar results were obtained by Boron [14]. However, there are no report of C-banding study on A. marmid and C. macrostomus. No significant differences have been observed between sexes both in the amount and in the location of heterochromatin. C-banded metaphases are shown in *Fig.1* and *2*. As shown, the amount of heterochromatin in these two species was considerably less than euchromatin, similar results have been obtained by Rabova et al.[10]. However, reported that the amount of heterochromatin was up to 60-70 % of the total chromosomes in North American Cyprinids [8].

The amount of heterochromatin is of interest to our long terms goals of understanding the chromosomal evolution and cytotaxonomy. In general extensive heterochromatin content indicates extensive potential to vary or vice-versa. From this point of view, we are far from satisfaction to evaluate evolutionary mechanism C-band pattern of chromosomal speciation in fish due to inadequate amount of data.

Gül et al.[7] in *Chalcalburnus tarichi* (2n=50) and Rabova et al.[10] in *Vimba vimba* (2n=50) and in *V. elongate* reported that there were small hetero-

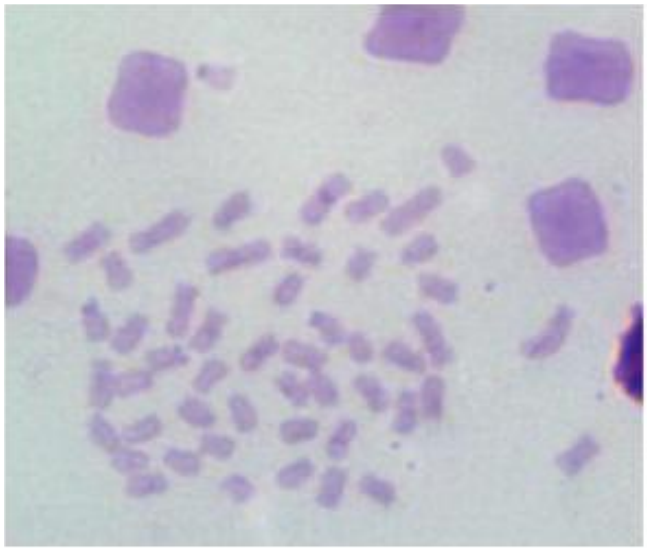

Fig. 1. C-banding pattern of *Acanthobrama marmid*
Şekil 1. *Acanthobrama marmid*'de C-band kalıbı

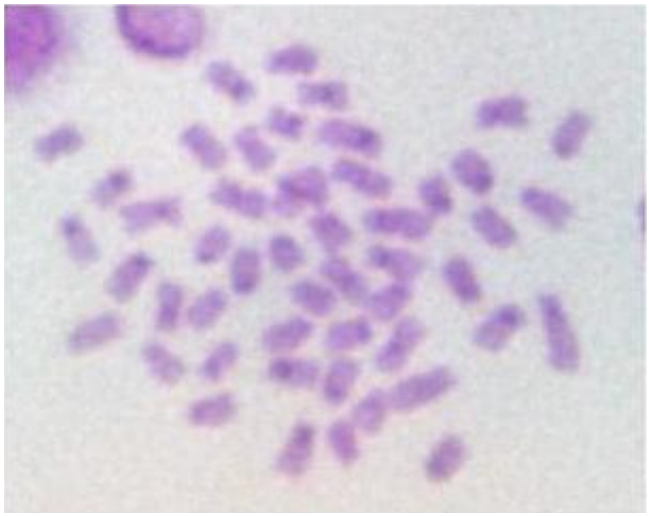

Fig. 2. C-banding pattern of *Cyprinion macrostomus*
Şekil 2. *Cyprinion macrostomus*'da C-band kalıbı

chromatic blocks in pericentromeric regions of all chromosome. In addition Rabova et al. reported in *V. vimba* that the largest subtelo-acrocentric pair of chromosomes showed size heteromorphism for distal blocks of heterochromatin and all the chromosomes in variably posses is C-bands around the centromeres [10]. These results are in agreement with our results in the present study.

Most species invariably possess C-bands at or around the centromeres (procentric) and frequently at the chromosome tips (telomeric). C-bands may also be found along chromosome arms (interstitial) and as entirely heterochromatic short arms of acrocentric chromosomes. The variation in such "short-arm" heterochromatin was particularly impressive in the rodent genera [8]. It was clear from inspection of the karyograms that many chromosomes in both species were very similar in relative size, centromer position, and C-banding. This was especially true for most of the chromosomes with short-arm heterochromatin.

In general chromosome arm number variation which differentiate species in both genera. The extensive heterochromatin content indicates a considerable potential to vary. Many natural populations contain substantial amounts of telomeric and interstitial heterochromatin which exist in a polymorphic state. Crossover events may occur away from these heterochromatin blocks [15]. Thus, the immediate effect of this is to modify the relative frequency of gene combination and resulting differentiation of related species.

Actually it has been possible to demonstrate the effect of heterochromatin on crossing-over by different ways i.e. ***i.*** By the use of classic gene markers in an organism. ***ii.*** By the use of inversion markers. ***iii.*** By an analysis of chiasmic pattern. [iv.] By PCR.

It has long been evident that the amount of heterochromatin was not necessarily constant from individual to individual within a species. However, it is important to give detailed attention to expensive heterochromatic variation now recognized in many natural populations. This variation extends all the way from large, C-banded blocks to the level of individual polytene bands. Increases and decreases in C-band material appear to be common in most species.

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