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Cytogenetic Investigations on the Wild Common Carp (*Cyprinus carpio* L.) from Vinh-Phu Province of Capital North Vietnam

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ABSTRACT

Cytogenetic investigations were carried out on wild common carp (Cyprinus *carpio L.*) caught from the Hong river (Red river) in North Vietnam. Diploid chromosome number, Karyotypic configuration and the number of Nucleolus Organizer Region (NOR)-bearing chromosomes was determined. Silver staining of mitotic chromosomes of common carp revealed an existence of two non-homologous sub-metacentric chromosomes bearing NORs on their entire upper shorter arms. Sequential counterstain enhanced fluorescent Chromomycin A3 also confirmed the same results.

Key Words: Common carp; Diploid chromosomes; Cytogenetics; Metaphase plates

INTRODUCTION

Cyprinus carpio L. is a teleostean species having a tetraploid origin. Inspite of the fact that carp possesses a large number of very small chromosomes, it has been fairly well studied by the cytogenetic researchers. In a majority of these studies, a diploid chromosome number has been reported to be 2n = 100 in common carp (Raicu *et al*., 1972; Denton, 1973; Zan & Song, 1980; Blaxhall, 1983; Labat *et al.*, 1983; Rab *et al*., 1989; Larka & Rishi, 1991; Anjum & Jankun, 1994; Anjum, 1995). Diploid chromosome number of native carp from Amur river has been divided into eight well-defined groups on the basis of their morphology and a standard karyotype has also been proposed (Rab *et al*., 1989). C-banding and Silver-NOR staining has also been employed in some cytogenetic studies on common carp (Takai & Ojima, 1982; Ruifang *et al*., 1985; Sola *et al*., 1986).

In most of these cases, two functionally active NORs have been detected. However, Sola *et al.* (1986) have postulated that only one pair of NOR-bearing chromosomes is normally active while the other pair is probably inactive and thus invisible in carp which has phylogenetically a tetraploid origin. Studies to confirm the number and localization of NORs in some Cyprinids have also been carried out by using the counterstain-enhanced fluorescent Chromomycin A3 technique (Mayr *et al*., 1986). Present investigations were aimed at studying the chromosome number, karyotypic arrangement and number of active NORs in the wild carp from Vietnam.

MATERIALS AND METHODS

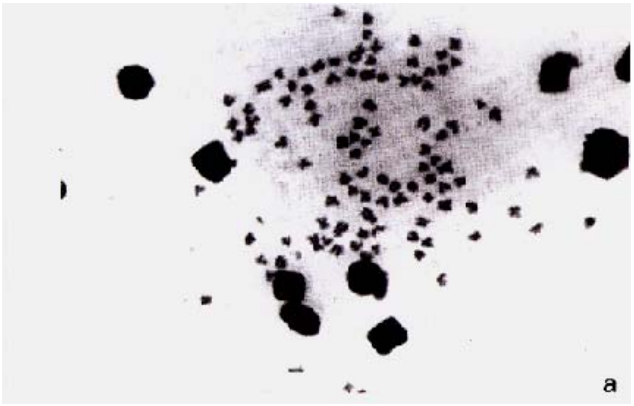
Twenty four specimens of wild common carp were collected from the section of Hong river flowing through Vinh-phu province of North Vietnam. These specimens were received in the Cytogenetics Laboratry of the Institute of Freshwater Ecology and Inland Fisheries, Berlin, Germany.

Standard procedures for chromosomal preparation from the head kidney tissue were used (Rab & Roth, 1989). Silver-NOR staining was performed according to the One-step method developed by Howell and Black (1980). Sequential fluorescent staining with Chromomycin A3 was subsequently carried out as described by Sola *et al*. (1992). Photomicrography of selected metaphase plates was carried out on the Nikon microscope (Model OPTIPHOT-2) using fluorescent lamp HBO 100 with filter set BV 2a.

