

## Assessment of disturbance at three spatial scales in two large tropical reservoirs

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### ABSTRACT

Large reservoirs are an increasingly common feature across tropical landscapes because of their importance for water supply, flood control and hydropower, but their ecological conditions are infrequently evaluated. Our objective was to assess the range of disturbances for two large tropical reservoirs and their influences on benthic macroinvertebrates. We tested three hypotheses: i) a wide variation in the level of environmental disturbance can be observed among sites in the reservoirs; ii) the two reservoirs would exhibit a different degree of disturbance level; and iii) the magnitude of disturbance would influence the structure and composition of benthic assemblages. For each reservoir, we assessed land use (macroscale), physical habitat structure (mesoscale), and water quality (microscale). We sampled 40 sites in the littoral zones of both Três Marias and São Simão Reservoirs (Minas Gerais, Brazil). At the macroscale, we measured cover percentages of land use categories in buffer areas at each site, where each buffer was a circular arc of 250 m. At the mesoscale, we assessed the presence of human disturbances in the riparian and drawdown zones at the local (site) scale. At the microscale, we assessed water quality at each macroinvertebrate sampling station using the Micro Disturbance Index (MDI). To evaluate anthropogenic disturbance of each site, we calculated an integrated disturbance index (IDI) from a buffer disturbance index (BDI) and a local disturbance index (LDI). For each site, we calculated richness and abundance of benthic macroinvertebrates, Chironomidae genera richness, abundance and percent Chironomidae individuals, abundance and percent EPT individuals, richness and percent EPT taxa, abundance and percent resistant individuals, and abundance and percent non-native individuals. We also evaluated the influence of disturbance on benthic macroinvertebrate assemblages at the entire-reservoir scale. The BDI, LDI and IDI had significantly greater average scores at São Simão than at Três Marias Reservoir. The significantly greater differences in IDI scores for São Simão Reservoir were reflected in 10 of the 13 Ekman-Birge dredge biotic metrics and in 5 of 13 of the kick-net biotic metrics. We also observed clear ranges of disturbances within both reservoirs at macro (BDI) and mesoscales (LDI) and in water quality, but an insignificant range in MDI results. However, we found no significant relationship between the benthic macroinvertebrate metrics and the BDI, LDI, and IDI among sites within a single reservoir. Hence, we believe that benthic macroinvertebrate distributions in those reservoirs were influenced by other factors or that reservoir macroinvertebrates (dominated by chironomids) were poor indicators of disturbance at the site scale.

**Key words:** Environmental quality; benthic macroinvertebrates; bioindicators; physical habitat.

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### INTRODUCTION

Reservoirs are complex artificial aquatic ecosystems with multiple functions, such as electricity generation, water supply, navigation, flood control, and recreation (Prado, 2002; Tundisi, 2006). However, reservoirs reduce river and floodplain connectivity (Tiemann, *et al.*, 2004) and water level fluctuations that are common in reservoirs erode shorelines and simplify littoral habitats (Miranda *et al.*, 2010). Furthermore, lower water levels and erosion caused by altered riparian and littoral vegetation reduce the availability of habitat elements (*e.g.*, woody material,

macrophytes) (Karr, 2006; Gaeta *et al.*, 2014). All these disorders eventually alter the composition, abundance, and diversity of aquatic biota, in the reservoirs and upstream and downstream of the dams. Therefore, it is important to develop integrated management of the multiple uses of reservoirs and to limit watershed disturbances that lead to such alterations.

To facilitate more complete characterization of ecological conditions, assessments of environmental quality are increasingly based on a systematic approach, integrating physical, chemical, and biological aspects (Barbour *et al.*, 1999; Callisto *et al.*, 2004). To characterize the environ-

mental quality of lacustrine ecosystems, an assessment of disturbance gradients is usually carried out based on the analysis of a single reservoir (Petesse *et al.*, 2007, Terra and Araújo, 2011) or a comparison of different reservoirs (Molozzi *et al.*, 2012, 2013a, Petesse *et al.*, 2014).

Human activities at the landscape level are major threats to the ecological integrity of aquatic ecosystems, because they alter water quality and biota in complex and synergistic ways (Allan, 2004; Macedo *et al.*, 2014a). The replacement of non-disturbed landscapes with new human-altered landscapes has altered ecosystems globally (Meyer and Turner, 1994; Vorosmarty *et al.*, 2010). Hence, quantifications of land use are valuable indicators of environmental conditions.

