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DIEGO LACERDA DE SOUZA

INFLUÊNCIA DO USO DO SOLO NAS CONCENTRAÇÕES DE METAIS EM TRÊS SUB-BACIAS DO SUDESTE DO BRASIL

CAMPOS DOS GOYTACAZES

FEVEREIRO 2018

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Dissertação apresentada no Centro de Biociências e Biotecnologia da Universidade Estadual do Norte Fluminense Darcy Ribeiro como parte das exigências para a obtenção do título de Mestre em Ecologia e Recursos Naturais.

Orientador: Carlos Eduardo de Rezende

Co-Orientadora: Cristiane dos Santos Vergilio

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Ama-se mais o que se conquista com esforço.

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RESUMO

As diferenças no uso do solo causadas pelas atividades antrópicas e naturais afetam a dinâmica da transferência de materiais para os ambientes aquáticos. Em áreas agrícolas, a conversão da vegetação nativa e o uso de pesticidas e fertilizantes podem causar contaminação desses ambientes. Assim, o presente estudo teve como objetivo avaliar a influência das diferenças no uso do solo nas concentrações de metais (Al, Ba, Cr, Cu, Fe, Mn, Pb, Ti e Zn) em três sub-bacias (BND – Rio Braço Norte Direito; BNE – Rio Braço Norte Esquerdo; IT – Rio Itapemirim) do rio Itapemirim, utilizando diferentes matrizes ambientais (solo, água, sedimento e ictiofauna). Índices e fatores geoquímicos como o fator de contaminação (FC), índice de contaminação (IC), fator de enriquecimento (FE) e análise dos componentes principais (PCA) foram utilizados para avaliar as influências antrópicas no sedimento dos rios. A sub-bacia do BND apresenta predominância de grandes fragmentos de floresta nativa, enquanto que no BNE estão as maiores áreas destinadas à cafeicultura e a outras a atividades agrícolas, e a sub-bacia do IT apresenta principalmente áreas de pastagens. As diferenças no uso do solo refletiram em diferentes concentrações dos metais nas matrizes avaliadas, onde em geral BNE>BND>IT. Da mesma maneira, os valores do FC, IC e FE apontam para maior contaminação do BNE, sendo a ordem desses índices em geral BNE>IT>BND. A análise dos componentes principais mostrou uma menor associação de Cr, Cu, Ni e Zn com os principais suportes geoquímicos (Al, Fe, Mn, Ti e C orgânico), indicando uma possível influência antropogênica nas suas concentrações. Os resultados do presente estudo indicam que a mudança do uso do solo pra áreas agrícolas, o possível uso de pesticidas e fertilizantes e o aumento do escoamento superficial estão atuando como fontes de metais para a água, sedimento e biota, em especial no BNE. Além disso, a insuficiência do tratamento de esgoto nos municípios do Caparaó Capixaba também pode estar atuando como fontes de metais para as sub-bacias da região.

Palavras-chave: Sedimento, índices geoquímicos, cafeicultura, contaminação.

ABSTRACT

Differences in land use caused by anthropogenic and natural activities affect the dynamics of the transfer of materials to aquatic environments. In agricultural areas, the conversion of native vegetation and the use of pesticides and fertilizers may cause contamination of these environments. This study aimed evaluating the influence of land use differences on the concentrations of metals (Al, Ba, Cr, Cu, Fe, Mn, Pb, Ti and Zn) in three sub-basins (BND – Braço Norte Direito River, BNE – Braço Norte Esquerdo River; IT – Itapemirim River) of the Itapemirim Basin, whose land use is predominantly focused on agricultural activities, using different environmental matrices (soil, water, sediment and ichthyofauna). Indices and geochemical factors such as contamination factor (CF), pollution load index (PLI), enrichment factor (EF) and principal component analysis (PCA) were used to evaluate the anthropogenic influences on river sediment. The BND sub-basin has a predominance of large fragments of native forest, while in the BNE there are the largest areas for coffee cultivation and others for agricultural activities, and the IT sub-basin presents mainly pasture areas. The differences in land use reflected in different concentrations of metals in the matrixes evaluated, where in general BNE > BND > IT. Likewise, CF, PLI and EF values point to a higher BNE contamination, the order of these indices being BNE > IT > BND in general. The PCA showed a lower association of Cr, Cu, Ni and Zn with the main geochemical supports (Al, Fe, Mn, Ti and organic C), indicating a possible anthropogenic influence in their concentrations. The highest concentrations of metals in the BNE are related to the larger areas destined to agriculture, mainly coffee cultivation. The results of the present study indicate that the change of the land use to agricultural areas, the possible use of pesticides and fertilizers and the increase of surface runoff are acting as sources of metals for water, sediment and biota, especially in the BNE. In addition, the insufficiency of sewage treatment in Caparaó Capixaba municipalities may also be acting as sources of metals for the sub-basins of the region.

Keywords: Sediment, geochemical indexes, coffee cultivation, contamination.

1. INTRODUÇÃO

Os solos desempenham papel importante nas condições geoquímicas dos ambientes aquáticos de água doce, uma vez que, partículas e material dissolvido nas suas formas orgânica e inorgânica podem atingir os ecossistemas aquáticos a partir do escoamento superficial. Nesse sentido, os diferentes usos do solo como na indústria, agricultura, mineração, urbanização, e mesmo a manutenção de áreas de vegetação nativa podem influenciar nessa dinâmica, uma vez que a conversão do uso do solo para suprir as demandas humanas remove a cobertura natural que leva a intensificação do escoamento superficial (BAI *et al.*, 2010; HOU *et al.*, 2017; JOSHI; BALASUBRAMANIAN, 2010; ZHANG *et al.*, 2014).

Dentre os contaminantes, os metais têm grande importância devido as diferentes funções em vias metabólicas para os vegetais e animais. Às atividades humanas, desde a revolução industrial, têm sido responsáveis por um aumento significativo destes elementos, gerando problemas de ordem sanitária, desequilíbrio ecológico e saúde humana (CRUTZEN, 2006; PRICE *et al.*, 2011).

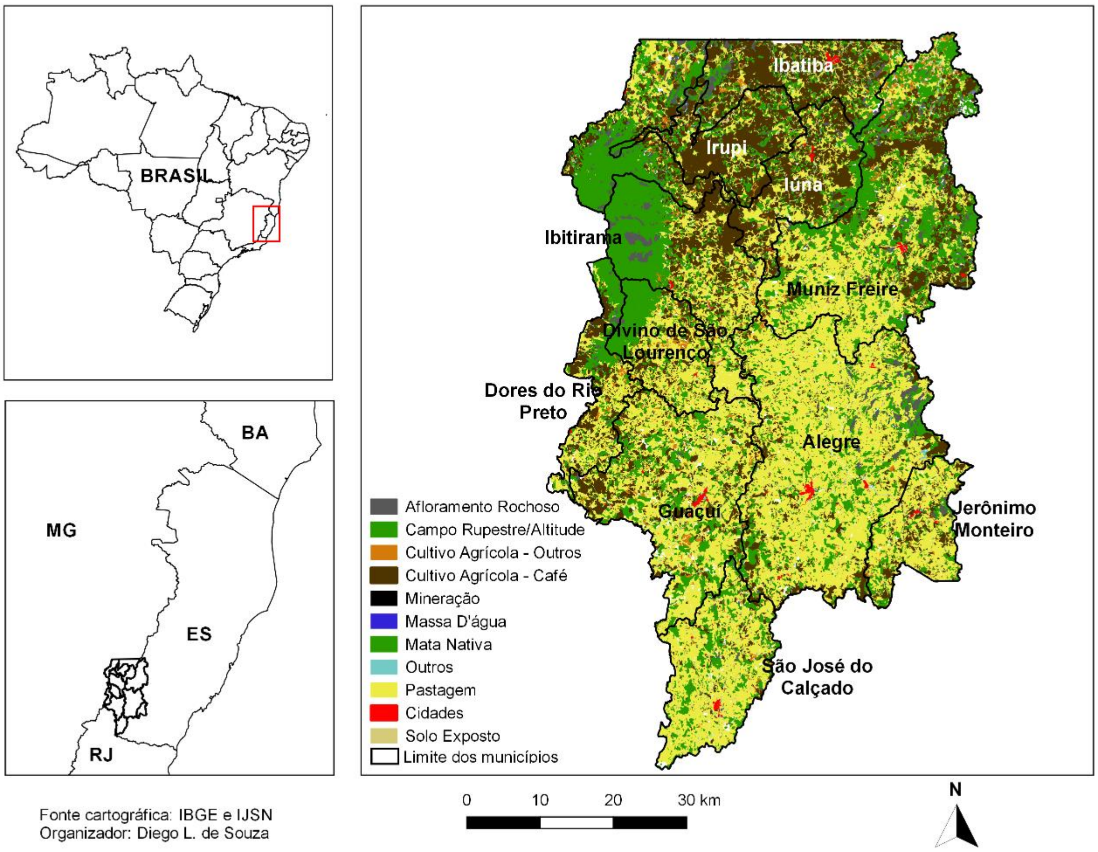
Nas áreas agrícolas, a conversão da vegetação original por áreas de lavouras, aumenta o revolvimento dos solos, o escoamento superficial, além da aplicação de pesticidas e fertilizantes que contém metais em sua composição, pode causar contaminação dos ambientes aquáticos por esses elementos. A falta de conhecimento dos agricultores na aplicação desses produtos acaba agravando o problema (BEDOR *et al.*, 2009). Além disso, em muitas situações a aplicação dos defensivos não é realizada da maneira correta, e em quantidades maiores do que a recomendada, provocando a contaminação das culturas e das áreas circundantes (ROCHA *et al.*, 2015).

O uso de pesticidas para impedir danos em colheitas é histórico, sendo estimada a entrada no mercado de aproximadamente 32 mil formulações desde o início do seu registro (MATTICE, 2010). Atualmente, os pesticidas ocupam o segundo lugar mundial dentre as substâncias químicas com extenso uso no meio ambiente, perdendo apenas para os fertilizantes (“Infographic: Pesticide Planet”, 2013). Os termos “pesticidas” e “agrotóxicos” abrangem um amplo grupo de compostos que são tipicamente categorizados com base na sua ação incluindo herbicidas, fungicidas, inseticidas, nematicidas, dentre outros. Os primeiros pesticidas consistiram de substâncias que continham grande quantidade de elementos tóxicos como o arsênio, cobre, chumbo, cádmio, mercúrio e que foram intensamente usados até o final da década de 40 (MATTICE, 2010). A sua aplicação levou a acumulação de metais no solo e no sedimento dos ecossistemas aquáticos, os quais poderiam remobilizar dependendo das condições climáticas e geoquímicas, levando a uma incorporação desses elementos na biota. Em função da alta toxicidade e persistência no ambiente foram introduzidos no mercado os pesticidas sintéticos orgânicos, inicialmente com a descoberta dos organoclorados, como o DDT, que teve seu uso substituído pelos atuais organofosforados, triazinas, ácidos fenoxiacéticos, carbamatos, triazóis, piretróides (MCKNIGHT *et al.*, 2015).

Assim como os pesticidas, os fertilizantes também apresentam metais em sua composição como cádmio, chumbo, arsênio, cobalto, ferro, manganês, zinco, cobre e níquel. A aplicação desses produtos leva ao aumento da concentração desses elementos nos solos e nas águas subterrâneas (ATAFAR *et al.*, 2010). A partir disso, estes elementos podem ser absorvidos pelas plantas e acumulados por animais, chegando até o homem através de ingestão direta ou transferidos ao longo da cadeia alimentar (TAYLOR; PERCIVAL, 2001). A aplicação de pesticidas e fertilizantes, além de contribuir com a entrada direta de metais no ambiente, pode torna-los mais biodisponíveis, uma vez que a sua utilização diminui o pH dos solos, favorecendo a maior mobilidade desses elementos (DEVRIES *et al.*, 2002).

