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Evaluation of toxicity, mutagenicity and carcinogenicity of samples from domestic and industrial sewage

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HIGHLIGHTS

- Toxicity, mutagenicity and carcinogenicity potential were studied in two water bodies.
- The Mumbuca stream and Perdizes river are target of different anthropic contaminants.
- The economic crisis of local ceramics has resulted in lower discharge of heavy metals.
- *D. melanogaster* is a sensitive indicator of environmental impacts in water bodies.

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ABSTRACT

Physico-chemical and toxicological analyses are of fundamental importance to determine water quality. The objectives of the present study were to evaluate the toxicity, mutagenicity and carcinogenicity of samples from the Mumbuca Stream and the Perdizes River, through both SMART and the wts test, respectively, in somatic cells of *Drosophila melanogaster* and to quantify the amount of heavy metals and other pollutants, which are indicative of environmental quality. Water samples were collected (M1, M2, P1, P2 and MP) and submitted to physico-chemical analysis, calculating the water quality index for each sampling site. In order to evaluate the toxicity, mutagenicity and carcinogenicity of the samples, third instar larvae descended from the crossing between virgin female *wts*/TM3, sb^1 and *mwh*/*mwh males* (wts test) and ST and HB (SMART) crosses were treated with samples from P1, P2, M1, M2 and MP sites. The physico-chemical analysis and the biological assay allowed us to conclude that undetected values for heavy metals and the low frequency of mutant spots (SMART) and epithelial tumor (wts) in treated flies from the Mumbuca Stream and Perdizes River may be due to the reduction of ceramic activities in the municipality. The physico-chemical analyzes identified altered the environmental quality parameters, which directly influenced the survival of *D. melanogaster* treated with samples of M2 and MP, which according to the WQI were classified as regular and poor environmental quality, respectively. The altered parameters may be due to clandestine domestic sewage sent downstream of the effluent.

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1. Introduction

The discharges of domestic, agricultural and industrial sewage linked to the deforestation of riparian forests are among the greatest anthropogenic impacts within the environment (Liu et al.,

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2017; Freitag et al., 2017; Salehi et al., 2017; Meena et al., 2017). As a consequence, biodiversity loss may occur, resulting from both indirect and direct effects, such as the ones caused by toxic compounds (Goulart and Callisto, 2003; Chaves et al., 2016; Zhang et al., 2017).

Because of this, the Brazilian National Environmental Council (CONAMA), via resolution 357, dated from 2005, stated that pollutant release into the environment would be prohibited. After the resolution was released, water bodies were then put under a normative, having its quality assessment established to monitor aquatic environments, through numerous tests, including physico-chemical analysis.

Both physico-chemical and toxicological tests are essential in determining water quality, pollutant risks and the effects on aquatic environments, which are mainly measured by biological assays using model organisms (Salvo et al., 2017; Guo et al., 2016; Dvorak et al., 2015; Goswami et al., 2014).

Drosophila melanogaster (Meigen, 1830) has gained prominence as a research model (Nepomuceno, 2015), because it is sensitive to several environmental pollutants (Singh and Chowdhuri, 2018; Chauhan et al., 2017; Zhou et al., 2017). The homology among genes with recognition to human diseases has contributed to the usage of *D. melanogaster* as a model in studies based on toxicological genetics (Kim et al., 2011).

Both the SMART (Graf et al., 1984; Graf and Van Schaik, 1992) and the Epithelial Tumor Detection (wts test) tests have been greatly performed on *D. melanogaster* (Nepomuceno, 2015). SMART is based on mutant identification resulted from heterozygosis loss of genetic markers (*mwh* and *flr*^{3}) in imaginal disk cells, as a consequence of xenobiotics with either mutagenic or promutagenic activity. SMART test allows the quantification of events resulting from mitotic recombination (Spanó et al., 2001).

SMART is usually a test for mutagenesis analysis (Oliveira et al., 2017; Machado et al., 2016; de Rezende et al., 2011), while the wts test is usually related to carcinogenesis studies (Morais et al., 2016b, 2016c; Furtado and Nepomuceno, 2012; Orsolin et al., 2012; Costa et al., 2011; Orsolin and Nepomuceno, 2009). Although both SMART and the wts test have already been used in environmental quality analyzes, there is no research taking both into account in the same study.