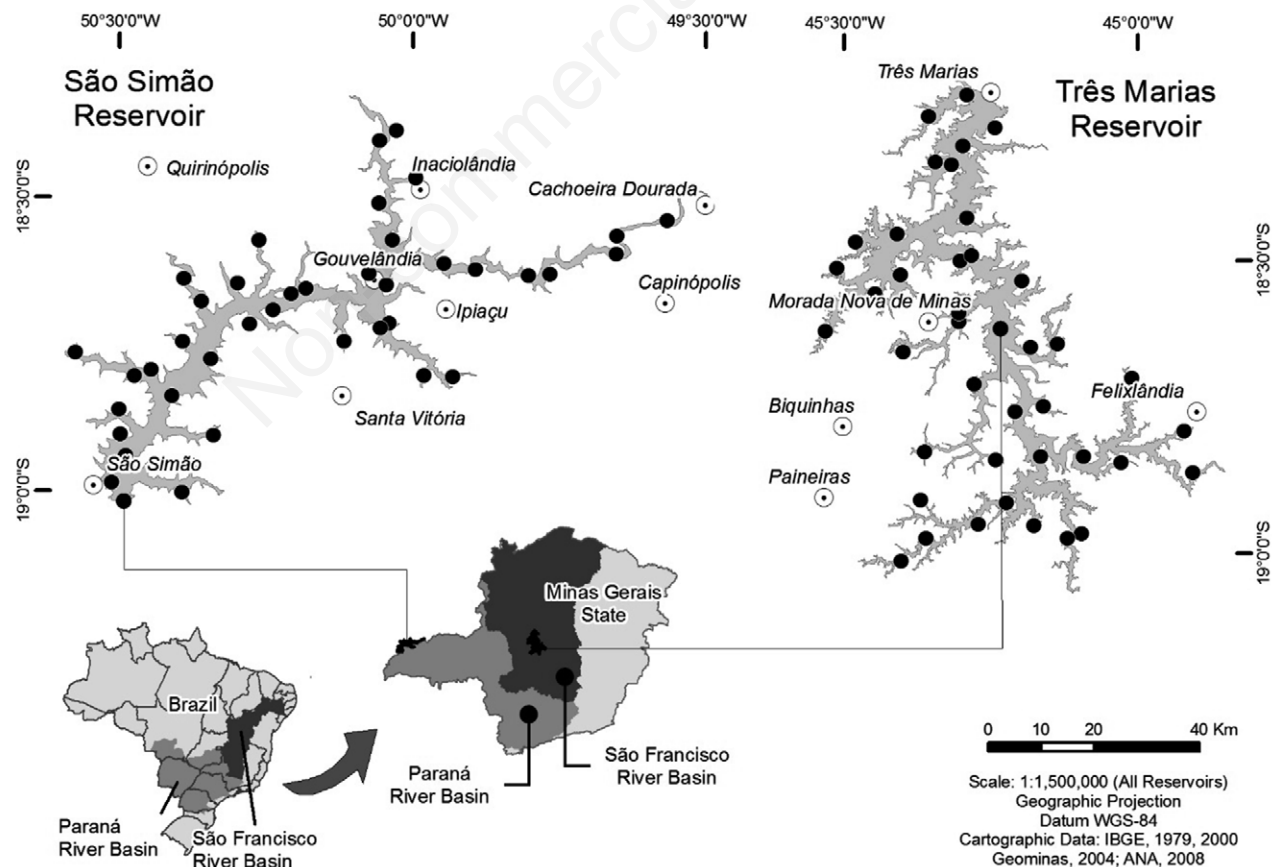
We evaluated the environmental quality of tropical reservoirs (Três Marias and São Simão, Southeast Brazil) using an integrative approach that considered land use, physical habitat structure, water quality, and biological parameters. We determined and compared disturbances levels for these two reservoirs and assessed the effects of environmental stressors on benthic macroinvertebrate assem-

blages at three spatial scales: buffer land use (macroscale), physical habitat transect structure (mesoscale), and sampling station water quality (microscale). We hypothesized that: i) a wide variation in the level of environmental disturbance can be observed among sites in the reservoirs; ii) each reservoir would exhibit a different degree of disturbance level; and iii) the magnitude of disturbance would influence the structure and composition of benthic macroinvertebrate assemblages.

## METHODS

### Study areas

Both reservoirs are among the largest in Brazil. Três Marias Reservoir is located in the upper São Francisco River Basin (Fig. 1). It has a surface area of 1040 km<sup>2</sup>, a total volume of 21 billion m<sup>3</sup>, and a maximum depth of 58.5 m (CEMIG, 2012). Its waters come mainly from the São Francisco River and its tributaries (Godinho and Godinho, 2003). It began operation in 1962 with the purposes



**Fig. 1.** Três Marias and São Simão Reservoirs depicting the 40 sample sites in each, and their locations in the Upper São Francisco and Paranaíba River Basins, in Minas Gerais, Brazil.

of flood control, improving navigation, encouraging industrial development and irrigation, and providing hydropower (Freitas and Filho, 2004).

São Simão Reservoir has a surface area of 722 km<sup>2</sup>, a total volume of 11 billion m<sup>3</sup>, and a maximum depth of 126 m. It is formed by the regulation of the Paranaíba River and its tributaries (Pinto-Coelho, 2013) and began operating in 1978 to provide hydropower (Souza and Souza, 2009). Watershed land uses of the reservoirs are mostly agricultural (São Simão ~70%; Três Marias ~20%) and pasture (Três Marias ~40%; São Simão ~20%) (Macedo *et al.*, 2014b).

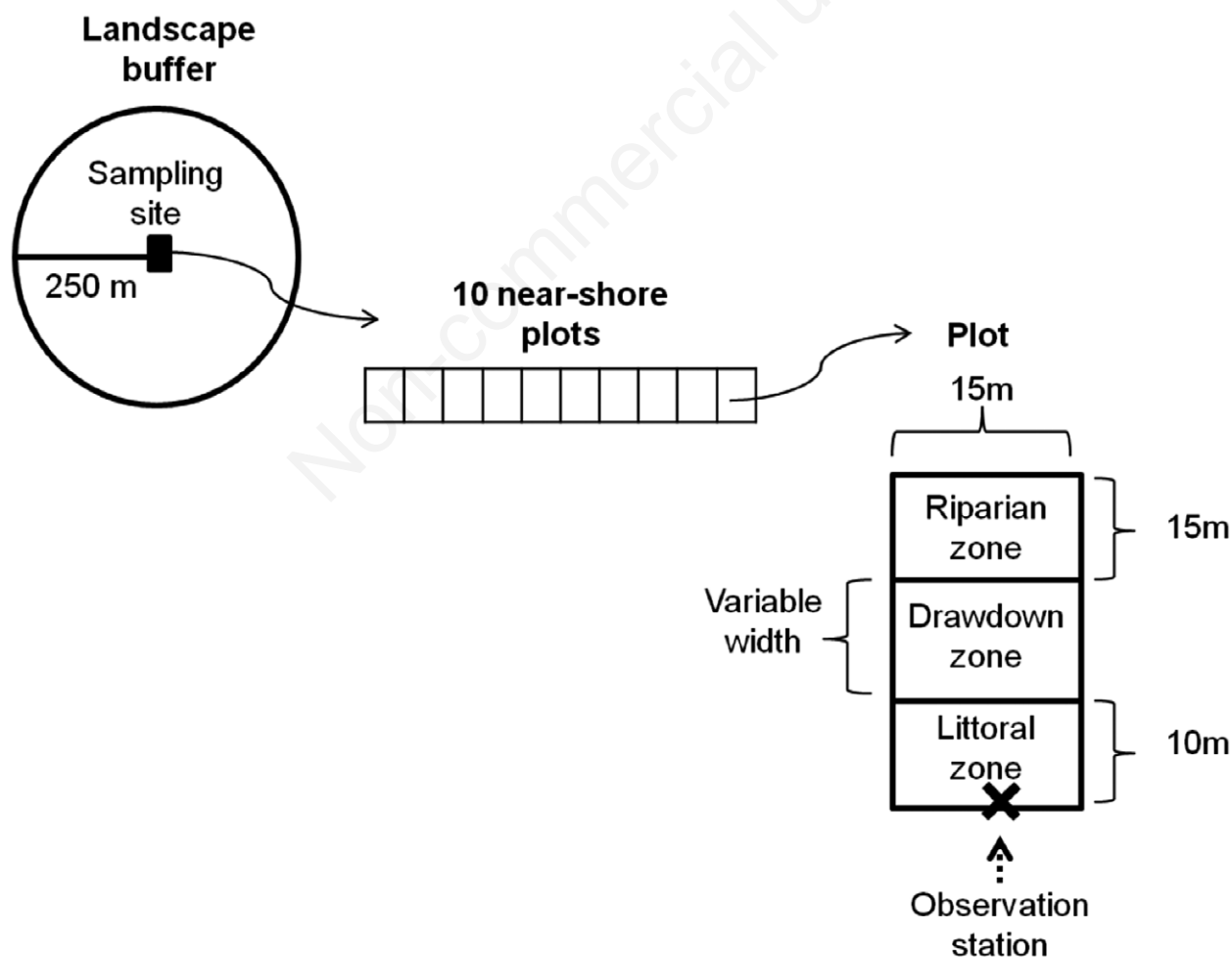
#### Sampling site selection and definition of spatial scales