Uma importante área agrícola no Brasil é a microrregião do Caparaó, localizado no sudoeste do Espírito Santo. A população da região é 181 mil habitantes, que vive em sua maioria na zona urbana (aproximadamente 60% da população) dos municípios (Alegre, Divino de São Lourenço, Dores do Rio Preto, Guaçuí, Ibatiba, Ibitirama, Irupi, Iúna, Jerônimo Monteiro, Muniz Freire e São José do Calçado) (ESPÍRITO SANTO, 2005; IBGE, 2017).

Na região, cerca de 63% do uso do solo é destinado para atividades agropecuárias, sendo 42% pastagens e 21% cultivos agrícolas. Predominantemente, a cafeicultura ocupa 19% da área, enquanto que a vegetação nativa cobre 20%. A mineração (0,3%) (principalmente extração de mármores, granitos e de areia), áreas urbanas (0,36%), afloramentos rochosos (3,56%) e solos expostos (0,78%) correspondem a 4,7% do uso do solo (Figura 1) (IJNS, 2006).



Fonte cartográfica: IBGE e IJSN
Organizador: Diego L. de Souza

Figura 1: Mapa do uso do solo da região do Caparaó no sudoeste do Espírito Santo.

A cafeicultura é também a principal atividade econômica da região, gerando 70% da renda. Mas outras atividades como a pecuária e o cultivo de hortaliças também são importantes economicamente. A implantação da cafeicultura, a partir da segunda metade do século XIX, deu início a um intenso processo de desmatamento na região. Isso se junta com práticas de uso degradante do solo, alteração da paisagem, aplicação abusiva de pesticidas e contaminação do solo e da água, que foram indicados desde 2002 como os principais problemas ambientais da região (ESPÍRITO SANTO, 2005).

O Caparaó está inserido em três bacias hidrográficas (do Rio Itapemirim, Rio Itabapoana e do Rio Doce), sendo que a bacia do Rio Itapemirim, que possui uma área total de 5.900 km^2, abriga a maior parte dos municípios da região, ocupando cerca de 61% da área do Caparaó (IJNS, 2006). O Rio Itapemirim (IT) se forma no município de Alegre, a partir da união dos seus dois principais afluentes, o rio Braço Norte Esquerdo (BNE) (com uma sub-bacia de 1.432 km^2) e o Braço Norte Direito (BND) (com uma sub-bacia de 510 m^2). Após a sua formação, o rio Itapemirim recebe águas de outros afluentes na região, formando uma sub-bacia parcial independente das demais, com uma área de 442 km^2 (IJSN, 2006).

As diferentes sub-bacias do rio Itapemirim possuem também diferentes características de uso do solo. As áreas de mata cobrem 30% do uso do solo na sub-bacia do BND, estando nessa região localizadas as principais áreas de preservação da região, como parte do Parque Nacional do Caparaó e o Parque Estadual da Cachoeira da Fumaça. No BNE e no IT, por sua vez, as áreas de vegetação nativa cobrem uma área menor, estando distribuídas em sua maioria em pequenos fragmentos. Na sub-bacia do BNE, estão as maiores áreas destinadas à agricultura, assim como as maiores áreas de solo exposto, dentre outros usos. Já a sub-bacia parcial do rio Itapemirim destaca-se pelas áreas de pastagem (aproximadamente 65%) (IJSN, 2006).

O Estado do Espírito Santo é o segundo maior produtor de café do Brasil a região do Caparaó chega a contribuir com até 19% da produção estadual (IBGE, 2017; ESPÍRITO SANTO, 2005). O fato de boa parte da área da região ser utilizada para atividades agrícolas sugere o uso difuso de pesticidas e fertilizantes. No ano de 2012, o Estado era o sétimo maior consumidor de pesticidas por área plantada do País, e no ano de 2014 quando foram levantados os últimos dados ocupava a 13ª posição. Quanto a aplicação de fertilizantes o estado ocupa a 2ª posição nacional com um total de aproximadamente 190 kg/ha (IBGE, 2017).

Tendo em vista o fato de que os metais provenientes da aplicação dos defensivos e fertilizantes poderem ser incorporados pelas culturas, e consequentemente ser transferidos para os consumidores (LEBLANC *et al.*, 2000). Torna-se necessário estudos que avaliem a presença desses elementos na região do Caparaó Capixaba, bem como a sua relação com as atividades agrícolas. Nesse sentido, o monitoramento através da avaliação de matrizes ambientais como peixes, sedimento, água e solo são fundamentais na avaliação integrada da contaminação ambiental. Com isso, é necessário compreender melhor as fontes, a dinâmica e os efeitos que os metais podem causar no ambiente através do estudo de suas concentrações e da influência do uso do solo nesse processo.

2. OBJETIVOS

2.1. Objetivo geral

Este trabalho teve como objetivo realizar uma análise integrada das concentrações de metais na região do Caparaó, avaliando diferentes matrizes ambientais (solo, água, sedimento e ictiofauna) na região da Bacia do Rio Itapemirim (principal Bacia Hidrográfica da região do Caparaó) em dois períodos do ano (seco e chuvoso) visando traçar a influência do uso do solo na concentração de metais na interface ecossistema terrestre e ambiente aquático.

2.2. Objetivos específicos

I. Determinar os níveis de metais (Al, Ba, Cr, Cu, Fe, Mn, Pb, Ti e Zn) em diferentes matrizes ambientais (solo, água, sedimento e ictiofauna) em três sub-bacias da bacia hidrográfica do rio Itapemirim, a fim de avaliar a influência do uso do solo nas concentrações desses elementos.

II. Comparar os níveis de metais encontrados nas matrizes ambientais com valores estabelecidos por órgãos normativos, visando avaliar se as concentrações encontradas estão dentro do estabelecido como critério de qualidade.

III. Avaliar os fatores de contaminação e de enriquecimento do sedimento, bem como a associação dos elementos com os principais suportes geoquímicos (Al, Fe, Mn e Carbono orgânico), a fim de identificar possíveis fontes.

ARTIGO

Land use influence on the metal concentrations of three southeast Brazil sub-basins

Land use influence on the metal concentrations of three southeast Brazil sub-basins

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Abstract

The particle transference to aquatic environments is affected by differences in land use. The conversion of the native groundcover to crop in agricultural areas, together with the use of pesticides and fertilizers, leads to the contamination of aquatic bodies. In this study we evaluated the effects of land use on the metal concentrations (Al, Ba, Cr, Cu, Fe, Mn, Pb, Ti, and Zn) of different environmental matrices (soil, water, sediment, and ichthyofauna) in three sub-basins (BND, BNE, and IT) of an important Brazilian coffee producing area. In general, the metal concentrations in environmental matrices were BNE>BND>IT. The contamination factors (CF), pollution load index (PLI), and enrichment factor (EF) bolstered the findings indicating the BNE as the sub-basin that has received the most impact, represented by the following order: BNE>IT>BND. The principal component analysis revealed a lower association of Cr, Cu, Ni, and Zn with the main geochemical supports (Al, Fe, Mn, Ti, and organic carbon) that indicate anthropogenic influences related to these elements. The higher metal concentrations in the BNE, affecting the water, sediment, and fish, correspond to an area of extensive agricultural activities, involving mainly coffee cultivation, and are related to superficial runoff from soils and crops applied with pesticides and fertilizers and a deficiency of municipal sewage treatment. In contrast, the lower geochemical indices in the BND are related to the higher percentage of native forest cover, which reduces the transport of particles to the sub-basin.

Key words: Sediment, geochemical indexes, coffee cultivation, contamination.

1. Introduction

The dynamics of metal biogeochemistry in aquatic environments is an important issue due to the toxicity, persistence and the essentiality of some elements (COOKE; ANDREWS; JOHNSON, 1990; DENISEGER *et al.*, 1990). The metals discharged into an aquatic body are distributed through the water column, interact with biota, or can be buried in the bottom sediment (SIN *et al.*, 2001). Processes such as adsorption, colloidal polymerization, precipitation, and co-precipitation lead to the deposition of metals in the sediment, and relatively small portions remain free as dissolved in water (GAUR *et al.*, 2005). In sediments these elements can be remobilized due to physical disturbances and geochemical alterations, reaching the water column when it is available to accumulate in the food chain (EGGLETON; THOMAS, 2004; ZHANG *et al.*, 2014). Therefore, sediments are both a sink and a source of metals to aquatic environments (SUPERVILLE *et al.*, 2014).

Different kinds of vegetal cover can influence superficial runoff and thereby interfere in the chemical form as well in the bioavailability of metals to the aquatic system, metal concentrations in the aquatic environment are associated with land use configurations in each river basin. Urban and agricultural soils generally presents higher heavy metal concentrations due de use of fertilizers and pesticides representing a source of these elements, while urban soils, distinct human activities can affect metal concentrations (ANCION; LEAR; LEWIS, 2010; ATAFAR *et al.*, 2010; CHRISTENSEN; NAKAJIMA; BAUN, 2006; GU *et al.*, 2012; JOSHI; BALASUBRAMANIAN, 2010; ZHAO *et al.*, 2010). Moreover, the overuse of products by farmers, possibly due to a lack of information regarding these practices, often intensifies the problem (ATAFAR *et al.*, 2010; DEVRIES *et al.*, 2002; NOURI *et al.*, 2008; TAYLOR; PERCIVAL, 2001). In this way, elements can be absorbed by plants, accumulated by animals, and can reach humans through the food chain (TAYLOR; PERCIVAL, 2001).

Brazil is the world’s largest coffee producer and ***Espírito Santo*** State (ES) in the southeast is the second largest producer state (IBGE, 2017). The sub-basins evaluated in this study are located in the ***Caparaó*** region, southwestern ***Espírito Santo*** State, an area responsible for nearly 19 % of the state’s coffee production. Since the arrival of coffee cultivation at the beginning of the 19th century, this region has undergone impacts related to changes in land use characterized by the expansion of agricultural areas, the application of pesticides and fertilizers, which may have caused the metal contamination of aquatic environments by metals (ESPÍRITO SANTO, 2005; IJNS (2006); PEDLOWSKI *et al.*, 2012).

Due to the historical coffee plantations areas and the probable associated use of pesticides and fertilizers, that might be a potential source of metals, itis necessary to investigate the concentrations of these chemicals in the region. From this perspective, the aim of this study was to evaluate the influence of agricultural lands, mainly those used for coffee production, on the metal concentrations in the sediment, water, and fish of three sub-basins with distinct land use.

2. Material and methods

2.1. Study area and sampling

The present study compared the metal concentrations and land use patterns among three sub-basins: the ***Itapemirim*** River (IT), the ***Braço Norte Esquerdo*** River (BNE), and the ***Braço Norte Direito*** River (BND) (Figure 1). The ***Itapemirim*** River is formed by the union of the BNE and BND rivers, and together they integrate the ***Itapemirim*** basin with a total area of 5,900 km^2. Sampling occurred in the high elevation coffee plantation area of the IT basin, in the upstream section, which corresponds to about 40% of its total area. The studied area was formed by the BNE sub-basin, which is the largest of the three with an area of 1,432 km^2, followed by the BND, with 510 km^{2,} and the IT sub-basin, with 442 km^2. However, as the IT sub-basin receives water from the BNE and BND, its total area reaches 2,384 km^2 (Figure 1). Both BND and BNE have dams that are located nearly of the junction to form IT River (Figure 1A).

The rivers' maximum flows (BND: 29.6 m^3/s, BNE: 31.9 m^3/s, IT: 78.7 m^3/s) occur from October to April, whereas the minimum flows (BND: 3.0 m^3/s, BNE: 6.9 m^3/s, IT: 15.1 m^3/s) occur from June to September. The mean annual flow of are 12.20, 17.90 and 42.70 m^3/s, while the mean temperature in BND, BNE ant IT is 24.0 ºC, 26.2 ºC, and 27.2 ºC, respectively and the annual precipitation is about 1400 mm where rainy period from October to April (mean = 164 mm) and dry period from June to September (mean = 34 mm). The total population reaches about 131.000 inhabitants in the three sub-basins (IBGE, 2010; ANA, 2017).