RESULTS

The diploid chromosome number of common carp specimens from Vietnam was found to be 2n = 100. Giemsa stained metaphase spread showing a diploid set of 100 chromosomes is presented in Fig. 1. The Karyotypic configuration comprised of 8 pairs of metacentric, 17 pairs

Fig. 1. Metaphase spread of common carp (2n = 100)



of submetacentric and 25 pairs of subtelo-and acrocentric chromosomes (Fig. 2). Silver-NOR staining of metaphase spreads in all the cases revealed an existence of two non-homologous submetacentric chromosomes (of two different sizes) bearing Nucleolus Organizer Regions (NORs) on their entire upper shorter arms (Fig. 3). A sequential staining of the same metaphase spreads with Chromomycin A3 resulted in the heterochromatin staining of NORs which appeared as bright zones against a dark background (Fig. 4).

Fig. 2. Karyotypic Configuration of 2n = 100

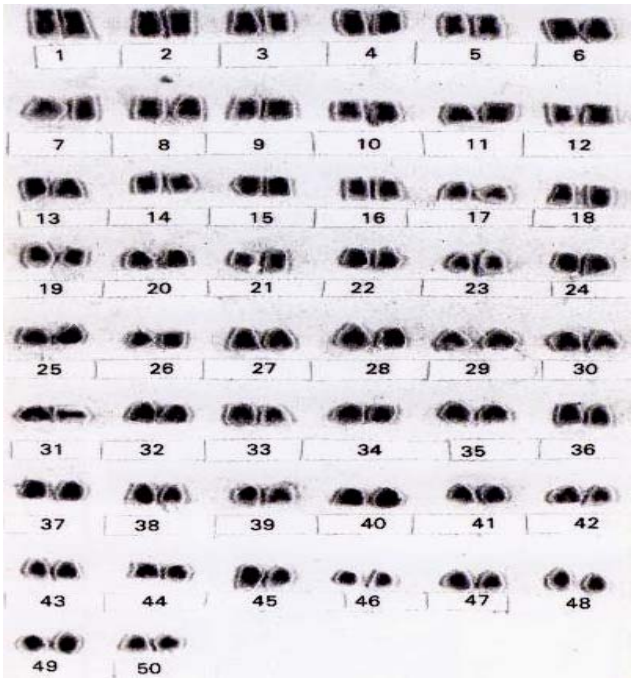


Fig. 3. Silver nitrate stained metaphase plate of common carp showing two Non-homologus NOR-bearing Chromosomes (arrows).

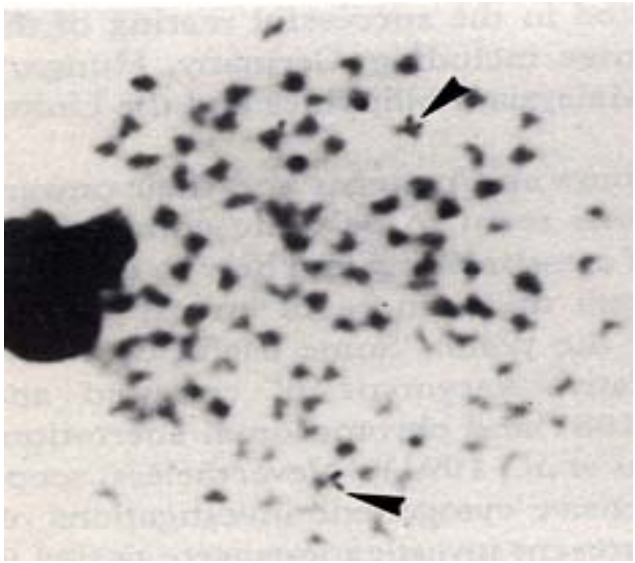
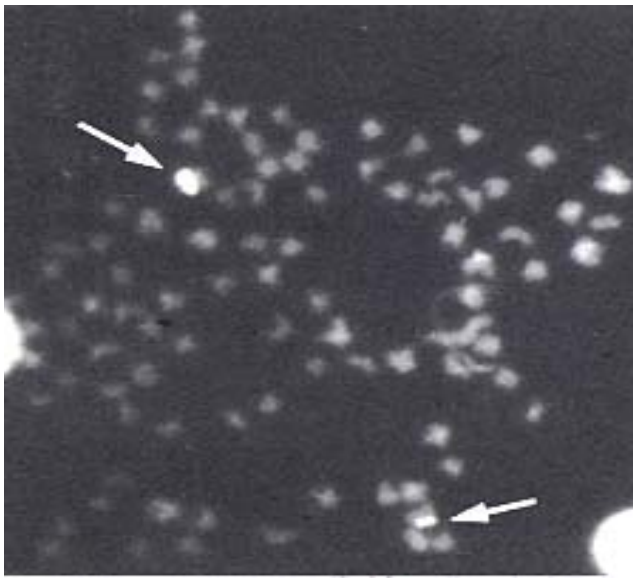