All field data were collected in the littoral zone and adjacent shoreline riparian areas of each reservoir. We followed USEPA (2011) to select sites. Based on random selection of an initial sampling site, the subsequent 39 other

sites were marked systematically along a map margin at intervals of 55.715 km (Três Marias) and 30.508 km (São Simão). We sampled in the littoral zone of the reservoirs at the end of the rainy season in April 2011 at Três Marias and April 2013 at São Simão, assuming that during this month water levels, habitat complexity and taxa diversity would be highest.

Following the definition of Frissell *et al.* (1986) and Allan (1995), we initially defined the parameters that would comprise each spatial scale. The water variables at the kick net and grab sampling stations were assumed to be the smallest scale environmental factors to influence macroinvertebrates (microscale). We considered buffer land use as the largest scale environmental factors to influence macroinvertebrates (macroscale), and site physical habitat structure as an intermediate or mesoscale influence (see below).

Land use was assessed in 250 m wide buffers at each site (Fig. 2). Each buffer was determined by a 500-m di-



**Fig. 2.** Field sampling design with 10 near-shore stations at which data were collected to characterize physical habitat (riparian, littoral and drawdown zones) in Três Marias and São Simão Reservoirs. Adapted from USEPA (2011).<sup>2</sup>

ameter circle centered on the site. The buffers were delimited on an image obtained through a TM sensor on-board the Landsat 5 satellite, taken during the sampling periods. Inside each buffer, polygons for the definition and quantification of cover categories were delimited and classified. The Kosmo 2.0 program (Open Geographic Information System) was used to define the patterns observed in each polygon. Images obtained from Google Earth 6.0 (Google Corporation) were employed as ancillary data in this assessment.

### Field data collection methods

We used the USEPA (2011) protocol to characterize physical habitat structure. We applied the protocol along 150 m of the margin, in 10 consecutive transects at each of the 40 sites, producing a total of 400 samples for each reservoir. Each site was composed of continuous sections of littoral (15 x 10 m), riparian (15 x 15 m), and drawdown zones (15 x Y m), where Y represents the length of the drawdown zone, which varied among transects because of differing margin slopes at the habitat transects. The drawdown zone extended a variable distance landward, depending on the amount of reservoir level drop compared with typical high water levels (modified from USEPA, 2011). At each transect, human impacts in the riparian zone were recorded. Subsequently, these data were used to calculate an index of the near-shore anthropogenic disturbance intensity as described by Kaufmann *et al.* (2014c). At each macroinvertebrate sampling station, we collected a water sample in the littoral zone. Water samples were taken at an average depth of  $1.6 \pm 1.2$  m and  $17.0 \pm 17.3$  m off shore in Três Marias. The sampling average depth in São Simão was  $1.4 \pm 1.1$  m, with an average distance of the drawdown zone of  $24.7 \pm 14.3$  m. We measured water temperature ( $^{\circ}\text{C}$ ), pH, electrical conductivity ( $\mu\text{S cm}^{-1}$ ), and total dissolved solids ( $\text{g L}^{-1}$ ) *in situ* with a multiparameter Yellow Spring, model YSI 6600 meter. We determined total depth with a hand-held SONAR unit, euphotic zone depth with a Secchi disk, and turbidity (NTU) with a turbidimeter (Digimed - model DM-TU). We measured pheophytin ( $\mu\text{g L}^{-1}$ ) following Lorenzen (1967), chlorophyll-a ( $\mu\text{g L}^{-1}$ ) and total nitrogen ( $\text{mg L}^{-1}$ ) following Golterman *et al.* (1978), and total phosphorus ( $\text{mg L}^{-1}$ ) and orthophosphate ( $\text{mg L}^{-1}$ ) according to Mackereith *et al.* (1978) and APHA (1998), respectively.

We collected benthic macroinvertebrates in the reservoir littoral zone through use of both an Ekman-Birge grab ( $0.02 \text{ m}^2$ ) and a kick-net sampler following USEPA (2011). Kick net samples were taken in the wetted margin and grab samples were taken at the same depths at which the water samples were taken. At each sampling site we collected one dredge sample and one with kick-net sample, totaling 40 samples for each sampler per reservoir. For kick-net samples, we set a sampling area of approxi-

mately  $0.06 \text{ m}^2$ , in which the sediment was disturbed by hand or foot and directed to the net. This sampling method was applied in shallower regions, close to the margins of the reservoirs, where there is greater diversity of substrates and higher density of flooded vegetation.