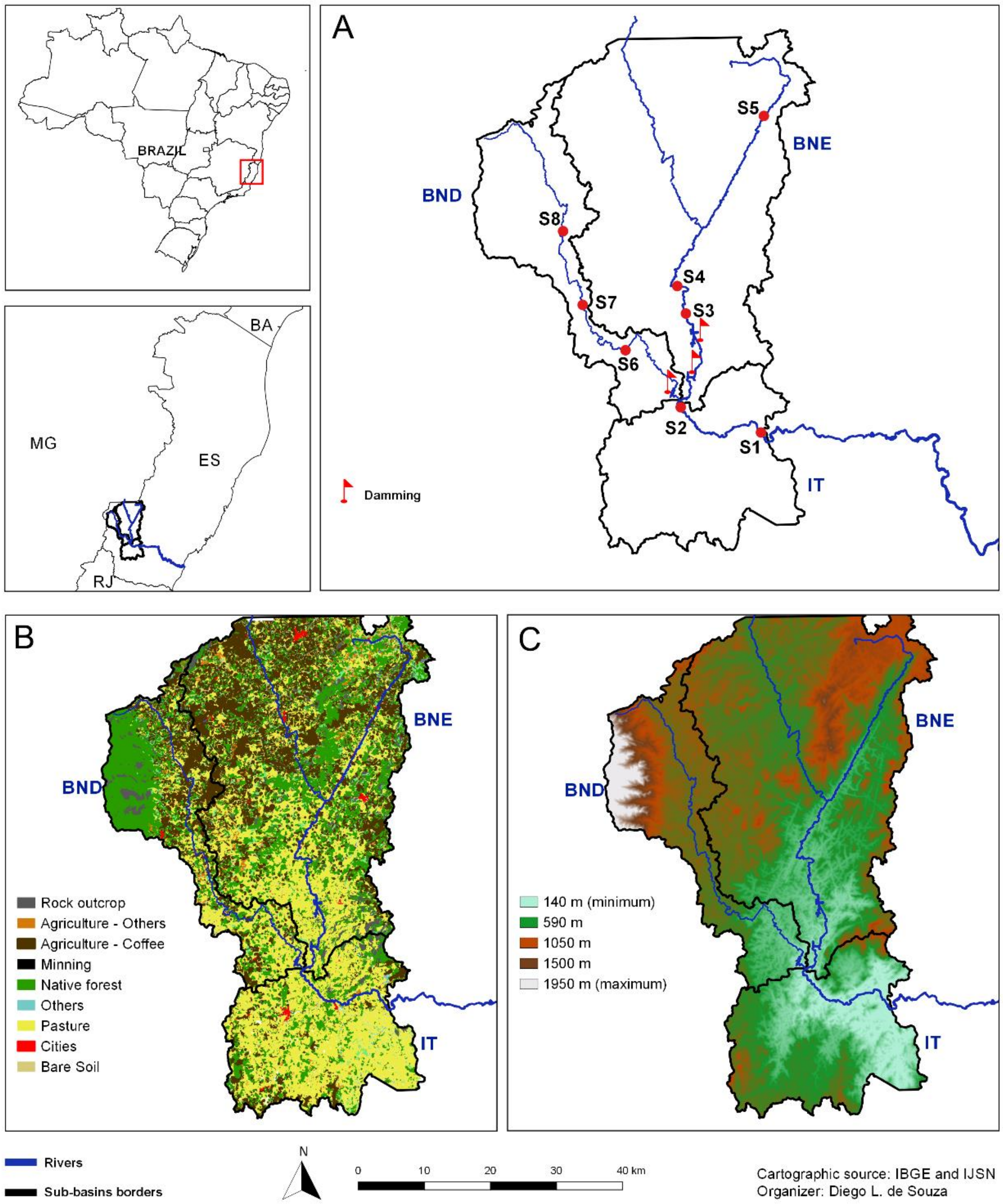

Figure 1: Map of the study area. A. Sampling sites in the Itapemirim (IT), Braço Norte Esquerdo (BNE), and Braço Norte Direito (BND) sub-basins. B. Land use map of the sub-basins. C. Elevation profile of the sub-basins.

For the assessment of metal concentrations, aliquots of surface sediment and water were collected at eight sampling sites: two in the main course of the ***Itapemirim*** River (S1 and S2), three in each one of the two main tributaries, the ***Braço Norte Esquerdo*** (S3 – S5) and ***Braço Norte Direito*** rivers (S6 – S8). These assessments were carried out in two sampling periods (July 2016 and January 2017), which represent the dry and the rainy seasons. Adjacent soil was also collected at each water and sediment sampling site to verify the influences of soil metal concentrations in the rivers (Figure 1A; Table 1). These sub-basins were chosen because of differing land use characteristics. The BND sub-basin contains the most preserved areas, the BNE is composed of mostly of coffee plantations, and the IT is characterized mainly by pasture areas (Figure 1B). The sub-basins also differ in relief, with the IT composed of a flatter relief and the lowest elevations, the BNE has maximum altitudes of approximately 1,500 meters, while the BND is the most elevated sub-basin with altitudes reaching 1,950 meters (Figure 1C).

Table 1: Coordinates of the sampling sites of the Braço Norte Direito (BND), Braço Norte Esquerdo (BNE), and Itapemirim (IT).

River	Site	Latitude	Longitude
BND	S8	20°28'7.20"S	41°40'26.09"W
	S7	20°34'11.50"S	41°38'49.93"W
	S6	20°37'57.97"S	41°35'9.92"W
BNE	S5	20°32'46.04"S	41°30'34.74"W
	S4	20°18'55.00"S	41°22'49.19"W
	S3	20°35'2.27"S	41°29'51.53"W
IT	S2	20°42'43.15"S	41°30'26.09"W
	S1	20°44'53.13"S	41°23'28.36"W

At each sampling site, the physical-chemical parameters of pH, electrical conductivity, dissolved oxygen, turbidity, and temperature were measured. The surface sediments were collected in plastic bags and kept under refrigeration at -20 ºC until the analysis. Sediment samples were freeze-dried and one portion was fractionated for granulometric characterization in a Laser Diffraction Particle Size Analyzer (Shimadzu SALD-3101), while the rest was fractionated (<2 mm). The fractionated samples were homogenized with grail and pistil for subsequent metal determinations. Water samples were collected in plastic bottles, acidificated to 1% v/v with HNO_3, and kept under refrigeration (-20ºC) until the metal analysis.

Ichthyofauna sampling was carried out in both dry and rainy seasons at three sampling sites: one in the main course of the IT (S1), and the others in its tributaries, the BNE (S4) and BND (S7), with electric fishing equipment. The collected fish were anesthetized in benzocaine 300 mg/L, weighed, measured (total length), and necropsied for removal of muscle tissue for metal analysis. The omnivorous fish species Acará (*Geophagus brasiliensis)* was chosen due to its abundance in the study area (SARMENTO-SOARES; MARTINS-PINHEIRO, 2013).

2.2. Land use characterization of the sub-basins

The land use description in each sub-basin was made compiling data from of land use file vectors of 2006 of the ***Jones dos Santos Neves Institute*** (JSNI) using Quantum Gis software. The summarized data was expressed as percentages of land use classes (pasture, native forest, cities, rock outcrop, mining, agriculture, bare soil, and other uses) in each sub-basin. The digital elevation model was obtained from the Topodata Project, which maintains a database of the elevation and geomorphometric variables for Brazil (MORISSON and ROSSETTI, 2012).

2.3. Metal quantification

Sediment and soil determinations were performed according to USEPA (1996) method 3052 by dissolving an aliquot of 0.5 g (dry weight) in a solution of HNO_3 65% (9 mL) + HF 48 % (4 mL) + HCl 37% (4 mL) by microwave (Mars 5 Xpress CEM-Corporation) for 40 minutes, heated at 180 °C with a ramp time of 10 minutes, and held for 30 minutes. Next 25 mL of 4% H_3BO_3 solution (m/v) was added to complex the possible HF residue. The final extract was reheated to 180 °C as described above. After cooling, the extracts were filtered (Whatman 40), resulting in a final volume of 50 mL with 0.5% v/v HNO_3. The total concentrations metal in water (dissolved + particulate) samples were processed according to USEPA (1994) method 3015 by microwave (Mars 5 Xpress-CEM Corporation) in HNO_3 65% (22.5 mL water + 2.5 mL HNO_3) at 165 °C for 30 minutes with a ramp time of 10 minutes and then held for 20 minutes, filtered with filter paper (Whatman 40), and diluted to a final volume of 25mL with 0.5% v/v HNO_3.

To determine metal concentrations in fish muscle (n = 102, with total length ranging from 5.9 to 20 cm and weight from 5.35 to 132 g), an aliquot of 1.0 g (dry weight) solubilized in 10 mL of 65% HNO_3 at 150 °C for 12 h was used. After this step it was reheated up to 190 °C until almost the dry point. Subsequently, the samples were filtered with filter paper (Whatman 40) and resuspended at 15 mL with 0.5% v/v HNO_3 for analysis (USEPA, 1992). Water, sediment, and muscle tissue samples were made in duplicates, considering a variation below 15%. Metal determinations (Al, Ba, Cr, Cu, Fe, Mn, Ni, Pb, Ti, and Zn) were performed with spectrometry coupled plasma optical emission (ICP - OES Varian - 720 ES) and the methods' detection limit (ppm) was: Al = 0.007; Ba = 0.003; Cr = 0.004; Cu = 0.004; Fe = 0.007; Mn = 0.003; Ni = 0.004; Ti = 0.003 and Zn = 0.003.

Standard reference material 1646a of estuarine sediments was used to determine methodological accuracy, following the same procedures mentioned above. The recovery results between 70 and 110% were considered (Table 2).

Table 2: Validation of the analytical procedure using Nist Standard Reference Material® 1646a of estuarine sediments (mg/kg), n= 3.

	Al	**Ba**	**Cr**	**Cu**	**Fe**	**Mn**	**Ni**	**Pb**	**Ti**	**Zn**
Found value	2191.4	175.0	37.7	9.2	2110.8	202.0	17.2	8.3	483.0	45.9
Certified value	2300.0	210	40.9	10.1	2010.0	234.5	23	11.7	456.0	48.9
Recovery %	**95**	**83**	**92**	**91**	**105**	**86**	**75**	**71**	**106**	**94**

2.4. Carbon concentration and isotopic composition of sediment

The carbon concentration (%) and isotopic composition ($\delta^{13}C$ ‰) was determined after removing carbonate using approximately 12 mg of dry and homogenized sediments, in silver vials. This analysis was performed in an organic elemental analyzer (Flash 2000) coupled with an isotope ratio mass spectrometer (Thermo Scientific Delta V Advantage). The methods' validation was made by standard OAS/isotope-low organic soil (elemental microanalysis) with a recovery of 94 % and sampling precision above 95%.

2.5. Assessment of sediment contamination

Pollution indices can be used in geochemical analysis for the assessment of sediment contamination. However, these approaches require the background values of the study areas. As no such data on background concentrations exists, the lower metal concentrations found in the sediment of each sub-basin were used as background values in this paper because these concentrations were assumed to be those closest to the background values. In this study, four different indices were used: Contamination factor, Pollution load index, Enrichment factor and the sediment quality guidelines stablished by NOAA: TEL (threshold effects levels) and PEL (probable effects levels).

2.5.1. Contamination factor (CF)

The CF is the ratio between each metal concentration in the samples and the background value:

$$\mathrm{CF} = \frac{(Metal_{Sample})}{(Metal_{Background})}$$

The CF values are interpreted as follow: CF < 1 indicates low contamination; 1 < CF < 3 is moderate contamination; 3 < CF < 6 is considerable contamination; CF > 6 is very high contamination (HAKANSON, 1980).