Fig. 4. Sequential Chromomycin A3 staining of the same metaphase spread showing bright heterochromatic staining of small NOR-bearing Chromosomes (arrows).



Exactly the same number and patterns of NOR-bearing chromosomes were observed through fluorescent staining as were observed through the Silver Nitrate staining.

DISCUSSION

Cytogenetic analysis of wild carp from Vietnam revealed a diploid chromosome number of 2n = 100. In a large majority of previously carried out investigations of common carp, similar findings i.e. 2n = 100, have been reported (Raicu *et al*., 1972; Denton, 1973; Zan & Song, 1980; Blaxhall, 1983; Rab *et al*., 1989; Larka & Rishi, 1991; Anjum & Jankun, 1994; Anjum, 1995). Some variations in the diploid chromosome number have, however, been reported in certain cases, pertaining to the presence of two or more micro chromosomes (Ohno *et al*., 1967; Sola *et al*., 1986; Al Sabti, 1986; Brzuska, 1988).

Karyotypic configuration also varied in several cases, mainly because of very small size of carp chromosomes which are very often very difficult to arrange in separate classes. Silver-NOR staining has revealed an existence of two sub-metacentric chromosomes (belonging to two different pairs) having different sizes and bearing Nucleolus organizer regions (NORs) on their entire upper shorter arms. Similarly an expression of two functionally active NOR-bearing chromosomes has previously been detected by some researchers through the use of silver staining. (Takai & Ojima, 1982; Ruifang *et al*., 1985; Mayr *et al*., 1986; Sola *et al*., 1986; Anjum, 1995). Certain intra-specific differences have very clearly been observed for the number of NOR-bearing chromosomes within the colored (Koi) carp

population where the presence of either only a single large or two smaller NOR-bearing homologous sub-metacentric chromosomes was noticed in different member of the same population (Anjum *et al.*, 1997).

Although a frequent occurrence of two functionally active NOR-bearing chromosomes in common carp has been observed in a majority of cases but some skeptic opinions also exist for the existence of two additional NOR-bearing chromosomes which are thought to be inactive in this species of tetraploid origin (Sola *et al*., 1986). Such findings of Sola *et al*. (1986) are based upon the hypothesis that diploidization of gene expression in *Cyprinus carpio* L. could be a consequence of a reduction in the rate of rRNA transcription and therefore only one pair of NORs is active/visible in common carp. As two non-homologous chromosomes belonging apparently to two different pairs have been found to possess active NORs in the present study, it could be postulated that only one partner from each pair is active and visible while the other additional pair of NORs which may also exist, is functionally inactive and therefore invisible due to reduction in the clusters of rRNA.

In the present investigations, sequential staining of the metaphase spreads with chromomycin A3 confirmed the same number and localization of NOR-bearing chromosomes as were detected through Silver-NOR staining. Thus, an ample amount of evidence has become available to support Sola *et al*. (1986)'s findings that only one pair of functionally active NOR-bearing chromosomes is visible in the common carp.

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