### Laboratory methods

We washed the macroinvertebrate samples through 1.0 and 0.5 mm mesh sieves. We identified benthic macroinvertebrates to family using several keys (Peterson, 1960; Boffi, 1979; Pérez, 1988; Merritt and Cummins, 1996; Carvalho and Calil, 2000; Fernández and Domínguez, 2001; Costa *et al.*, 2006; Mugnai *et al.*, 2010). Chironomidae were identified to genus according to Trivinho-Strixino (2011). For each site, we calculated richness and abundance of benthic macroinvertebrates, richness and abundance of Chironomidae, percent of Chironomidae individuals, abundance and percent of EPT (Ephemeroptera, Plecoptera, Trichoptera) individuals, richness and percent of EPT taxa, and abundance and percent of resistant individuals according to Merritt and Cummins, 1996; Moreno and Callisto, 2004; Morais *et al.*, 2014 (molluscs, chironomids, oligochaetes), and abundance and percent of non-native individuals (*Melanoides tuberculatus*, *Corbicula fluminea*, *Macrobrachium amazonicus*).

### Disturbance indices

We constructed three disturbance indices to evaluate anthropogenic impacts on the reservoirs at macro, meso and microscales after Ligeiro *et al.* (2013). We considered the impact of land use as a macroscale disturbance (BDI - Buffer Disturbance Index), site physical habitat disturbance as a mesoscale disturbance (LDI - Local Disturbance Index), and the chlorophyll-a and phosphorus concentration at the macroinvertebrate sampling stations as a microscale disturbance (MDI - Micro Disturbance Index). For calculating the BDI, the land use categories received a weight related to the intensity of disturbance (Ligeiro *et al.*, 2013):

$$\begin{aligned} \text{BDI (Buffer Disturbance Index)} = & \\ & 4 \times (\% \text{ residential}) + 2 \times \\ & (\% \text{ agricultural areas} + \% \text{ bare soil}) + \\ & (\% \text{ pasture} + \% \text{ Eucalyptus}). \end{aligned}$$

We included the percent *Eucalyptus* in this calculation because of its negative effects on benthic macroinvertebrate diversity and the toxicity of its allelopathic substances (Canhoto and Laranjeira, 2007; Remor *et al.*, 2013). FEMAT (1993) mandated 100-m riparian buffers alongside fish bearing streams in the Pacific Northwest USA, and Leal *et al.* (2016) reported that disturbance associated with forest cover and forest change within 100-m buffers



were strongly associated with several physical habitat variables in Amazonian streams.

We calculated the LDI from an index of the near-shore anthropogenic disturbance intensity and extent (*RDis\_IX*) described by Kaufmann *et al.*, (2014c). This index incorporates the separate sums of proximity-weighted tallies of 3 types of agricultural disturbance and 9 types of non-agricultural disturbance, based on field observations of their presence and proximity. It is composed of metrics described by Kaufmann *et al.*, (2014a), and calculated separately for the riparian and drawdown zones:

- *bfxHorizDist*=Drawdown exposure (horizontal distance in meters) from the shoreline to the high-water mark.
- *hifpAnyCirca*=Proportion of stations with at least one type of human activity within transects.
- *hiiAg*=Human Disturbance Intensity – Agricultural types (sum of mean proximity-weighted tallies for 3 separate types of Agricultural activities)
- *hiiNonAg*=Human Disturbance Intensity – Non-Agricultural types (sum of mean proximity-weighted tallies of 9 types of Non-Agricultural human activities)
- *hiiAll*=Human Disturbance Intensity – (sum of proximity-weighted tallies of all 12 types of human activity)

We adapted *RDis\_IX* for our separate riparian and drawdown zone data by apportioning the separate tallies of human disturbance from drawdown and riparian transects according to the proportion of drawdown exposure in ten 15 x 15-m transects adjacent to the wetted shoreline. We calculated the adapted variable *RDis\_IX\_syn* by first summing the values of the disturbance metrics (*e.g.* *hiiNonAg*) from the drawdown and riparian transects, after weighting each by the proportions of the 15-m band that were, respectively, within and not within the drawdown zone. We then averaged these drawdown-apportioned values over the 10-paired drawdown and riparian transects. Finally, *RDis\_IX\_syn* was calculated from *hiiAg\_syn*, *hiiNonAg\_syn* and *hifpAnyCirca* according to Equations 1 and 2 of Kaufmann *et al.* (2014c), and we defined  $LDI = RDis\_IX\_syn$ . Kaufmann *et al.* (2014a) reported that the precision of *RDis\_IX* relative to its potential range was moderate and its among-lakes variance relative to its repeat-sampling variance indicated minor to moderate effects of sampling noise. Those measures of precision justified its use in a national lake assessment in the USA (Kaufmann *et al.*, 2014c).

The MDI was calculated as the Trophic State Index (TSI) for reservoirs proposed by CETESB (2000), and is based on total phosphorus (TP) and chlorophyll-a (CI) determined at the sampling station scale:

$$TSI(TP) = 10 \times (6 - (1.77 - 0.42 \times (\ln TP) \ln 2^{-1}))$$

$$TSI(CI) = 10 \times (6 - ((0.92 - 0.34 \times (\ln CI) \ln 2^{-1})))$$

The Trophic State Index was calculated by averaging TSI (CI) (Três Marias=1.06±0.52; São Simão=3.58±7.18) and TSI (TP) (Três Marias=0.002±0.001; São Simão=0.002±0.002). Since Carlson (1977), trophic state indices have been used globally to assess the effects of nutrient enrichment on lake productivity and compare lake trophic state.