2.5.2. Pollution load index (PLI)

For the entire sampling site, PLI has been determined as the *n*th root of the product of the *n* CF:

$$\mathrm{PLI} = (\mathrm{CF}_1 \mathrm{\ x\ } \mathrm{CF}_2 \mathrm{\ x\ } \mathrm{CF}_3 \mathrm{\ x} \ldots \mathrm{x\ } \mathrm{CF}_{\mathbf{n}})^{1/\mathrm{n}}$$

This index provides comparative means for assessing the level of metal pollution. When PLI > 1 pollution exists, and when PLI < 1, it is assumed that there is no pollution (TOMLINSON *et al.*, 1980).

2.5.3 Enrichment factor (EF)

The enrichment factor is frequently used in geochemical studies to determine the degree of anthropogenic metal pollution:

$$\mathrm{EF} = \frac{\left(\frac{\mathrm{Metal}}{\mathrm{Al}}\right)\mathrm{sample}}{\left(\frac{\mathrm{Metal}}{\mathrm{Al}}\right)\mathrm{background}}$$

Aluminum was used as the normalizing element because of its conservative feature and low mobility, which reduce the effect of grain size and mineralogy. According to Sakan *et al.* (2009), the EF values are interpreted as follows: EF < 1 indicates no enrichment; < 3 is minor enrichment; 3 – 5 is moderate enrichment;

5 – 10 is moderately severe enrichment; 10 – 25 is severe enrichment; 25 – 50 is very severe enrichment; and > 50 is extremely severe enrichment.

2.6. Multivariate analysis

To evaluate the spatial variability of the elements between the sampling sites and the element's associations with possible sources of metals in the sub-basins, multivariate cluster analysis (CA) and principal component analysis (PCA) were conducted. Cluster analysis was applied to experimental data standardized through z-scale transformation to avoid misclassification due to differences in data dimensionality and PCA was conducted for the data in log scale. Hierarchical agglomerative cluster analysis was performed using Ward's method with squared Euclidean distances (VAROL; ŞEN, 2009). Both analysis was conducted using STATISTICA software, version 8.0.

2.7. Statistics

The normality of data was verified using plots of the quantiles (qqplot) and when necessary normalized using simple visual transformations (boxcox). The data obtained from metal and isotopic analysis were submitted to analyses of variance and the means compared by Tukey test considering $p < 0.05$ using R program. Due the low number of sampling sites, the statistical analysis was conducted only to compare the seasonal variability.

3. Results and discussion

The three studied sub-basins show distinct land uses (Figure 2). The data compiled shows that the BNE sub-basin is mainly influenced by cities, coffee plantation areas, other agricultural activities, and bare soil. The BND sub-basin is characterized by large fragments of native forest while the BNE areas of native forest are small fragments, as can be observed in figure 1. Meanwhile, pasture areas are located mainly in the IT sub-basin (~60%), and mining activities are solely present in the BNE and IT sub-basins (Figure 2). In the region, the most developed mining activities are the extraction of ornamental rocks like marble, granite, and sand. Such practice serves to revolve rock, soil, or sediment, leading to the release of metals to the environment and altering the geochemical characteristics of these matrices (CHEN, HAIYANG *et al.*, 2016; ZHANG *et al.*, 2014).

Many land uses comprise less than 10% of the sub-basins and include mining, cities, bare soil, agriculture (others), and rock outcrop. Coffee plantations cover 8% of the IT sub-basin, 18% in BND, and 23% in BNE. Native forest covers about 30% of BND and 17% of IT and BNE. Pasture is the main land cover class in the three sub-basins, reaching about 60% of the IT sub-basin (Figure 2).

Therefore, according to the land use and carbon isotope values, it is possible to differentiate the three studied sub-basins: (1) BND with a predominance of larger fragments of native forest areas, (2) BNE with its predominance of coffee plantations and other agricultural activities, and (3) IT with a pasture predominance.

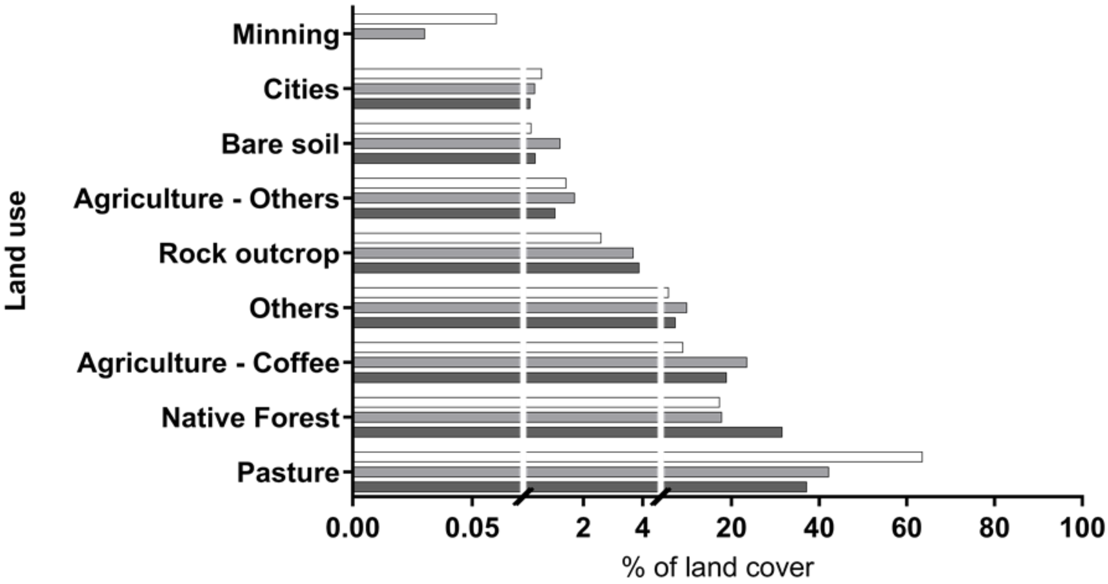

Figure 2: Percentage of each land use classes in the Braço Norte Direito (BND), Braço Norte Esquerdo (BNE), and Itapemirim (IT) sub-basins.

Water physical-chemical parameters indicated a tendency of soil influence in the water column (Table 3). The conductivity and turbidity levels were higher during the rainy season, due to the increase in precipitation and the transport of particulate material and ions to the aquatic environment. Instead, the pH (with some exceptions) and dissolved oxygen values were lower during the rainy season. This is related to the dissolved and particulate organic matter carried to rivers and the resulting consumption of O_2 and production of CO_2, a process that forms carbonic acid in water, reduces the pH, and depletes the dissolved oxygen. In addition, the higher turbidity in the rainy season reduces the light entering the water column, thereby inhibiting photosynthesis and its role in the reduction of dissolved oxygen (Table 3) (TUNDISI; MATSUMURA, 2011). The physical characteristics of the sediments also showed a tendency of particulate transport to the rivers that coincided with the beginning of the rainy season (Table 3).

Table 3: Physical chemical parameters in water (conductivity, pH, temperature, dissolved oxygen, and turbidity) and sediments (granulometry, organic carbon, and isotopic carbon composition) of the Braço Norte Direito (BND), Braço Norte Esquerdo (BNE), and Itapemirim (IT) Rivers in dry and rainy seasons. ± - Standard deviation.

			Water				**Sediment**			
Season	**River**	**Site**	**Conductivity (µS/cm)**	**pH**	**DO (mg/L)**	**Turbidity (NTU)**	**Sand (%)**	**Silt and Clay (%)**	**C (%)**	**δC13 ‰**
Dry	BND	S8	24.7	6.98	7.33	2.89	96.8	3.2	0.15	-24.5
		S7	27.0	7.11	7.02	2.91	97.8	2.2	0.08	-24.2
		S6	22.0	7.78	6.94	2.33	100.0	0.0	0.07	-23.4
	BNE	S5	36.4	6.67	4.20	1.82	100.0	0.0	0.05	-22.6
		S4	54.9	6.29	8.80	4.28	73.7	26.3	0.71	-23.0
		S3	59.1	7.42	6.95	2.87	95.4	4.6	0.19	-22.6
	IT	S2	45.4	7.05	6.47	2.69	100.0	0.0	0.18	-21.9
		S1	47.8	6.91	8.23	3.49	96.5	3.5	0.03	-21.2
	Mean	BND	24.6 ± 2.5	7.3 ± 0.4	7.1 ± 0.2	2.7 ± 0.3	97.3 ± 0.7	2.7 ± 0.7	0.12 ± 0.05	-24.4 ± 0.2
		BNE	50.1 ± 12.1	6.8 ± 0.6	6.7 ± 2.3	3.0 ± 1.2	84.6 ± 15.4	15.4 ± 15.4	0.45 ± 0.37	-22.8 ± 0.3
		IT	46.6 ± 1.7	7.0 ± 0.1	7.4 ± 1.2	3.1 ± 0.6	98.3 ± 2.4	1.7 ± 2.4	0.10 ± 0.11	-21.5 ± 0.5
Rainy	BND	S8	26.6	6.40	5.20	2.62	93.5	6.5	0.14	-24.5
		S7	23.4	7.27	6.22	3.03	100.0	0.0	0.08	-23.7
		S6	40.0	5.98	6.41	-	93.7	6.3	0.21	-23.8
	BNE	S5	50.6	6.85	5.23	3.82	99.9	0.1	0.04	-22.5
		S4	58.8	7.21	7.34	9.03	99.9	0.1	0.08	-23.4
		S3	60.8	6.86	6.15	9.77	14.5	85.5	2.23	-22.1
	IT	S2	56.7	6.48	6.21	5.55	99.9	0.1	0.05	-22.2
		S1	57.9	6.09	7.54	5.26	100.0	0.0	0.05	-22.0
	Mean	BND	30.0 ± 8.8	6.6 ± 0.7	5.9 ± 0.7	2.8 ± 0.3	95.7 ± 3.7	4.3 ± 3.7	0.15 ± 0.07	-24.0 ± 0.4
		BNE	56.7 ± 5.4	7.0 ± 0.2	6.2 ± 1.1	7.5 ± 3.2	57.2 ± 60.4	42.8 ± 60.4	1.16 ± 1.52	-22.7 ± 0.9
		IT	57.3 ± 0.8	6.3 ± 0.3	6.9 ± 0.9	5.4 ± 0.2	99.9 ± 0.1	0.1 ± 0.1	0.05 ± 0.01	-22.1 ± 0.1

A change in particle size mean values was observed, mainly in the BNE but these values are strongly influenced by Site 3. In the dry season Site 3 was composed of 95 % sand, while during the rainy season it contained 85 % silt and clay fraction

(Table 3). This observation may be related to S3's proximity to a dam (Figure 1A), as river flow reductions increase fine particle sedimentation (MAGILLIGAN; NISLOW, 2005). The percentage of organic carbon in both seasons was BNE > BND > IT and these values increased in the rainy season in BNE and BND. These results can be explained by the fact that BNE presents high amounts of fine particles that adsorb organic matter (ANDREWS *et al.*, 2004; CHEN, MEILIAN; HUR, 2015). The carbon isotopic composition showed the same pattern in both seasons, with the following ratio order of the rivers: IT > BNE > BND (Table 3). The isotopic carbon composition of sediments is an indicator of vegetal cover, since basins with predominance of C3 plants show a lighter isotopic ratio (-30 to -27 ‰) of carbon isotopes, while a mixture of plants with C3 and C4 metabolism display a heavier isotopic ratio (-28 to -19 ‰) (MARTINELLI *et al.*, 1999). The most negative values in ratio of carbon isotopes observed in BND is related to the native forest area, whereas the higher ratios in IT demonstrate the pasture cover of its sub-basin (Table 3).

The differences between the sub-basins are also observed in soil metal concentrations. IT and BNE showed the same order of elements Fe > Al > Ti > Ba > Mn > Zn > Cr > Pb > Ni > Cu, while a different order was observed in BND: Fe > Al > Ti > Mn > Ba > Cr > Zn > Ni > Cu > Pb. Some of these concentrations were statistically different between the sub-basins ($p<0.05$) (Table 4). The higher concentrations of Cr, Cu, Fe, Ni, Ti, and Zn were found in BND, while IT showed higher values of Ba, Mn, and Pb, and BNE the highest values of Al. The concentrations of almost all elements were within the limits of Brazilian Environmental Legislation for soils - CONAMA 420/2009, only Ba exceeded the limits for agricultural soils at site 5 (BNE), and in IT (sites 1 and 2) and Ni exceed the established value at site 7 (BND), indicating that these values can be harmful to the environment (Table 4).

