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KARYOTYPE ANALYSIS OF THE FISH SPECIES Stizostedion volgensis (Percidae, Pisces) CAUGHT AT DIFFERENT LOCALITIES ON THE DANUBE

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Cytogenetic investigations of the fish species Stizostedion volgensis (Percidae, Pisces), were performed, using specimens collected from four different localities on the Danube (near Beška, Zemun, Višnjica and Grocka) during three consecutive years (1986, 1987 and 1988). Karyotype analysis showed that the diploid chromosome number was 2n=48. The karyotype consisted of 8 pairs of submetacentric (SM), 6 pairs of subacrocentric (SA) and 10 pairs of acrocentric (A) chromosomes.

The frequency of frequency of structural chromosome changes, exhibited as breaks and gaps was analysed. The highest frequency of these changes occurred in the fishes caught at the localities downstream of Belgrade and the mouth of the river (Sava), near Višnjica and Grocka.

On the basis of previous criteria and the proposed "critical level (3,0 - 3,5%) the values for break and gap-type changes that were above these, point to the presence of genotoxic agents in the river water.

The results obtained indicat the probable existence of a genotoxic risk, as these localities appear to be periodically or permanently contaminated with genotoxic agents,

Key words: Karyotype, Stizostedion volgensis, genotoxicity, chromosomal aberrations

INTRODUCTION

The data on the karyotype of pikeperch in the available literature are somewhat variable (Table 1). The diploid chromosome number in these fish species is definitely 2n=48 (Nygren et al., 1968; Bozhko et al., 1978; Živković et al., 1987; Fišter, 1992), but in relation to chromosome morphology, different data have appeared. Thus, Bozhko et al. (1978), reported that the caryotype of (Lucioperca lucioperca, L. (Stizostedion lucioperca in accordance with new nomenclature, consist of 4 M, 22 SM, 14 SA and 6 A; but Živković et al. (1987) suggests that the karyotype in Stizostedion volgensis which is the eastern variant of the species consists of: 16 M and SM, 12 SA and 20 A. Some data from the

earlier literature connot be considered, probably due to bad chromosome preparation techniques at that time and are only of historical importance, like the finding that the diploid chromosome number is 2n=24 (Svardson et al., 1939) and 2n=46 (Lieder, 1963)

Table 1. Karyotype data of the fish species Stizostedion volgensis and Stizostedion lucioperca (Lucioperca lucioperca L.)

Authors	Species	2n	Karyotype	NF
Svaedson et al. (1939)	Lucioperca lucioperca	24		
Lieder (1963)	Lucioperca lucioperca	46		
Nygren et al. (1968)	Lucioperca lucioperca	48	48 A	48
Bozhko et al. (1978)	Lucioperca lucioperca	48	4 M + 22 SM + 14 SA + 6 A	86
Živković et al. (1987)	Stizostedion volgensis	48	16 M, SM + 12 SA + 20 A	76
Fišter	Stizostedion volgensis	48	16 SM + 12 SA + 20 A	76

Using karyotype analysis, the presence of various structural changes might be detected, but most frequently, chromosome breaks and gaps occur. As well as some other authors Brogger (1982), suggested that increasing frequencies of breaks and gaps, were a useful parameter, an indicator of genotoxic contamination.

Chemical and other aggressive genotoxic agents influence hereditary structures and chromosome changas occurred as a result of their activity, e. g.: in tissue cultures, in experimental animals (Soldatović et al., 1980; Zimonjić et al., 1980; Zimonjić, 1982; Fišter et al., 1986; Fišter et al., 1987; Zimonjić et al., 1990), in fish from contaminated water (Perin et al., 1978; Alink et al., 1980; Fišter, 1992; Fišter et al., 1994), as well as in drinking water (Williamson, 1992; Van Hoof, 1982; Meiner, 1988; Onodera et al., 1993; Djelić et al., 1995). The reason for increasing chromosome changes might be also "anonimous agents", as for example inections viruses (Soldatović et al., 1981) and different sources of radiation (Bender et al., 1974; Countryman and Heddle, 1976; Ajdačić et al., 1986). However, the reason for increasing frequencis of breaks and gap-type changes might be found inside organisms themselves and they are of a genetic nature, the chromosome constitution itself. The age of the organisms and strain of the laboratory animals (Lilp et al., 1981) should also be taken into account.