The range of disturbance evaluation was based on the location of the sites in an XY graph, with sites at the origin having no disturbance in macro or mesoscales. The more distant the points were from the origin, the greater the disturbance. To evaluate the anthropogenic impacts of the reservoirs at both macro and mesoscales combined we calculated the IDI (Integrated Disturbance Index), which was modified from Ligeiro *et al.* (2013). The IDI score is the Euclidean Distance between the position of each sampling site relative to the origin of the plane. We used the Pythagorean Theorem to define the IDI:

$$IDI \text{ (integrated disturbance index)} = [(BDI \cdot 3^{-1})^2 + (LDI \cdot 0.75^{-1})^2]^{1/2}$$

To standardize the range of the two axes, we divided BDI values by 3 and LDI values by 0.75.

## Data analyses

To check for autocorrelation of biotic data, we performed a Mantel test in Primer software (Clarke and Gorley, 2006). We constructed Bray-Curtis similarity matrices from macroinvertebrate taxa abundances and Euclidean distance from geographic coordinate data and checked if there was a relationship between the two matrices.

To compare both reservoirs regarding physical and chemical characteristics we performed t-tests on normal and homocedastic data or Mann-Whitney tests on data that did not comply with these assumptions. We also conducted Spearman correlations between physical and chemical water quality variables and IDI, BDI, LDI, MDI, and biological data to check for relationships. These tests were made from log transformed ( $x+1$ ) data.

We performed multiple regressions of biological metrics against the disturbance indices (BDI, LDI, MDI, and IDI). Before the analyses, the biological data were log transformed ( $x+1$ ); the BDI, LDI and IDI data were arcsine transformed (Zar, 1996). Pearson correlations between LDI and BDI scores were performed to test the presence of significant relationships. We also conducted t-tests to evaluate whether the indices and the biotic metrics values differed between reservoirs. All statistical analyses were performed in Statistica 7.0 software (Statsoft, 2004) and Primer 6.13 (Clarke and Gorley, 2006).

## RESULTS

### Physical and chemical parameters

The evaluation of physical and chemical parameters indicated significant differences ( $P < 0.01$ ) between both reservoirs for several major variables. São Simão Reservoir (SS) had higher values than Três Marias Reservoir (TM) for turbidity ( $TM = 4.1 \pm 2.2$ ;  $SS = 13.5 \pm 11$  NTU), total dissolved solids ( $TM = 0$ ;  $SS = 18.8 \pm 4.5$  mg/L) and alkalinity ( $TM = 2.1 \pm 0.2$ ;  $SS = 396.5 \pm 79.5$  mg/L), and lower photic zone depth ( $TM = 1.4 \pm 0.5$ ;  $SS = 0.5 \pm 0.3$  m).

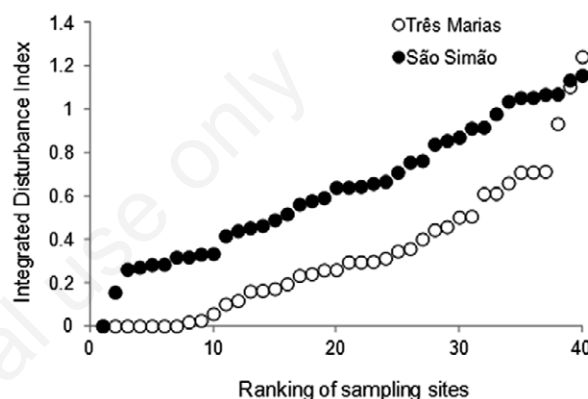
We found weak but significant ( $P < 0.05$ ) correlations between some water quality and biological variables (Tab. 1) but no significant correlations between IDI, BDI, or LDI and water quality. As expected, MDI was significantly correlated with chlorophyll a ( $r = 0.86$ ) and phosphorus ( $r = 0.46$ ) but was not related to other physical or chemical variables.

### Reservoir disturbances measures

The buffer land use surrounding Três Marias Reservoir was primarily natural land uses consisting of Cerrado (natural Savanna) (30.07%) and forest (26.70%). *Eucalyptus* (16.31%), pasture (10.68%), agriculture (10.59%), bare soil (4.09%), flooded vegetation (1.15%), and human structures (0.41%) were also observed. In contrast, most land uses around São Simão Reservoir were human modified, consisting of pasture (45.42%), agriculture

(24.87%), bare soil (17.89%), forest (6.95%), field (3.41%), human structures (1.31%), and flooded vegetation (0.15%).

Those differences in buffer land use resulted in significantly higher BDI scores for São Simão Reservoir (Três Marias:  $0.58 \pm 0.49$ ; São Simão:  $1.17 \pm 0.46$ ;  $P < 0.01$ ). The LDI (Três Marias:  $0.16 \pm 0.24$ ; São Simão:  $0.32 \pm 0.28$ ;  $P = 0.01$ ) and IDI (Três Marias:  $0.34 \pm 0.31$ ; São Simão:  $0.64 \pm 0.30$ ;  $P < 0.01$ ) also indicated greater disturbance of São Simão (Fig. 3). At São Simão Reservoir, nearly all



**Fig. 3.** Integrated Disturbance Index score for sites in Três Marias and São Simão Reservoirs ranked by increasing scores.

**Tab. 1.** Spearman significant correlations ( $P < 0.05$ ) between physical and chemical variables and biological metrics obtained from Ekman-Birge grab and kick-net samples for Três Marias and São Simão Reservoirs.