The comparison of this study with other soil metal concentrations showed that Cr concentrations found in BND were higher than mean values from urban soils, urban road dusts, and agricultural soils in China, which showed the highest values of most of the metals compared. Pb concentrations in BND, BNE, and IT were higher than the concentrations found in other studied agricultural areas in Brazil and in the USA. Ni and Zn levels in BND, BNE, and IT were also higher than other studies in Brazil and lower than other countries compared. Meanwhile, the Cu and Mn values in the present study were lower in comparison with those of most other countries and other levels found in Brazil (Table 4). The variations in metal concentrations in soils generally reflect pedogenic factors and anthropogenic sources (DAVIES; WIXSON, 1987). The general lower concentrations of metals found in the Caparaó region (ES) in the present study, in comparison with other countries, with exception of Cr and Pb, is due to the occurrence of the three sub-basins above crystallin pre cambrian rocks (EMBRAPA, 1978). These rocks generally present lower concentrations of metals compared with other rock types (PAYE *et al.*, 2010).

Table 4: Metal concentrations in soils in both seasons from the Braço Norte Direito (BND), Braço Norte Esquerdo (BNE), and Itapemirim (IT) sub-basins and the comparison with normative legislation of Brazil and other studies. ± - Standard deviation; <LD - Below limit method's detection. Different letters represent statistically significant values ($p<0.05$).

River	Site	Al (mg/g)	Ba (µg/g)	Cr (µg/g)	Cu (µg/g)	Fe (mg/g)
BND	S8	31 ± 2	135 ± 20	55 ± 6	11 ± 1.2	54 ± 8
	S7	26 ± 4	80 ± 12	111 ± 14	24 ± 2.1	56 ± 3
	S6	19 ± 3	117 ± 16	57 ± 7	10 ± 0.2	48 ± 6
BNE	S5	20 ± 3	390 ± 58	22 ± 3	4 ± 0.2	23 ± 3
	S4	53 ± 8	65 ± 9	35 ± 2	3 ± 0.4	39 ± 1
	S3	30 ± 4	89 ± 6	22 ± 2	3 ± 0.3	49 ± 6
IT	S2	25 ± 3	266 ± 38	14 ± 2	7 ± 0.8	23 ± 3
	S1	18 ± 2	358 ± 39	22 ± 2	7 ± 0.2	38 ± 1
Mean	BND	25 ± 6 a	111 ± 28 b	74 ± 32 a	15 ± 7.7 a	53 ± 4 a
	BNE	34 ± 17 a	181 ± 181 ab	27 ± 8 ab	4 ± 0.8 c	37 ± 13 b
	IT	22 ± 5 a	312 ± 65 a	18 ± 6 b	7 ± 0.1 b	30 ± 10 b
Comparison with literature	China			58.9	31.3	
	USA			-	18.5	
	Spain			20.3	17.3	
	Brazil (RO e MT)			39.4	16.5	
	Brazil (ES)			41.0	5.5	
CONAMA 420/2009	Warning		150	75	60	
	Agricultural soils		300	150	200	

River	Site	Mn (µg/g)	Ni (µg/g)	Pb (µg/g)	Ti (mg/g)	Zn (µg/g)
BND	S8	338 ± 36	15 ± 1.2	11 ± 1	13 ± 1.2	48 ± 6
	S7	145 ± 20	33 ± 4.1	16 ± 1	8 ± 1.2	41 ± 6
	S6	167 ± 16	18 ± 0.9	11 ± 1	8 ± 1.1	36 ± 3
BNE	S5	283 ± 35	6 ± 0.5	13 ± 1	4 ± 0.5	24 ± 2
	S4	95 ± 4	11 ± 0.3	10 ± 1	9 ± 0.3	36 ± 2
	S3	131 ± 18	6 ± 0.1	15 ± 2	7 ± 0.3	28 ± 9
IT						
	S2	146 ± 13	5 ± 0.5	14 ± 2	2 ± 0.3	30 ± 4
	S1	303 ± 45	9 ± 0.2	12 ± 1	9 ± 1.0	38 ± 2
Mean	BND	216 ± 105 a	22 ± 9 a	13 ± 3 a	10 ± 3.1 a	42 ± 6 a
	BNE	170 ± 100 a	8 ± 3 b	13 ± 2 a	6 ± 2.7 a	29 ± 6 a
	IT	225 ± 111 a	7 ± 3 b	13 ± 1 a	6 ± 4.7 a	34 ± 6 a
Comparison with literature	China		27.5	37.7		113.2
	USA		18.2	11.0		53.0
	Spain		20.5	17.5		57.5
	Brazil (RO e MT)		1.3	8.1		6.8
	Brazil (ES)	131.6	6.6	8.8		22.6
CONAMA 420/2009	Warning		30	72		300
	Agricultural soils		70	180		450

China: Wei and Yang, (2010); USA: Holmgren *et al.,* (1993); Spain: Rodríguez Martín, Arias and Grau Corbí, (2006); Brazil (RO and MT): Santos and Alleoni (2013); Brazil (ES): Paye *et al.,* (2010).

As soil surface runoff can carry metals to rivers, the water column, fish, and sediment were also evaluated in the present study. In water, the order of metal concentrations in BND was Fe > Zn > Al > Cu > Ba > Mn; in BNE, Fe > Al > Zn > Mn > Ba > Cu; and in IT, Fe > Al > Zn > Cu > Ba > Mn (Table 4), Cr, Ni, Pb and Ti were below the detection limit. The highest concentrations of the elements (Al, Ba, Fe, and Mn) was observed in the BNE River, while the BND had the highest concentrations only for Cu, Ni, and Zn (Table 5). Ba concentration, which was higher in the dry season in the BND River (mainly due to Site 7), was not observed in this sub-basin during the rainy season, when levels in the BNE River increased. Site 7 also presented higher values of Zn in the dry season and this fact may be related to its proximity to the ***Ibitirama*** municipality, which discharges raw sewage directly into the water. Likewise, Site 3 also shows higher concentrations during this season, and this site is similarly close to the city of ***Jerônimo Monteiro***, which, according to Corrêa Martins, Souza and Silva Souza (2016), treats about 60% of its sewage, with the remainder discharged directly into the environment. Some of these concentrations were statistically different between the sub-basins ($p<0.05$) (Table 5).

Al, Cu, Fe, and Zn are into the levels established by Brazilian Legislation CONAMA 357/2005 for Level 2 aquatic bodies in both seasons for the three sub-basins (BND, BNE, and IT) being necessary conventional treatment before being destined for domestic use, preservation of aquatic communities, plant irrigation, and human consumption). Hence, they can be harmful to organisms if the correct treatment is not applied (Table 5).

The comparison with the literature show that the concentrations found in these sub-basins are higher than those found in other studies in Brazil that analyzed concentrations in water from areas affected mostly by agricultural activities (Table 5). However, the levels were lower than the found in rivers undergoing impact (mainly from urbanization and industrialization) in other countries (the Tsurumi, Tigris, and Odra Rivers). Therefore, the higher metal concentrations found in water in this study when compared with other studies in Brazil might be related to the agricultural activities areas of the sub-basins and the lack of municipal sewage treatment (Table 5).

Table 5: Metal concentrations in water (mg/L) of the Braço Norte Direito (BND), Braço Norte Esquerdo (BNE), and Itapemirim (IT) Rivers in dry and rainy seasons, the comparison with normative legislation in Brazil, and values found in other studies. ± - Standard deviation; <LD – Below limit method's detection. Different letters represent statistically significant values (p<0.05).

Season	**River**	**Site**	**Al**	**Ba**	**Cu**	**Fe**	**Mn**	**Zn**
Dry	BND	S8	107 ± 15	11 ± 0.1	15 ± 2	663 ± 26	10 ± 0.3	47 ± 3
		S7	342 ± 5	477 ± 11.3	11 ± 0	951 ± 23	15 ± 0.2	836 ± 32
		S6	110 ± 12	13 ± 1.6	11 ± 1	422 ± 8	8 ± 1.1	22 ± 3
	BNE	S5	1047 ± 152	20 ± 1.6	11 ± 1	1379 ± 116	39 ± 5.3	43 ± 7
		S4	89 ± 12	14 ± 0.7	20 ± 3	369 ± 11	8 ± 1.1	101 ± 12
		S3	84 ± 12	16 ± 0.1	7 ± 1	341 ± 13	9 ± 1.0	36 ± 2
	IT	S2	204 ± 16	18 ± 0.7	12 ± 0	288 ± 9	14 ± 1.1	83 ± 9
		S1	174 ± 6	18 ± 0.3	15 ± 0	247 ± 4	12 ± 0.4	199 ± 22
	Mean	BND	186 ± 135	167 ± 269	12 ± 2	679 ± 265	11 ± 3.5	302 ± 463
		BNE	407 ± 555	17 ± 3	13 ± 7	696 ± 591	19 ± 17.6	60 ± 36
		IT	174 ± 21	18 ± 1	13 ± 2	268 ± 29	13 ± 1.7	141 ± 82
Rainy	BND	S8	35 ± 3.3	<3 ± <3	9 ± 0.5	348 ± 41	8 ± 1.2	3 ± 0.4
		S7	37 ± 2.3	<3 ± <3	9 ± 0.8	308 ± 36	<4 ± <4	3 ± 0.2
		S6	266 ± 11.2	<3 ± <3	8 ± 1.2	965 ± 7	12 ± 0.8	<3 ± <3
	BNE	S5	408 ± 0.1	22.1 ± 1.2	9 ± 0.3	1361 ± 126	29 ± 3.4	<3 ± <3
		S4	142 ± 18.3	17.3 ± 0.2	9 ± 1.2	1520 ± 49	32 ± 1.5	68 ± 9.5
		S3	403 ± 53.8	25.5 ± 0.1	8 ± 1.2	1318 ± 12	39 ± 1.7	<3 ± <3
	IT	S2	53 ± 7.5	15.4 ± 0.2	11 ± 6.0	354 ± 47	13 ± 0.9	<3 ± <3
		S1	36 ± 5.3	18.6 ± 0.4	9 ± 0.6	274 ± 12	7 ± 0.5	10 ± 0.7
	Mean	BND	113 ± 132.9	<3 ± <3	9 ± 0.8	540 ± 368	10 ± 2.4	3 ± 0.3
		BNE	318 ± 152.2	21.6 ± 4.1	9 ± 0.3	1400 ± 106	33 ± 5.3	68 ± -
		IT	45 ± 11.8	18.6 ± 2.2	10 ± 1.8	314 ± 57	10 ± 4.3	10 ± -
	CONAMA 357/2005		100.0	700.0	9.0	300.0	100.0	180.0
Comparison with literature	Tsurumi River, Japan				654	362	264	
	Tigris River, Turkey				165	388	467	37
	Odra River, Poland				54	1,861	353	535
	Nhue River, Vietnam				14		207	61
	Pardo River, Brazil				2		43	12
	Sinos River, Brazil		35		6	36	48	3
Dry and Rainy	Mean	BND	150 ± 126 a	167 ± 268.5 a	11 ± 2.3 a	609 ± 297 a	11 ± 2.8 a	182 ± 366 a
		BNE	362 ± 367 a	19 ± 4.2 b	11 ± 4.9 a	1048 ± 541 a	26 ± 14.1 a	62 ± 30 a
		IT	117 ± 5 a	18 ± 0.2 b	12 ± 2.8 a	291 ± 20 b	12 ± 0.3 a	98 ± 10 a

Tsurumi River, Spain: Mohiuddin *et al.,* (2009); Tigris River, Turkei : Varol and Şen (2012); Odra River, Poland: Adamiec and Helios-Rybicka (2002); Nhue River, Vietnam: Kikuchi *et al.,* (2009); Pardo River, Brazil: Alves *et al.,* (2014); Sinos River, Brazil: Weber *et al.,* (2013).