By analysis of the frequency of breaks and gap-type chromosome changes in untreated laboratory animals (in the controls) and in the untreated tissue cultures (controls) as well as in fish from free waters (far from industrial plants, human habitats and other sources of related contamination), Fišter (1992) proposed criteria for a assessment of the probability of genetic risk on the basic of the proposed "critical level". Increasing values - in the area above the critical zone, indicate the presence of genotoxic agents in the natural environment of fishes.

MATERIAL AND METHODS

Specimens of the fish species Stizostedion volgensis were caught at four different localities of the Danube (near Beška, Zemun, Višnjica and Grocka)

during three consecutive years (1986, 1987 and 1988) and were cytogenetically analysed.

Abouth thirty mitotic figures of metaphase chromosomes were examined in every individual fish. Metaphase chromosomes were obtained by a preparation of kidney tissue, according to the method of Fontana et al. (1970) which was slightly modified.

RESULTS

Cytogenetic karyotype analysis of the fish species Stizostedion volgensis showed that the diploid chromosome number was 2n=48 (Figure 1).The karyotype of this fish species consisted of: 8 pairs of submetacentric (SM), i. e. 16 submetacentric chromosomes: 6 pairs of subacrocenric (SA), i. e. 12 subacrocentric chromosomes and 10 pairs of acrocentric (A), i. e. 20 acrocentric chromosomes; NF=76 (Figure 1).

The frequency of structural chromosome aberrations of the break and gap-type are presented in Table 2 and Figure 2.

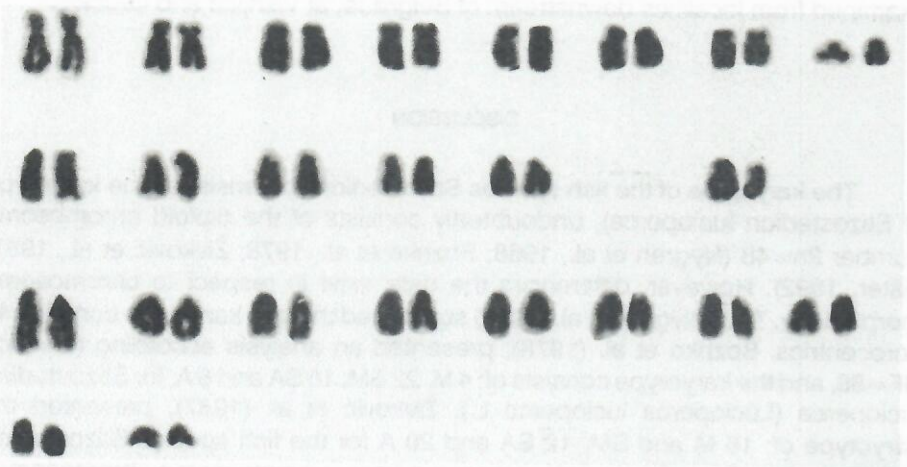

Figure 1. Karyotype of the fish species Stizostedion volgensis (Percidae)

In the fisches which were caught in the Danube at Beška in 1986, 290 mitotic figures of the metaphase chromosome were examined and breaks and gaps detected in 1,03%. In 1987, in the fishes from the same locality, 267 mitotic figures of chromosomes were analysed and 0,74% changes of the same type were recorded. Cytogenetic examination of 210 metaphase chromosome figures in individual fishes, which were caught at Beška in 1988, revealed 0,95% breaks and gaps.