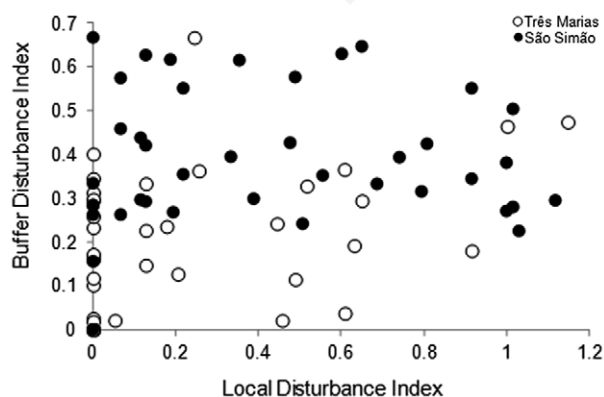
Sampler	Water quality x biological metrics	Três Marias	São Simão
		r	r
Ekman-Birge Grab	Alkalinity x % EPT individuals	0.42	
	Alkalinity x % EPT richness	0.39	
	Alkalinity x % resistants		0.35
	Alkalinity x % Chironomidae individuals		0.36
	Conductivity x EPT richness	0.32	
	Pheophytin x benthos richness	0.34	
Kick-net	Alkalinity x benthos richness	-0.32	
	Alkalinity x abundance of EPT individuals		-0.35
	Electrical conductivity x benthos abundance		-0.34
	Electrical conductivity x abundance of resistants		-0.33
	Electrical conductivity x Chiromidae abundance		-0.31
	Pheophytin x % EPT individuals		-0.40
	Pheophytin x abundance of resistants		0.32
	Pheophytin x Chiromidae abundance		0.32
	Phosphorus x benthos abundance		-0.32
	Phosphorus x % EPT individuals		-0.32
	Phosphorus x % EPT richness		-0.32
	Secchi depth x benthos richness	0.33	
	Temperature x % EPT richness		0.44
	Total dissolved solids x abundance of EPT individuals		-0.33
	Turbidity x % EPT individuals		-0.37

Richness, number of unique taxa at the lowest level identified; genera for Chironomidae, families for all others.

sites (33) had some kind of human influence, even if in small amounts. At sites with higher LDI values, there were many anthropogenic influences, such as buildings, walls, dikes or revetments, trash, roads, or man-made beaches. Only two São Simão sites had high levels of pasture. At Três Marias, 19 sites had some kind of human influence, two of which had high levels of agriculture.

The significantly higher differences in IDI scores for São Simão Reservoir were reflected in 10 of the 13 Ekman-Birge dredge biotic metrics. Compared with Três Marias, significantly lower values were obtained in São Simão for benthos richness ( $P<0.001$ ), abundance of EPT individuals ( $P<0.05$ ), % EPT individuals ( $P<0.001$ ), and EPT richness ( $P<0.001$ ). Similarly, benthos abundance ( $P<0.001$ ), abundance of resistant ( $P<0.001$ ), % resistant ( $P<0.001$ ), Chironomidae abundance ( $P<0.001$ ), Chironomidae richness ( $P<0.01$ ), and % Chironomidae individuals ( $P<0.001$ ) were significantly higher for this reservoir. In addition, 5 of 12 kick-net biotic metrics were significantly higher for São Simão than Três Marias: benthos abundance ( $P<0.001$ ), abundance of resistant ( $P<0.01$ ), % Chironomidae individuals ( $P<0.01$ ), abundance of Chironomidae ( $P<0.001$ ), and abundance of EPT individuals ( $P<0.001$ ). No significantly lower values were found for the kick-net-based biotic metrics in São Simão.

We observed substantial variation in the level of disturbances among sites within both reservoirs at both the macro (BDI) and mesoscales (LDI) (Fig. 4). We found insignificant correlation between the BDI and LDI for São Simão Reservoir ( $r=0.03$ ), but a low and significant value for Três Marias ( $r=0.38$ ). In both reservoirs, all sites were oligotrophic with MDI scores showing a small range, between 20 and 36, except for the six ultra-oligotrophic sites in São Simão, with MDI scores less than 7, so this index was not used in the IDI.



**Fig. 4.** Buffer Disturbance Index scores, based on land use coverage in 250 m radius buffers at each sample site, versus Local Disturbance Index scores for the riparian/littoral assessment plots at each site.

### Structure and composition of benthic macroinvertebrate assemblages

The Ekman-Birge sampling at Três Marias Reservoir produced a total of 976 individuals (23 families), with an average density of 1084 individuals  $m^{-2}$  (Supplementary Tabs. 1 and 2). We collected 698 Chironomidae individuals (72% of total individuals) and 24 genera, with an average density of 795 Chironomids  $m^{-2}$ . The kick-net samples produced a total of 4464 individuals (21 families), with an average density of 1786 individuals  $m^{-2}$  including 2874 Chironomidae individuals (64% of the total) and 25 genera at an average density of 1150 individuals  $m^{-2}$ . We also collected the non-native species *Melanoides tuberculatus* (0.6%), *Corbicula fluminea* (0.6%), and *Macrobrachium amazonicus* (0.1%). We found *M. tuberculatus* in sites with low and high LDI values and *C. fluminea* in a site with a high BDI value. *M. amazonicus* was present in sampling sites with high BDI and LDI values.

At São Simão Reservoir we collected 3693 organisms and 14 taxa with the Ekman-Birge grab, including 3536 Chironomidae (96% of the total) distributed in 26 genera. The kick-net sampling yielded 8921 organisms and 25 genera, including 7218 Chironomidae individuals (81% of the total) and the non-native *M. tuberculatus* (0.01%).

We found no abundance autocorrelation of biotic data for Três Marias ( $R=0.01$ ;  $P=0.55$ ) or for São Simão ( $R=0.12$ ;  $P=0.07$ ) reservoirs. The buffer (BDI), site (LDI), and micro (MDI) disturbance index scores did not correlate with the site biological metric scores for either reservoir (Tab. 2). Hence, it was not possible to establish relationships between the benthic macroinvertebrates and micro, meso or macroscales or integrated disturbances within individual reservoirs.

### DISCUSSION

The studied reservoirs suffer many human impacts, and we identified different disturbance levels in near-shore areas at meso and macroscales of both reservoirs. Landscape modifications resulting from human interventions yielded a gradient of ecological conditions as a result of land use, as others studies have reported for lakes (Whittier et al., 2002b; Dodds et al., 2006; Kaufmann et al., 2014c).