Monte Carmelo municipality, located in the state of Minas Gerais, Brazil is known for its ceramic-dependent economy, which was associated with a continuous process of pollution by the disposal of both domestic and industrial litter (Morais et al., 2016a; Campos et al., 2015; Campos et al., 2015). Heavy metals were also found, with limits higher than the one allowed by CONAMA (2005), being associated with chromosomal changes in native species (Morais et al., 2016a; Campos Júnior et al., 2015).

The objective of the present study was to evaluate the toxicity, mutagenicity and carcinogenicity of two water bodies, the Mumbuca Stream and the Perdizes River, located in the Monte Carmelo municipality, Minas Gerais, Brazil, with a continuous process of pollution by the disposal of both domestic and industrial wastewater (Morais et al., 2016a; Campos et al., 2015; Campos Júnior et al., 2015) through both SMART and the wts test using *D. melanogaster*. The physico-chemical conditions and the water quality of the streams were also determined through a water quality index, in order to identify the main polluting source in the municipality.

These water resources were selected for environmental evaluation based on the evidences of discharges of domestic and industrial effluents by anthropic activities on the banks of these bodies of water.

2. Material and methods

2.1. Sampling sites

Sampling sites (Fig. 1) were located in the Monte Carmelo municipality (18º44′5” S and 47º29′47” W), from the state of Minas Gerais, Brazil. Monte Carmelo has about 45 thousand inhabitants within an area of 1343035 km^{2} (IBGE, 2010). A high amount of the population is dependent somehow on the streams located nearby the city.

Five sites were chosen based on the presence or absence of anthropic activities in every sampling unit. Site 1 (M1 - 18°44′19.6″ S and 47°29′47.4″ W) is located in the Mumbuca stream and is not influenced by anthropogenic degradation process, being used by the Municipal Department of Water and Sewage (DMAE), in both collection and distribution of water for local population use.

Site 2 (M2 - 18°42′31.9″ S and 47°29′37.8″ W) is also located in the Mumbuca stream and is targeted for disposal of domestic and industrial pollutants.

Site 3 (MP - 18°41′48.1″ S and 47°26′57.3″ W) is located in meeting point of both the Mumbuca stream and Perdizes river, also receiving inputs of pollutants.

Site 4 (P1 - 18°39′48.7″ S and 47°29′14.3″ W) and site 5 (P2 - 18°40′30.4″ S and 47°28′7.10″ W) are both located in Perdizes river. Previous data have howed poor environmental quality at the chosen sites (Campos et al., 2015).

2.2. Physicohemical analysis of water

A total of 5 samples of water were collected in each site of sampling. Physicochemical parameters of suspended solids and biochemical oxygen demand (BOD) of water were analyzed according to the procedures reported by Boyd and Tucker (1992).

The evaluation of other physical and chemical parameters occurred under the standard methods guidelines from Examination of Water and Wastewater (APHA-AWWA-WPCF, 1998).

Analyses of toxic metals were made using a Varian Spectra AA-220FS atomic absorption spectrometry. Reference solutions of toxic metals were arranged in order of analysis, prepared with 0.9 mol/L of HNO and CFA-C 10% v/v (pH 8.0) (limits of detection/LOD of 0.001 mg/L).

All analyzes were carried out at the Chemistry Laboratory of the Federal University of Uberlândia, MG, Brazil.

The results of the physicochemical analyses were compared with the tolerance limits for each parameter according to Guideline 357/2005 of CONAMA (CONAMA, 2005). These permissible limits are compatible with the standards set by the World Health Organization (WHO, 2011).

The analyze]s were done during the dry season (April to September). The rainy season was not analyzed due to the low rainfall during the year (2016).

2.3. Water quality index (WQI)

Water quality index determination was carried out according to the parameters of the Instituto Mineiro de Gestão das Águas (IGAM, 2013).

Data analyses were performed using the IQADATA Software, available from the University of Santa Cruz do Sul, Rio Grande do Sul state, Brazil.

The water quality index is determined by the product of quality established for each parameter such as: dissolved oxygen, fecal coliform, pH, biochemical oxygen demand, nitrate, phosphate, water temperature, turbidity and total solids (Yogendra and

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