Soil particles can reach the sediment, and the evaluation of metal concentrations in this compartment may indicate anthropogenic influences on the river system (ZHANG *et al.*, 2014). This was observed analyzing the metal concentrations in the sediment of the rivers in the present study (Table 6). During the dry season, the BNE river showed higher concentrations of Al, Mn, Pb, and Ti; the BND of Cr, Cu, Fe, Ni, and Zn; and the IT for Ba. With the beginning of the rainy season, while the concentrations of almost all elements (except Pb) increased in the three rivers (IT, BNE, BND), in the BNE river, this was accomplished by the presence of a higher number of elements (Al, Cu, Fe, Mn, Ni, Pb, Ti, and Zn), while only Cr showed highest levels in BND and Ba again in the IT river. Furthermore, in the

Table 6: Metal concentrations in sediments of the Braço Norte Direito (BND), Braço Norte Esquerdo (BNE), and Itapemirim (IT) Rivers in both dry and rainy seasons and comparisons with TEL, PEL, and other studies. ± - Standard deviation. <LD - Below limit method detection. Different letters represent statistically significant values (p<0.05).

Season	River	Site	Al (mg/g)	Ba (µg/g)	Cr (µg/g)	Cu (µg/g)	Fe (mg/g)	Mn (µg/g)	Ni (µg/g)	Pb (µg/g)	Ti (mg/g)	Zn (µg/g)
Dry	BND	S8	14.1 ± 0.5	84.8 ± 7.4	26.17 ± 0.79	1.16 ± 0.06	27.1 ± 1.7	221.9 ± 16.3	7.12 ± 0.28	12.0 ± 1.2	2.55 ± 0.3	15.8 ± 1.3
		S7	16.1 ± 1.7	179.1 ± 8.2	29.62 ± 1.33	3.56 ± 0.28	24.2 ± 0.4	128.5 ± 1.1	6.73 ± 0.73	11.0 ± 0.7	1.07 ± 0.0	14.5 ± 1.5
		S6	17.9 ± 1.8	256.1 ± 16.4	19.22 ± 1.59	2.80 ± 0.10	18.3 ± 2.4	138.8 ± 17.0	5.42 ± 0.74	12.4 ± 0.9	0.97 ± 0.1	10.1 ± 0.8
	BNE	S5	21.4 ± 2.4	334.4 ± 12.8	11.28 ± 0.61	1.29 ± 0.09	12.2 ± 0.4	215.4 ± 25.0	2.78 ± 0.15	15.8 ± 0.1	0.69 ± 0.0	5.7 ± 0.1
		S4	23.5 ± 0.6	106.9 ± 13.9	8.77 ± 0.17	2.97 ± 0.29	28.1 ± 0.2	236.6 ± 11.2	3.91 ± 0.26	14.5 ± 1.4	4.35 ± 0.0	9.9 ± 0.9
		S3	15.3 ± 2.0	312.3 ± 1.2	7.52 ± 0.23	1.57 ± 0.04	14.6 ± 0.4	117.0 ± 3.2	2.61 ± 0.07	15.4 ± 0.8	0.87 ± 0.0	8.1 ± 1.0
	IT	S2	13.0 ± 1.9	253.9 ± 8.0	3.99 ± 0.22	0.28 ± 0.03	10.2 ± 1.0	117.1 ± 10.4	1.71 ± 0.11	15.9 ± 0.1	0.63 ± 0.0	4.9 ± 0.4
		S1	14.4 ± 0.9	302.1 ± 2.4	3.75 ± 0.43	<0.02 ± <0.02	8.1 ± 0.8	85.0 ± 4.4	1.06 ± 0.02	10.4 ± 1.3	0.65 ± 0.1	3.3 ± 0.2
	Mean	BND	16.0 ± 1.9	173.3 ± 85.8	25.0 ± 5.30	2.5 ± 1.23	23.2 ± 4.5	163.1 ± 51.2	6.42 ± 0.89	11.8 ± 0.7	1.5 ± 0.9	13.5 ± 3.0
		BNE	20.1 ± 4.3	251.2 ± 125.5	9.2 ± 1.91	1.9 ± 0.90	18.3 ± 8.6	189.7 ± 63.8	3.10 ± 0.71	15.2 ± 0.7	2.0 ± 2.1	7.9 ± 2.1
		IT	13.7 ± 1.0	278.0 ± 34.0	3.9 ± 0.17	0.3 ± -	9.2 ± 1.5	101.1 ± 22.7	1.38 ± 0.47	13.1 ± 3.9	0.6 ± 0.0	4.1 ± 1.1
Rainy	BND	S8	20.7 ± 0.6	195.6 ± 10.6	37.38 ± 0.11	5.75 ± 0.60	17.2 ± 0.8	87.8 ± 0.1	7.70 ± 0.38	5.9 ± 0.5	1.1 ± 0.0	15.89 ± 0.16
		S7	14.9 ± 0.7	157.6 ± 1.2	29.56 ± 2.77	3.34 ± 0.23	15.3 ± 1.2	159.2 ± 13.5	7.24 ± 0.64	6.4 ± 0.6	1.3 ± 0.2	17.31 ± 1.48
		S6	12.9 ± 1.3	392.5 ± 22.0	31.34 ± 1.14	3.95 ± 0.32	14.6 ± 0.9	165.9 ± 6.9	6.44 ± 0.17	9.6 ± 0.1	0.9 ± 0.0	15.17 ± 0.50
	BNE	S5	21.9 ± 2.2	430.2 ± 39.6	20.81 ± 2.57	3.06 ± 0.36	10.7 ± 1.5	87.4 ± 8.3	3.97 ± 0.37	9.4 ± 0.3	0.4 ± 0.0	18.64 ± 0.28
		S4	21.5 ± 1.4	335.4 ± 11.9	5.75 ± 0.42	1.30 ± 0.06	5.8 ± 0.2	58.7 ± 2.9	3.08 ± 0.05	9.0 ± 1.1	0.6 ± 0.0	7.08 ± 0.53
		S3	34.7 ± 4.8	133.1 ± 16.9	45.97 ± 1.75	16.75 ± 0.51	61.9 ± 1.7	845.4 ± 78.6	17.13 ± 0.65	15.8 ± 2.1	7.2 ± 0.2	82.43 ± 2.46
	IT	S2	22.2 ± 0.0	548.8 ± 16.1	11.04 ± 0.28	1.38 ± 0.13	11.3 ± 1.1	151.3 ± 1.1	4.12 ± 0.56	11.2 ± 0.5	2.1 ± 0.1	12.17 ± 1.47
		S1	14.2 ± 0.7	540.4 ± 10.6	8.30 ± 0.53	1.19 ± 0.02	9.1 ± 1.0	95.7 ± 0.5	4.88 ± 0.59	10.7 ± 0.5	0.9 ± 0.0	9.63 ± 0.62
	Mean	BND	16.2 ± 4.1	248.6 ± 126.1	32.8 ± 4.10	4.3 ± 1.26	15.7 ± 1.4	150.4 ± 45	6.8 ± 0.79	9.6 ± 2.82	1.3 ± 0.61	14.8 ± 2.48
		BNE	26.0 ± 7.5	299.6 ± 151.8	24.2 ± 20.32	7.0 ± 8.46	26.2 ± 31.1	260.1 ± 295	5.6 ± 5.69	13.3 ± 3.22	2.4 ± 2.81	22.0 ± 29.97
		IT	18.2 ± 5.7	544.6 ± 5.9	9.7 ± 1.94	1.3 ± 0.13	10.2 ± 1.6	112.3 ± 29	2.9 ± 1.85	12.0 ± 2.58	1.1 ± 0.72	7.5 ± 4.10
		TEL			37.3	35.7			18	35.0		18.0
		PEL			90.0	197.0			36	91.3		36.0
		Fraction										
Comparison with literature	Tigris River, Turkey*	<2mm			119	1941		1233	216	393		530
	Tigris River, Turkey	<2mm			79	78		591	133	243		148
	Paraíba do Sul River, Brazil	<63 µm			91	73		933				222
	Macaé River, Brazil	<63 µm	52	138	66	44	48	445	24	45		130
	Pardo River, Brazil	-			19	11		254	5	7		26
	Sinos River, Brazil	-	12		23		24	620		13		29
Dry and Rainy	Mean	BND	16.1 ± 2.8 b	210.9 ± 104.9 a	28.9 ± 6.0 a	3.43 ± 1.5 ab	19.5 ± 5.1 a	150.4 ± 44.6 a	6.78 ± 0.8 a	9.6 ± 2.8 a	1.33 ± 0.6 a	14.8 ± 2.5 a
		BNE	23.1 ± 6.3 a	275.4 ± 127.3 a	16.7 ± 15.3 ab	4.49 ± 6.1 a	22.2 ± 20.8 a	260.1 ± 295.3 a	5.58 ± 5.7 ab	13.3 ± 3.2 a	2.36 ± 2.8 a	22.0 ± 30.0 a
		IT	16.0 ± 4.2 b	411.3 ± 155.2 a	6.8 ± 3.5 b	0.95 ± 0.6 b	9.7 ± 1.4 a	112.3 ± 29.2 a	2.94 ± 1.8 b	12.0 ± 2.6 a	1.08 ± 0.7 a	7.5 ± 4.1 a

Tigris River, Turkey: Varol (2011) (* means of a site affected by a copper mine); Paraíba do Sul River, Brazil: Molisani *et al.,* (1999); Macaé River, Brazil: Molisani *et al.,* (2015); Pardo River, Brazil: Alves *et al.,* (2014); Sinos River, Brazil: Weber *et al.,* (2013).

rainy season, Cr values at Site 6 (BND) and Site 3 (BNE) exceeded TEL, while Zn exceed TEL at Site 5 (BNE) and PEL at Site 3 (BNE) (Table 6), indicating their harmful effects to organisms, as described by the USA National Oceanic and Atmospheric Administration (NOAA).

The higher metal concentrations during rainy season are due to superficial runoff, which carries soil particles to aquatic bodies. The higher levels observed in BNE might be associated with the high elevation areas of coffee plantations and other agricultural cultures, in addition to the higher percentage of bare soil and the lower size of forest fragments that contribute to this process (Figure 2). In addition, taking into consideration that the IT river receives water from the BND and BNE and has an independent sub-basin, it might be expected that its concentrations would be higher. However, the BNE and BND tributaries have dams that cause sedimentation of suspended particles and consequently reduce the transport of metals (Figure 1A) (MAGILLIGAN; NISLOW, 2005). This effect can also be observed through the grain sediment size of the IT river, which is composed of higher percentages of sand particles (Table 3). The BNE and IT rivers showed the same order of the elements in the sediment with Al > Fe > Ti > Ba > Mn > Pb > Zn > Cr > Ni > Cu, while in the BND some differences were observed, especially for Fe, Cr, Zn and Pb, with Fe > Al > Ti > Ba > Mn > Cr > Zn > Pb > Ni > Cu (Table 6).

Metal concentrations in surface sediments of the three sub-basins of this study (fraction <2mm) were similar to the levels of other rivers from agricultural areas such as ***Sinos***, ***Pardo,*** and ***Macaé*** (fraction <63µm) in Brazil, even analyzing the silt/clay and sand fractions. The BNE, BND, and IT also showed lower metal levels in comparison with the ***Paraíba do Sul*** River (fraction <63µm), considered a highly affected sub-basin due to the influx of untreated sewage, agricultural land use, and significant urbanized and industrial areas. Lower levels than in the present study were also observed in the Tigris River, a highly impacted river in Turkey that receives wastewater discharge from a copper mine. Due to this point source, the highest difference between the values for all elements was observed in comparing those of the present study with those of the Tigris River. These comparisons show that the sub-basins evaluated in this study have undergone the same degree of impact as other rivers in agricultural areas, but less impact than the Tigris River, with its point mining activity (Table 6).