Probably the highly altered land use and physical habitat at nearly all sites in São Simão Reservoir contributed to the absence of correlation between BDI and LDI. On the other hand, the correlation between these indices at Três Marias, although weak, probably results from the greater heterogeneity in disturbance degree (Fig. 4). The importance of evaluating anthropogenic pressures at multiple spatial scales is already known (Hughes et al., 2006; Johnson and Host, 2010), including understanding that

measurements at different spatial scales may not be comparable to each other (Turner *et al.*, 1989). Hence, the choice of spatial scales influences the results of landscape level assessments because important parameters and processes at one scale may not be observed in another, resulting in information loss (Henderson-Sellers *et al.*, 1985; Meentemeyer and Box, 1987). The occurrence of wide variation in the magnitude of disturbance at macro and meso-scales among sites within the two reservoirs and a higher general level of disturbance at São Simão allowed us to accept our first and second hypotheses.

We did not observe relationships between biotic metrics and disturbance levels at different scales among individual sites within each reservoir; however, we did observe such a relationship between reservoirs, similar to what Kaufmann *et al.* (2014b) reported for fish and riparian birds and Sanches *et al.* (2014) reported for non-native fish species richness and percent individuals. In the dredge samples, we observed higher values for typical metrics that indicate human impact (*e.g.*, abundance and richness of Chironomidae) at São Simão. Moreover, at Três Marias Reservoir, we found higher values for sensitive biotic metrics indicating that this ecosystem provides

conditions for survival of more different species and for sensitive taxa, corroborating the IDI results. Therefore, the third hypothesis was accepted also. Ligeiro *et al.*, (2014) highlighted the importance of benthic macroinvertebrates as bioindicators of environmental quality, mainly EPT assemblages as indicators of good water quality, whereas chironomids tend to demonstrate poor conditions, because of their capacity to exploit different food resources, reproduce quickly under a variety of conditions, and tolerate fine sediments, a common characteristic of the reservoirs studied (Callisto *et al.*, 2014).

Although MDI (station trophic state) scores did not correlate with biological metrics, three water quality variables had weakly significant correlations ( $P < 0.05$ ) with those metrics (Tab. 1). The positive correlation for alkalinity and EPT metrics may have occurred because moderate increases in alkalinity enhance the biomass of primary producers (periphyton and macrophytes), which can elevate the proportions of EPT (Koetsier *et al.*, 1996). Increased temperature can negatively affect the growth of macroinvertebrates if there is insufficient food, but it can reduce their development time and accelerate their growth if food is abundant and the temperature increases are not

**Tab. 2.** Regression results between the Integrated Disturbance Index and biological metrics obtained from Ekman-Birge grab and kick-net samples for Três Marias and São Simão Reservoirs. (Richness: number of unique taxa at the lowest level identified; genera for Chironomidae, families for all others).

Sampler	Biological metrics	Três Marias		São Simão	
		P	R <sup>2</sup>	P	R <sup>2</sup>
Ekman-Birge grab	Benthos abundance	0.4330	0.0442	0.373	0.0209
	Benthos richness	0.9677	0.0017	0.2385	0.0364
	Abundance of EPT individuals	0.4943	0.0374	0.8689	0.0007
	% EPT individuals	0.8103	0.0113	0.6607	0.0051
	EPT richness	0.7625	0.0146	0.6342	0.006
	% EPT richness	0.6561	0.0225	0.647	0.2132
	Abundance of resistants	0.6891	0.0199	0.5885	0.0153
	% resistants	0.9862	0.0008	0.4067	0.0182
	Abundance of non native	0.5569	0.0091	NP	NP
	% Non native individuals	0.4841	0.1296	NP	NP
	Chironomidae abundance	0.497	0.0371	0.4389	0.0159
	Chironomidae richness	0.6811	0.0205	0.7547	0.0026
	% Chironomidae individuals	0.6908	0.0198	0.4275	0.0167
Kick-net	Benthos abundance	0.727	0.0171	0.113	0.0648
	Benthos richness	0.4243	0.0453	0.7171	0.0035
	Abundance of EPT individuals	0.8936	0.0005	0.8216	0.0014
	% EPT individuals	0.2259	0.0384	0.6441	0.0057
	EPT richness	0.4568	0.0147	0.4211	0.0171
	% EPT richness	0.5881	0.0078	0.3683	0.0214
	Abundance of resistants	0.0600	0.0904	0.2409	0.036
	% resistants	0.474	0.0136	0.511	0.0114
	Abundance of non native	0.9076	0.0004	0.0885	0.0745
	% Non native individuals	0.7678	0.0023	0.0885	0.0745
	Chironomidae abundance	0.7919	0.0125	0.2399	0.0362
	Chironomidae richness	0.4264	0.045	NI	NI
	% Chironomidae individuals	0.9367	0.0722	0.4888	0.1269

NP, not present; NI, not identified; richness, number of unique taxa at the lowest level identified; genera for Chironomidae, families for all others.



extreme (Hughes and Davis, 1986; Bayoh and Lindsay, 2003; Feuchtmayr *et al.*, 2007). Pheophytin is a natural degradation product of chlorophyll-a (Streit *et al.*, 2005). Its negative correlation with percent EPT individuals in São Simão may indicate the effect on EPT of degraded terrestrial plant tissue from the riparian zone that had eroded into the reservoir littoral zone (Junet *et al.*, 2005). All three biological metrics were EPT based, suggesting their sensitivity to water quality changes, as has been documented elsewhere (Stoddard *et al.*, 2008; Moya *et al.*, 2011; Chen *et al.*, 2014).

Regarding the kick-net samples, the absence of significant differences in the metrics based on sensitive taxa between Três Marias and São Simão might also be related to the fact that we employed this equipment in areas that had recently been exposed in the dry season. In such environments, only resistant taxa and a significantly lower abundance of sensitive taxa, such as EPT, would be expected. The higher significance value of the abundance of EPT individuals in the kick-net based metrics for São Simão was mainly influenced by the high number of Leptohyphidae individuals collected in this reservoir (798) when compared with 110 individuals sampled in Três Marias. This family includes tolerant Ephemeroptera taxa (Firmiano, 2014) and this Leptohyphidae abundance pattern was much weaker in the dredge data (56 individuals in São Simão and 30 in Três Marias).