The specimens of pikeperch caught in the Danube, at Zemun in 1986, were cytogenetically analysed; 180 mitotic figures of chromosome were examined and

2,22% breaks and gaps were detected. In the fishes from the same locality, 234 metaphase were analysed in 1987, and 1,70% breaks and gaps were recorded. A total of 297 mitotic figures of chromosomes were examined in the fishes which were caught in the Danube at Zemun in 1988 and 1,01% exhibited the same type of structural aberration.

In the specimens of pikeperch which were caught in the Danube at Višnjica in 1986, karyotype analyses revealed breaks and gaps in 3,76%. A total of 270 mitotic chromosome figures were analysd in individual pikeperch caught a year later (1987), and 3,33% breaks and gaps were observed. In the fishes caught at the same locality 1988, 297 mitotic figures were examined and 3,36% structural changes of the same type were detected. In the individuals of pikeperch which were caught in the Danube at Grocka in 1986, cytogenetic analysis of 300 mitotic metaphase chromosome figures revealed 3,66% changes of the gap type and chromosome breaks. At the same locality, the next year, 240 mitotic figures were examined and 2,91% breaks and gaps were detected. In fishes of the same species were caught and 292 mitotic figures of chromosomes were examined revealing breaks and gaps in 3,42%.

The results are presented in Figure 2 according to the criteria of Fišter, (1992). The highest frequency for breaks and gaps was obtained in fishes examined from localities downstream of Belgrade, at Višnjica and Grocka.

DISCUSSION

The karyotype of the fish species Stizostedion volgensis (as the karyotype of Stizostedion lucioperca), undoubtedly consists of the diploid chromosome number 2n=48 (Nygren et al., 1968; Bozhko et al., 1978; Živković et al., 1987; Fišter, 1992). However, differences the data exist in respect to chromosome morphology. Thus Nygren et al. (1968) suggested that the karyotype contains 48 acrocentrics. Bozhko et al. (1978), presented an analysis according to which NF=86, and the karyotype consists of: 4 M, 22 SM, 16 SA and 6 A, for Stizostedion lucioperca (Lucioperca lucioperca L.). Živković et al. (1987), presented the karyotype of: 16 M and SM, 12 SA and 20 A for the fish species Stizostedion volgensis, but diversification of metacentric from submetacentric chromosomes was not made. In our opinion and on the basis of data obtained, there is no metacentric chromosome in the karyotype of the fish species Stizostedion volgensis, and the karyotype contains a relatively large number of acrocentrics, i. e.: 16 SM, 12 SA and 20 A; NF=76. It seems very possible that the karyotype of Stizostedion volgensis differs from the karyotype of Stizostedion lucioperca, but this needs further analysis.

Cytogenetic analysis showed that the frequencies of chromosome breaks and gaps showed statistically significant differences in relation to localities from which the fishes were collected during the three year investigation (Table 2, Figure 2).

The mean values obtained for the frequency of structural aberrations at Beška, were much lower than the proposed critical zone (Fišter, 1992) and there were about 1% (for 1986, 1,03%; for 1987, 0,74% and for 1988, 0,95%). Somewhat higher values were recorded in the fishes caught in the Danube at Zemun (for 1986, 2,22%; for 1987, 1,70% and for 1988, 1,01%), but none of them exceeded the range of the critical zone, i. e. the vaue of 3,5%. However, the mean values for gaps and breaks for the two localities (Beška and Zemun), were much lower than the proposed level for the critical zone.