The metal concentrations in sediments corresponded with soil levels (Figure 3). For the BND and IT, the sediment concentrations are generally lower than soil, while in the BNE, metal concentrations in the sediments are almost the same as those observed for soil. The differences in land use might be influencing this condition, since in BNE the higher superficial runoff, together with the use fertilizers and pesticides in coffee plantations and the lack of sewage treatment, the metals are carried directly to water, where they can accumulate in fish and sediments. In the BND sub-basin, however, large areas of native forest retain soil particles (Figure 1B) and the flat relief of the IT sub-basin region may reduce soil contributions and sediment concentrations (Figure 1C).

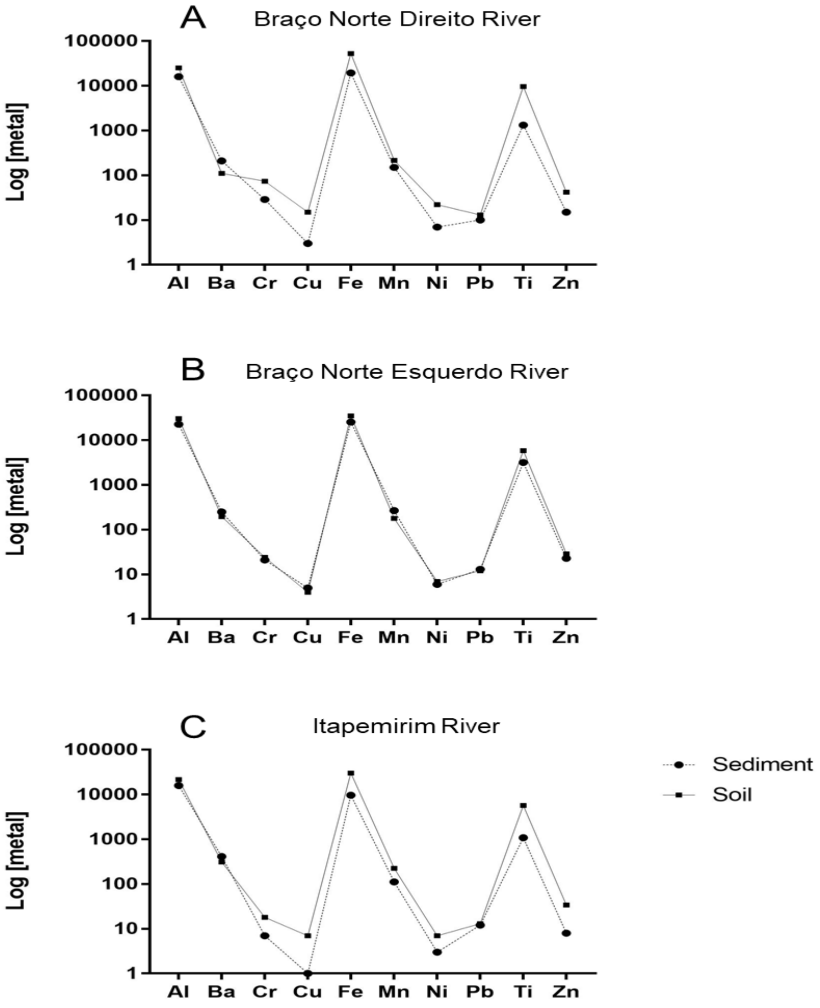

Figure 3: Logarithmical scale of metal concentrations in soil and sediment of the rivers. A – Braço Norte Direito River; B – Braço Norte Esquerdo River; C – Itapemirm River.

There is no published data of the metal concentrations in any of the investigated matrices in the present study area (BNE, BND, and IT). Therefore, the present data are important as a first reference for a relevant area involved in coffee production in Brazil. From this perspective, geochemical analysis such as Contamination Factor (CF), Pollution Load Index (PLI), and Enrichment Factor (EF) contribute to the understanding of the differences among the three sub-basins.

According to the Contamination Factor (CF) analysis, all elements presented CF values above 1, indicating moderate contamination of sediments in the three sub-basins. The BNE showed the higher CF values for almost all elements (with the exception of Cu, Ni, and Pb). The BNE's higher CF value is mainly influenced by S3. At this site some elements (Ni and Cr) had CF values from 3 to 6 or even above 6 (Fe, Cu, Mn, Ti, and Zn), indicating moderate and very high contamination, respectively (Table 7).

The Pollution Load Index (PLI) from the three sub-basins was above 1, indicating a degree of metal pollution. The BNE showed the highest PLI mean (followed by IT and BND), demonstrating a higher contamination potential. As with the CF, S3 in the BNE had the highest PLI values (4.43) in comparison with other sampling sites (Table 7).

Table 7: Metal contamination factors (CFs) and pollution load indices (PLIs) for sediments of the Braço Norte Direito (BND), Braço Norte Esquerdo (BNE), and Itapemirim (IT) Rivers and comparisons with other studies.

River	Site	Contamination factors (CFs)										PLI
		Al	**Ba**	**Cr**	**Cu**	**Fe**	**Mn**	**Ni**	**Pb**	**Ti**	**Zn**	
BND	S8	1.35	1.65	1.65	2.99	1.52	1.76	1.37	1.51	1.95	1.57	1.69
	S7	1.20	1.98	1.54	2.98	1.35	1.64	1.29	1.46	1.27	1.57	1.57
	S6	1.19	3.82	1.32	2.91	1.13	1.74	1.09	1.85	1.01	1.25	1.56
BNE	S5	1.42	3.58	2.79	1.69	1.97	2.58	1.30	1.40	1.29	2.12	1.90
	S4	1.47	2.07	1.26	1.66	2.92	2.51	1.34	1.30	5.67	1.48	1.92
	S3	1.63	2.08	4.65	7.13	6.57	8.19	3.79	1.73	9.30	7.89	4.43
IT	S2	1.35	1.58	2.00	2.99	1.33	1.58	2.76	1.31	2.21	2.54	1.88
	S1	1.09	1.66	1.61	4.30	1.06	1.06	2.81	1.02	1.24	1.94	1.58
Mean	BND	1.25	2.49	1.50	2.96	1.33	1.71	1.25	1.61	1.41	1.46	1.61
	BNE	1.51	2.58	2.90	3.49	3.82	4.43	2.14	1.48	5.42	3.83	2.75
	IT	1.22	1.62	1.81	3.64	1.20	1.32	2.79	1.16	1.73	2.24	1.73
Comparison with literature	Tigris River, Turkey*			1.86	34.68	1.11	2.37	2.93	5.79		6.80	4.19
	Tigris River, Turkey			1.23	1.40	1.00	1.13	1.80	3.58		1.90	1.49
	Macaé River, Brazil	1.00	0.21	0.25	1.22	1.23	0.58	0.71	2.65		0.83	0.75

Tigris River, Turkey: Varol (2011) (* means of a site affected by a copper mine); Macaé River, Brazil: Molisani *et al.,* (2015).

The enrichment factor showed a pattern similar to that of CF values (Table 8). Higher enrichment factor values for most of elements were found in the BNE, followed by the IT and BND. Cu was the most enriched metal in the three sub-basins, indicating anthropogenic influences that may be mainly due to agricultural activities, since this catchment has a low human density and hence lower sewage production (49 inhabitants per km^2) (IBGE, 2017). However, we cannot ignore the contribution of Cu from domestic sewage since sewage is recognized as an important source of this element in aquatic ecosystems, especially in developing countries (SODRÉ *et al.*, 2012). In this case, most of the cities in the study area do not have a domestic sewage treatment system, and sewage is released directly into river water, thus representing a possible source of metals and other contaminants. As observed for the CF and PLI, S3 showed the highest EF values for almost all elements (except for Ba) (Table 8).

The higher metal concentrations in the sediment reflected in the CF, PLI, and EF values for S3 are related to this site's proximity to a dam (Figure 1A). This result is consistent with the increased presence of fine particles (silt and clay), indicated by granulometric analysis (Table 3), because damming reduces river flow and intensifies the sedimentation process (MAGILLIGAN; NISLOW, 2005).

Table 8: Enrichment factors (EF) for sediments of the Braço Norte Direito (BND), Braço Norte Esquerdo (BNE), and Itapemirim (IT) Rivers and comparisons with other studies

River	Site	Enrichment factor (EF)									
		Al	**Ba**	**Cr**	**Cu**	**Fe**	**Mn**	**Ni**	**Pb**	**Ti**	**Zn**
BND	S8	1.00	1.23	1.23	2.21	1.12	1.31	1.01	1.12	1.44	1.16
	S7	1.00	1.66	1.28	2.49	1.13	1.37	1.08	1.22	1.06	1.31
	S6	1.00	3.21	1.10	2.45	0.95	1.46	0.92	1.56	0.85	1.05
BNE	S5	1.00	2.53	1.97	1.19	1.39	1.82	0.91	0.99	0.91	1.50
	S4	1.00	1.41	0.86	1.13	1.98	1.71	0.91	0.89	3.85	1.00
	S3	1.00	1.28	2.85	4.36	4.02	5.02	2.32	1.06	5.70	4.83
IT	S2	1.00	1.17	1.48	2.21	0.98	1.17	2.05	0.97	1.64	1.88
	S1	1.00	1.52	1.47	3.93	0.97	0.97	2.57	0.93	1.14	1.77
Mean	BND	1.00	2.03	1.20	2.38	1.07	1.38	1.00	1.30	1.12	1.18
	BNE	1.00	1.74	1.89	2.23	2.46	2.85	1.38	0.98	3.49	2.44
	IT	1.00	1.34	1.48	3.07	0.98	1.07	2.31	0.95	1.39	1.83
Comparison with literature	Tigris River, Turkey*			1.68	31.34	1.00	2.14	2.65	5.23		6.15
	Tigris River, Turkey			1.21	1.38	1.00	1.12	1.77	3.53		1.89
	Macaé River, Brazil	1.00	0.21	0.25	1.22	1.23	0.58	0.71	2.65		0.83

Tigris River, Turkey: Varol (2011) (* means of a site affected by a copper mine); Macaé River, Brazil: Molisani *et al.,* (2015).

The comparison of the present study's CF and PLI values with those of two other rivers renders interesting results (Tables 7 and 8). BND, BNE, and IT sub-basins presented higher CF, PLI and EF values than the ***Macaé*** River in Brazil, which is mainly affected by agricultural activities and urbanization, with exception of Pb. Similar or even higher CF and EF values for Cr, Fe, and Mn were observed in the present study for the BNE river, in comparison with the Tigris River (at a site affected by a copper mine), while lower values were found for Cu, Ni, Pb, and Zn. However, the PLI values of Site 3 in this study were higher than those of a contaminated site on the Tigris River due to the major number of elements with higher CF values. This comparison demonstrates that although the sub-basins evaluated presented lower metal concentrations in sediment than the ***Macaé*** River (Table 6), they may have undergone the most extensive impact. Moreover, the higher PLI values of Site 3, which surpassed those of a highly contaminated river site on the Turkish river, arouse concern. However, one must emphasize that while Molisani *et al.* (2015) used background values from rock mineralogy and analyzed a fraction of <63µm, while in this study the lower concentrations found in each sub-basin were utilized.

Multivariate cluster analysis (CA) was applied to group the sampling sites according to their similarity with respect to metal spatial variability (Figure 4). The eight sampling sites formed three clusters; the first cluster was composed of S8, S7, and S6 from the BND river, the second cluster of the sites S4, S5, S2, and S1 from the BNE and IT rivers, and the third cluster composed of S3. This analysis indicates that the metal variation is similar in the BNE and IT, while the BND shows behavior different from the other two sub-basins. This result can be related to the differences in the land use between the studied areas, since the BNE and IT show higher CF, PLI, and EF values and the same order of element concentrations in soil and sediment, possibly the result of similar associations between the metals. S3 represents an isolated cluster due to the disparity of the metal levels in comparison with other sampling sites in the BNE, a fact that can be attributed to the nearby dam.