It is likely that other elements also influenced benthic macroinvertebrates in the reservoir littoral zone, particularly the seasonal variation in water level. For example, in the previous dry season the Três Marias water level was nearly 10 m lower vertically and an average of 113 m lower horizontally than in the sampling period (CEMIG, 2012). Changes in the benthic fauna resulting from variations in reservoir water level were recorded in previous studies. A Wisconsin (USA) study assessed benthic macroinvertebrates before and after a drawdown episode and observed that part of the benthic fauna decreased rapidly on substrates exposed to the air; three months were necessary for recolonizing the habitat (Kaster and Jacobi, 1978). A similar study assessed benthic macroinvertebrates in reaches upstream and downstream of an Idaho (USA) dam (Munn and Brusven, 1991). The authors observed that the benthic macroinvertebrates downstream of the reservoir were strongly altered, resulting in high abundance of Chironomidae and low taxa richness. The variation in water level was considered a main effect in simplifying the assemblage. In a study carried out at the Three Gorges Dam in China, Shao *et al.* (2008) monitored benthic macroinvertebrates for three years. The authors concluded that variation in water level resulted in greater proportional abundance of chironomids and oligochaetes. Aura *et al.* (2010) in Kenya used an index of biotic integrity to evaluate benthic macroinvertebrates in impaired and fluctuating rivers, and

observed a replacement of sensitive taxa by tolerant taxa, such as *Chironomus* sp., *Lumbricus* sp. and oligochaetes.

Another factor that could have affected the results of our study was the two-year gap in sampling each reservoir. In April 2011, Três Marias had a useful volume of 99.0% and in April 2013 the useful volume of São Simão was 70.3% (ONS, 2016). This last value represents approximately 78% of the useful volume of São Simão in the same month in 2011 (90.3%). This 12.3% decrease in the useful volume of São Simão increased the drawdown zone of this reservoir in 2013 related to 2011, and could have contributed to overestimating the LDI data for this reservoir, because drawdown is used to construct this index.

Considering previous studies and our results, we believe that distribution of the benthic macroinvertebrates resulted from variation in water level more than the other disturbances we measured. The structure of benthic macroinvertebrates can be influenced more by low colonization rates than by variations in water level (Marchant and Hehir, 2002). But water level variation favors early colonizers, such as chironomids (Baxter, 1977). Chironomids use several food resources and reproduce rapidly, allowing them to occupy a wide variety of relatively unstable habitats (Trivinho-Strixino and Sonada, 2006). Thus, it is likely that chironomid dominance in the reservoir littoral zones resulted from their capacity to occupy different habitats and rapidly colonize recently flooded areas. These results are particularly important considering recent reservoir levels in Brazil. Recently, reservoir water levels have been lower than the historic average, some of them have reached the dead (inactive) volume of less than 15% of their levels at the end of the rainy season, which increases the probability of electrical brownouts or blackouts during the dry season.

Corroborating the results of our study, other studies in Brazil suggest that there is a relationship between the presence of non-native species and the construction of reservoirs (Pamplin *et al.*, 2006; Jorcin and Nogueira, 2008; Schiavone *et al.*, 2012; Sanches *et al.*, 2014). The alteration of river flow resulting from the construction of large reservoirs can favor the establishment of non-native species, which may affect native communities (Whittier *et al.*, 2002; Bunn and Arthington, 2002a). The continuous input of biological material coming from the main river and its tributaries can make reservoirs vulnerable to invasions by non-native species (Rocha *et al.*, 2011).

Because we used a kick-net where there was more flooded vegetation, it is likely that those sites had more biotic and abiotic elements used by organisms for their maintenance and growth. The quality of the substrate in rivers, lakes and reservoirs has been recognized as one of the factors that most influence the distribution of benthic macroinvertebrates (Molozzi *et al.*, 2012, 2013b; Macedo *et al.*, 2014a; Tupinambás *et al.*, 2014). Among them, aquatic

macrophytes offer shelter against predators, spawning sites, and food resources for aquatic organisms (Wilcox and Meeker, 1992). Considering this, the higher amount of macrophytes where the kick-net was used probably contributed to the greater abundance of individuals collected and the reduced dominance of chironomid larvae.

Because reservoirs are a challenge for water resource management (Jennings *et al.*, 1995) and stakeholders are interested in developing programs that can be expected to meet behavioral change goals at least cost (Bell *et al.*, 2013), we suggest that future studies compare larger sets of reservoirs that are minimally and highly disturbed (Molozzi *et al.*, 2011; Kaufmann *et al.*, 2014b, 2014c; Morais *et al.*, 2014, Petesse *et al.*, 2014). In such studies, it is important to assess different spatial scales of environmental quality, non-native species populations, and the influence of water level variations on water quality and biological responses.

## CONCLUSIONS

Our results showed that assessment of environmental quality at different spatial scales resulted in the classification of sampling sites along a range of disturbances for each reservoir. We observed higher values for biotic metrics that indicate human impact at São Simão Reservoir, which also experienced greater levels of human pressures. Nonetheless, we did not observe significant relationships between biotic metrics and disturbance indices at different scales. We believe that annual water level depletions during the dry season strongly influenced littoral zone benthic macroinvertebrates six to seven months later, affecting possible responses to differences in land use and habitat complexity at the water levels observed during our study. Those water level fluctuations favored assemblages dominated by early colonizers with short life cycles. Accordingly, water level drawdown would tend to affect all sites within the same reservoir to some degree, regardless of differences in near-shore or upland disturbances within the set of reservoir sample sites. Therefore, we recommend that assessments of reservoir environmental quality consider both different spatial scales and different degrees of water level variation as factors influencing benthic macroinvertebrates among different years within single reservoirs or among different reservoirs that experience different levels of drawdown. We also recommend sampling in the reservoir limnetic zone to assess possible differences between permanently *versus* recently flooded sites.

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