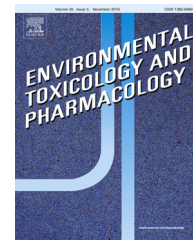


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# Biological monitoring and B chromosome frequency in Bagre (*Rhamdia quelen*) in southeast Brazil

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## ABSTRACT

The genus *Rhamdia* presents B chromosomes which appear to be present in most species of the genus and thus represent an important characteristic in the evolutionary process. Furthermore, variations in environmental conditions can induce the presence of B chromosomes generated by alterations in the cell cycle, due to the interference from pollutants. The present study aimed to evaluate the cytogenetic aspects of individuals of a population of *Rhamdia quelen* collected in three areas with differing standards of water quality in the River Uberabinha, a region of the County of Uberlândia, Minas Gerais, Brazil. The Piscine Micronucleus Test results indicate significant genotoxic and cytotoxic potential at the sampling Sites. The chromosome count yielded the modal number  $2n = 58$  with variance between zero and seven B chromosomes. The highest frequency of B chromosomes and the presence of karyotypes with seven supernumerary chromosomes occurred at Site 3, referring, thus, to the location of the highest genotoxic potential. There was a positive correlation between the presence of B chromosomes and the reduction in environmental quality. Therefore, the process of bioaccumulation of heavy metals in aquatic environments may be crucial to determine the presence of B chromosomes.

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## 1. Introduction

Biological monitoring represents the regular and systematic use of live organisms, known as biological indicators, to

evaluate changes in the environment or water quality (de Zwart, 1995; Sandrini et al., 2009). Chemical substances that do not belong to the natural environment interfere in the development of local organisms and subsequent survival rates (Van Der Oost et al., 2003; Frenzilli et al., 2009). The deleterious

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effects on the individuals of the indigenous population cannot easily be perceived since their damage tends to become apparent only over long periods of time. The actions of these pollutants, however, can be examined directly in various organisms, normally by analyzing the cells from organ tissues and using biomarkers. But genotoxic effects of these substances may not become apparent for various generations (Padrangi et al., 1995).

Schmid (1975) and Heddle (1993) characterized a cytogenetic attempt to represent the quantification of the Howell–Joly corpuscles, cytoplasmic structures known as micronuclei, originating from damage to DNA. The micronuclei are extra nuclear corpuscles formed during the process of mitosis. They are the result of chromosome fragments without centromeres or entire chromosomes that were not included in subsequent cell division although they had originated in the process of cell division. These micronuclei thus indicate clastogenic or aneugenic events (Albertini et al., 2000). Morphological nuclear alterations are recurrent problems in the segregation of chromosomes that may become tangled or joined together. These alterations may also be due to genetic amplification in the bridging-breaking-fusion cycle and the posterior elimination of DNA with the amplification of these chromosomes (Ergene et al., 2007).

Intra-specific type karyotype assessment permits the determination of evolutionary interferences and guides assumptions regarding the origins of the morphological and karyotypic structural variations for individuals of the same species situated in different regions. To carry out the technique for mitotic chromosome observation it is necessary to stimulate the cell division process and inhibit mitotic spindle formation during the metaphase phase to later obtain biological material to carry out the microscopic procedure of chromosome counting and pairing (Bertollo et al., 1978).

*Rhamdia quelen* is popularly known as “Jundiá”. This is an osseous fish of the order *Siluriforme* and family *Heptapteridae*. The species examined presents B chromosomes known as supernumerarys. These chromosomes have no defined function and their numbers in the *R. quelen* species can vary from one to seven. The presence of one or two of these elements, however, is most frequent. The supernumerarys can vary between micro and medium sizes (Swarça et al., 2000). The presence of supernumerary chromosomes in any species may be related to variations in environmental conditions because pollutants, especially heavy metals, can alter the cell cycle generating chromosome recombination (Eliseeva et al., 1990; Green, 1990).

The population examined in the present study was sampled in areas of the River Uberabinha, which drains a basin of more than 2000 square kilometers. Flowing through three counties, Uberaba, Uberlândia and Tupaciguara, the river draws water from areas of soybean and corn production as well as urban zones. The river includes specialized areas affected by urban development, an industrial park with residue dumping and the effluent from sewage treatment in the city of Uberlândia (Brites and Rantin, 2004). This being the case, waterborne pollutants present in the areas sampled of the Uberabinha may induce biological responses in the bony parts examined that could originate supernumerary chromosomes, as compared with samples taken from non-polluted

areas (reference site). The present study aimed to evaluate the cytogenetic aspects of individuals of a population of *R. quelen* collected in three reaches with differing standards of water quality in the River Uberabinha, a region of the County of Uberlândia, Minas Gerais, Brazil. The species was evaluated using the Piscine Micronucleus Test.

## 2. Material and methods

### 2.1. Sampling sites

The biological material was collected from three sampling sites (Fig. 1) that present adverse characteristics (taking into account the potential genotoxic activity) of the Uberabinha in the region of Uberlândia, Minas Gerais, Brazil. The Sites were located with the objective of sampling three distinct sites of the Uberabinha.

Site 1—Reference site located approximately 26 m above the bridge of the BR-050 highway as it exits the city of Uberaba, Minas Gerais, Brazil. This site drains a reserve area of native growth adjacent to the riverbed. Geographical coordinates of the location: 18°53′25.08″S and 48°13′32.41″W.

Site 2—The second sampling site is located below or downstream from the reference site. Geographical coordinates of the location: 18°54′11.21″S and 48°18′17.10″W.

Site 3—The third sampling site is located on the Capim Branco Farm, property of the Federal University of Uberlândia. Geographical coordinates of the location: 18°53′18.03″S and 48°20′11.16″W.

### 2.2. Biological material

A total of six samplings were made during the dry season (low water) and the wet season (high water), from February to September, 2012. Fish worms were used as bait. The fish were put in styrofoam containers with water from the location, and aerators were used. Sixty fish were identified by the Ichthyological Laboratory in Ribeirão Preto–São Paulo, Brazil, as being *R. quelen*. Thirty fish were collected during the dry season, and thirty during the wet season. These fish, of both sexes, were transported alive, during the night, to the Cytogenetic Laboratory of the Federal University of Uberlândia. The morphometric parameters of the fish were taken including weight and size (Fish were only considered that presented the same weight standard, approximately 800 g and 25 cm average length to ensure an isogenic standard in the samples).

### 2.3. Sample monitoring

Following standard procedures (APHA, 1998), the fish were kept in glass aquariums (24″ × 20″ × 20″) containing 60 L of unchlorinated water. A period of one day was observed for acclimatization before testing. The water of the aquariums was maintained saturated in oxygen by the aerators and the temperature was controlled (25 ± 2 °C).

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