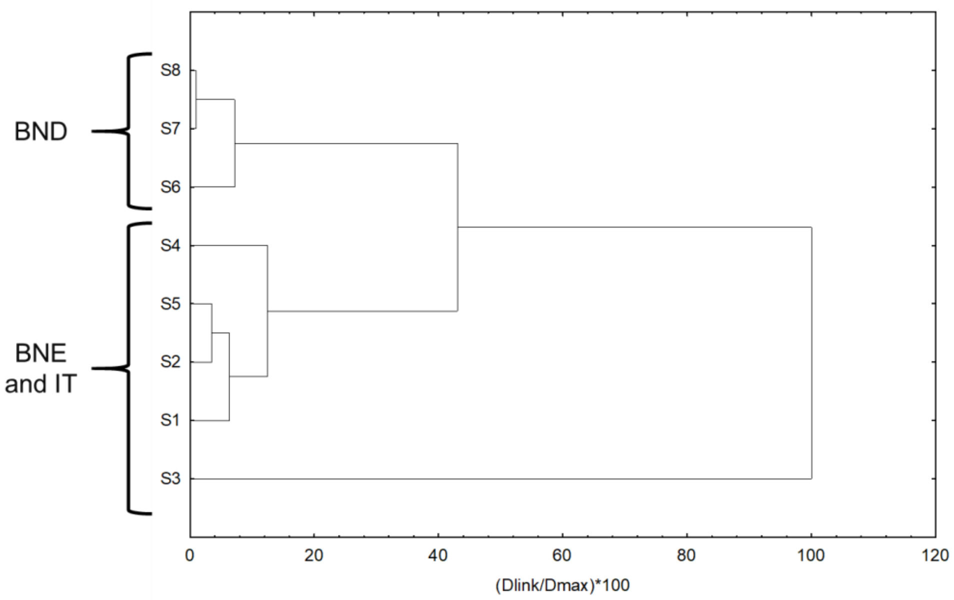

Figure 4: Dendrogram of multivariate cluster analysis of the sampling sites (1 to 8) of the Braço Norte Direito (BND), Braço Norte Esquerdo (BNE), and Itapemirim (IT) Rivers.

PCA was performed to compare the compositional pattern of sediment samples. The entire dataset revealed two main components that explained 77.1% of the total variance. The first principal component (Axis I) accounted for 58.5% of the total variance and was correlated (loading > 0.60) with Ba and sand fractions that indicate that Ba integrate the mineralogy of these particles. The second principal component (Axis II) accounted for 18.6% of the total variance and was correlated with Cr (loading > 0.60). The lower association of Cr, Ni, Cu, and Zn with the Al, Fe, Mn, Ti, and organic carbon indicate the influence of land use on these elements. In fact, the CF and EF values point to some contamination and enrichment in the different sub-basins (Figure 5A, Tables 7 and 8).

Case analysis was used to show the distribution of the sampling sites between the seasons (Figure 5B), indicating that Component 2 (Cr, Ni, Cu, and Zn) was associated with most of sampling sites in the rainy season and implicating superficial runoff as a source of these metals. Pb was the only element with a lower association to all others and the sand fraction, but also was negatively associated with Component 2. Thus, this element seems to originate from natural sources, a hypothesis borne out by its EF values, which show no enrichment in the BNE and IT.

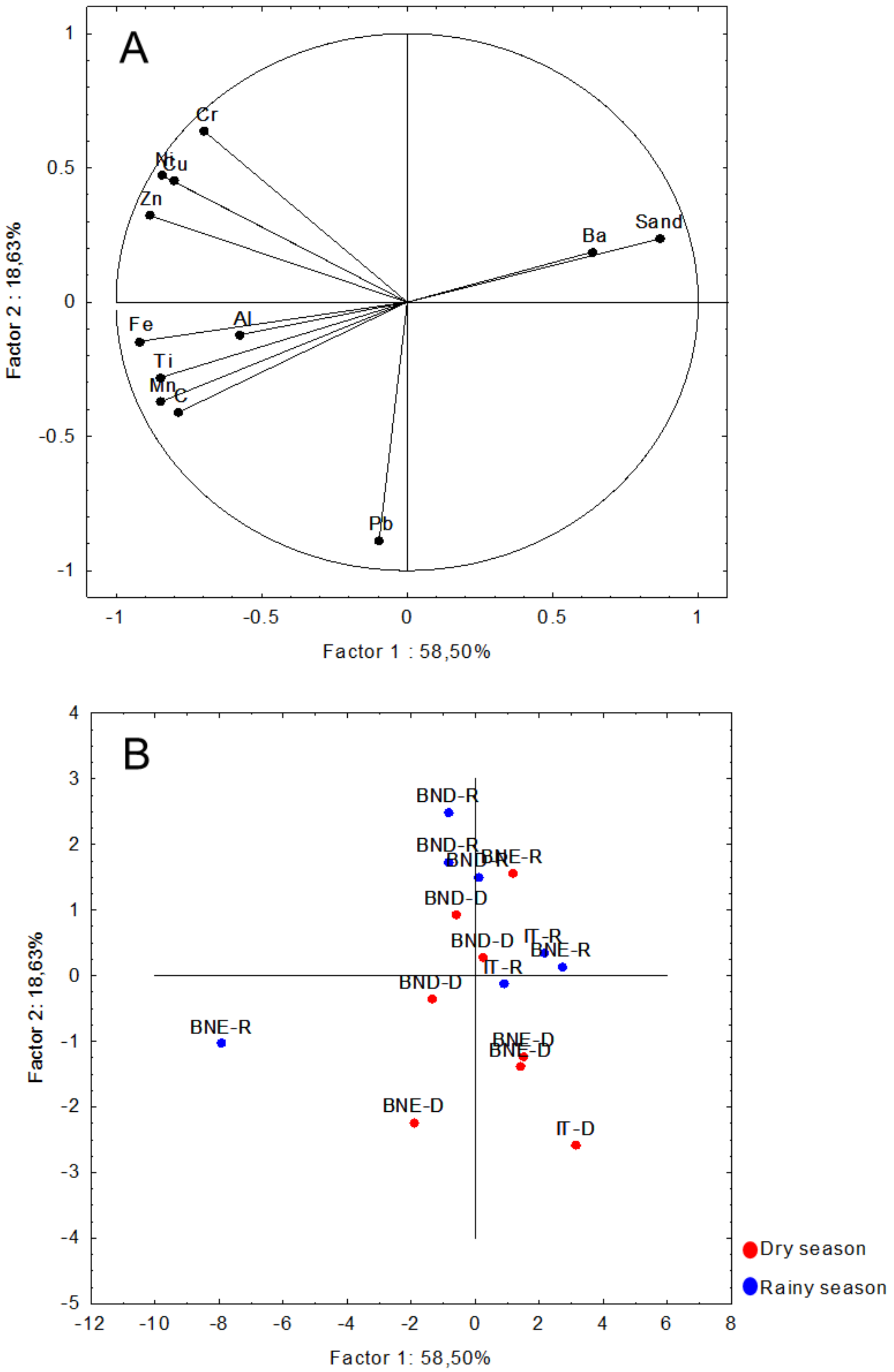

Figure 5: Principal component analysis of the parameters analyzed in the sediments of the Braço Norte Direito (BND), Braço Norte Esquerdo (BNE), and Itapemirim (IT) Rivers in dry and rainy seasons. A. Association between the elements in the three rivers. B. Sampling site distribution in the three rivers between the seasons. D – Dry season; R- Rainy season.

Metal concentrations in fish (*Geophagus brasiliensis*) also indicate the differences between the sub-basins (Table 9). In total, 102 fishes were collected, with total length ranging from 5.9 to 20 cm and weight from 5.35 to 132 g. Mean values were of 11.3 and 29.8, respectively. The order of metal concentrations in BND was Zn > Al > Mn > Fe > Ba > Ti > Cu > Cr, in BNE Mn > Al > Zn > Fe > Ba > Ti > Cu > Cr, and in IT Mn > Zn > Ba > Al > Fe > Cu > Cr > Ti. Ni and Pb were below the detection limit (Table 8). The BNE river showed the highest values of Al, Fe, and Mn, the BND for Cr and Zn, and the IT for Ba. These results reflected the metal concentrations observed in water. Fish from the BND and BNE showed the same mean values for Cu, which were higher than IT; Ti likewise was only detected in these two rivers (BND and BNE). The higher Cr values in the BND also reflect the soil and water concentrations in this sub-basin. Some of these concentrations were statistically different between the sub-basins (Table 9).

Table 9: Metal concentrations (µg/g dry weight) in Geophagus brasiliensis (n=102) of the Braço Norte Direito (BND), Braço Norte Esquerdo (BNE), and Itapemirim (IT) Rivers and comparisons with other studies. ± - Standard deviation. <LD - Below limit method detection. Different letters represent statistically significant values (p<0.05).

	River	Al	Ba	Cr	Cu
	BND	7.3 ± 14.1 a	3.9 ± 2.0 b	0.17 ± 0.67 a	0.24 ± 0.12 a
	BNE	10.3 ± 10.7 a	4.6 ± 2.2 a	0.09 ± 0.03 a	0.24 ± 0.12 a
	IT	4.0 ± 3.7 a	5.8 ± 2.1 a	0.08 ± 0.02 a	0.15 ± 0.04 b
Comparison with literature	Piracicaba River, Brazil	5.0		0.23	0.19
	Alagados Reservoir, Brazil	839.8		4.41	27.55
	Paraíba do Sul River, Brazil			3.35	5.70

	River	Fe	Mn	Ti	Zn
	BND	5.6 ± 12.6 a	6.1 ± 4.2 b	0.43 ± 0.72 a	9.5 ± 5.9 a
	BNE	6.5 ± 8.1 a	10.5 ± 4.5 a	0.42 ± 0.46 a	9.1 ± 4.5 a
	IT	3.6 ± 3.0 a	9.9 ± 5.4 b	<0.002 ± <0.002 -	8.2 ± 2.0 a
Comparison with literature	Piracicaba River, Brazil		0.4		8.4
	Alagados Reservoir, Brazil	20.4	6.8		23.18
	Paraíba do Sul River, Brazil				380

Piracicaba River, Brazil: Meche *et al.* (2010); Paraíba do Sul River, Brazil: Terra *et al.* (2008); Alagados Reservoir, Brazil: Voigt *et al.* (2015).

When comparing the fish metal concentrations from the present study with the literature, lower levels were found in comparison with other studies, as observed for water and soil. With exception of Mn, the concentrations found in this study were close to levels found in fish from a rural section of the ***Piracicaba*** River. The metal concentrations in the ***Alagados*** reservoir were higher than those found in this study. This reservoir is affected by agricultural and cattle breeding activities and the unregulated occupation of the reservoir margins (CLEMENTE *et al.*, 2010). The ***Paraíba do Sul*** River presents higher concentrations of metals in the muscle of *G. brasiliensis,* the same fish species used in the present study, with Cu and Zn levels at least 38 and 46 times higher than found in this study, respectively. Different land use features such as agriculture, the presence of urban and industrial areas, and sewage discharge can explain the highest concentrations in ***Paraiba do Sul*** River (MOLISANI, *et al.*, 1999; TERRA *et al.*, 2008). Therefore, *G. brasiliensis,* which is a Cichlid species that feeds near the bottom, may represent a good environmental monitor, since they reflect the geochemical characteristics of the environment (Table 8).

4. Conclusion

The results of the present study illustrate the influence of land use on metal concentrations in different matrices (water, sediment, and fish) in three sub-basins in the southeast of Brazil. The largest areas of agricultural activities, cities, and bare soil in the BNE may be responsible for its higher metal concentrations, in conjunction with more elevated PLI and EF values, mostly due to the different kinds of land use in this sub-basin. The conversion of land use in the studied area was the main factor responsible for the contamination of Cr, Cu, Ni, and Zn and the presence of some metal concentrations above the TEL and PEL raise concerns about possible harmful effects on organisms.

The present study is the first report of metal concentrations in different environmental matrices in an important agricultural region of Brazil. While the metal concentrations in the different environment matrices demonstrated in the present study do not indicate higher levels of contamination, they raise concerns about the increased use of pesticides and fertilizers and the need for sewage treatment.

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