Table 2. Frequency of chromosomal breaks and gaps in Stizostedion volgensis from some localities of the Danube

Locality	Year	Mitoses examined	Breaks and gaps	Breaks and gaps %
BEŠKA	1986.	290	3	1,03
	1987.	267	2	0,74
	1988.	210	2	0,95
ZEMUN	1986.	180	4	2,22
	1987.	234	4	1,70
	1988.	297	3	1,01
VIŠNJICA	1986.	292	11	3,76
	1987.	270	9	3,33
	1988.	297	10	3,36
GROCKA	1986.	300	11	3,66
	1987.	240	7	2,91
	1988.	292	10	3,42

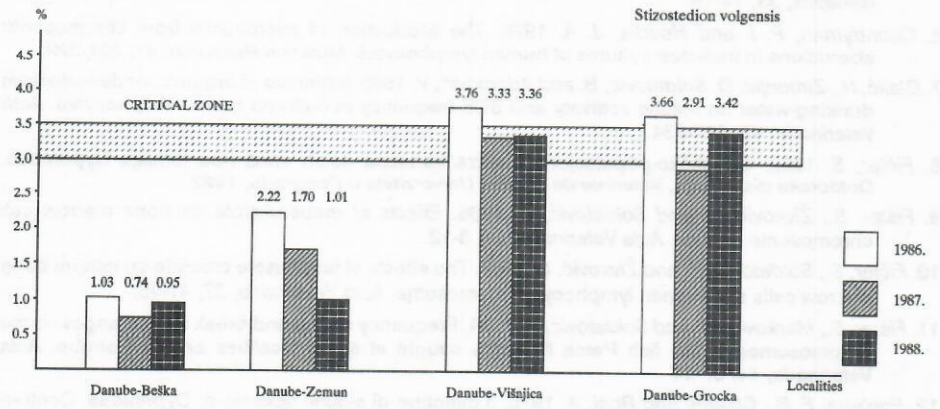

Figure 2. Frequencies of chromosomal breaks and gaps in Stizostedion volgensis specimens caught at four different localities of the Danube during three consecutive years

Statistically very significant increased mean values for these chromosome changes (compared to the northern localities), were obtained for the fishes from the Danube, caught at the locality of Višnjica. Thus, in 1986, the mean value was

3,76% and above the level of the proposed critical zone, whereas in 1987 and 1988, it was somewhat lower and in the critical area (3,33% and 3,36%).

In the fishes collected from the Danube, at Grocka, the mean frequency values for the breaks and gaps were within the range of the critical zone (for 1988, 3, 42%) or above them (for 1976, 3,66%). Statistically very significant differences were obtained when this locality was compared with the localities north of Belgrade and the river Sava, at Beška and Zemun. Similar results were obtained for the fish species Perca fluviatilis (Fišter et al., 1994).

On the basis of the data obtained, the existence of a genotoxic risk might be concluded, as some localities of the Danube (by Višnjica and Grocka), were permanently or periodically contaminated with genotoxic agents.

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ANALIZA KARIOTIPA RIBA VRSTE STIZOSTEDION VOLGENSIS (PERCIDAE, PISCES) UHVAĆENIH NA RAZLIČITIM LOKALITETIMA DUNAVA

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SADRŽAJ

Izvršena su citogenetička istraživanja vrste Stizostedion volgensis (Percidae, Pisces), na četiri lokaliteta Dunava (kod Beške, Zemuna, Višnjice i Grocke) u tri uzastopne godine (1986, 1987 i 1988). Analiza kariotipa je pokazala da se on sastoji od 2n=48 hromozoma i to: 8 pari submetacentrika (SM), 6 pari subakrocentrika (SA) i 10 pari akrocentrika (A).

Analizirana je učestalost promena tipa hromozomskih prekida i gapova i ustanovljeno je da se srednje vrednosti ovih promena bile najviše kod riba uhvaćenih na lokalitetima Dunava nizvodno od ušća reke Save i Beograda, kod

Višnjice . Prema ranije utvrđenom kriterijumu, na osnovu pretpostavljene "kritične zone" (3,0 - 3,5% ovih strukturnih promena), vrednosti iznad ove zone ukazuju na prisustvo genotoksičnih agenasa u rečnoj vodi.

Dobijeni rezultati ukazuju na moguće postojanje izvesnog genetičkog rizika, odnosno - na moguće povremeno ili stalno prisustvo genotoksičnih agenasa na ispitanim lokalitetima nizvodno od Beograda.

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SADRŽAJ

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