

Full Paper

# | Vol 7 || No. 4 || October-December 2015|

# Environmental Implications of a Stabilization Pond Effluent Released in Paraná River at Ilha Solteira, Brazil: The Quality of Water and Sediment

Douglas P. Pedroso<sup>a</sup>, Felipe A. Santos<sup>\*b</sup>, Alexandre O. Jorgetto<sup>a</sup>, João G. T. Queluz<sup>a</sup>, Bruno P. Rocha<sup>a</sup>, Sônia M. A. Jorge<sup>a</sup>, Marco Antonio U. Martines<sup>b</sup>, Gustavo R. Castro<sup>a</sup>

<sup>a</sup>Departament of Chemistry and Biochemistry, Institute of Bioscience, São Paulo State University, 18618-000 Botucatu – SP, Brazil. <sup>b</sup>Institute of Chemistry, Federal University of Mato Grosso do Sul, 79070-900 Campo Grande – MS, Brazil.

Article history: Received: 05 August 2015; revised: 26 October 2015; accepted: 08 November 2015. Available online: 25 December 2015. DOI: http://dx.doi.org/10.17807/orbital.v7i4.777

**Abstract:** Sewage treatment plants in Ilha Solteira City, which consist of stabilization ponds, have been modifying water and sediment characteristics around sewage outfall area. Water quality parameters such as dissolved oxygen (OD) and conductivity were directly influenced by the sewage outfall. OD values were reported before ( $6.36 \text{ mg L}^{-1}$ ) and after ( $2.94 \text{ mg L}^{-1}$ ) the sewage outfall. Measurement of organic matter content in the sediment showed the same pattern, with values of 1.34% and 4.80% for the SP1 and SP3 samples, respectively. A copper and zinc analysis indicated that metals are being transported through the water column, and SP3 is an important sedimentation zone. The total concentrations reported for copper are 6.48, 26.97, 37.54, 27.12 and  $18.55 \text{ mg kg}^{-1}$  for SP1, SP2, SP3, SP4 and SP5, respectively, and the total concentrations for zinc are 11.21, 48.53, 67.41, 58.62 and  $24.76 \text{ mg kg}^{-1}$ . Bureau Community of Reference (BCR) sequential extraction indicated the following bioavailability order of copper and zinc: SP1 < SP5 < SP4 < SP3 < SP2. At SP2, more than 70% of the copper and zinc is found in bioavailable fractions. Benthic organism's analysis indicates that the Oligochaetes correlation with organic matter content is related to the decrease in water quality.

Keywords: BCR sequential extraction; metal species; benthic organisms

#### **1. INTRODUCTION**

The quality of environmental media such as atmosphere, water, and soil has been severely impacted by human activities. Among these activities are the mining and textile industries, which usually contribute to the poisoning of the environment by releasing contaminated effluents containing toxic metals and dyes. Due to exponential urban growth, domestic sewage has become an important source of organic substances that contribute to the compromise of the natural water quality [1].

Domestic sewage has attracted the attention of scientists due to its environmental impacts such as poor recreational water quality due to the presence of pathogens, eutrophication, nutrient loading, toxicity to humans and ecosystems, the presence of chemicals and the accumulation of contaminants that can be transferred through the food chain [2-4]. The great majority of available sewage treatment methods are not effective for the removal of contaminants such as toxic metal/ species and pharmaceutical products [5-8], and because these contaminants are not biodegradable and/or possess a long life in environmental media, the contaminants can be assimilated by living organisms such as mussels and fishes [5, 9]. The presence of toxic metal species in aquatic systems, particularly in the surface sediments, represents potential risks, as these toxic metal species can be transferred to the water column due to changes in pH and redox parameters, thus becoming available to biological organisms [1, 2, 10, 1 1].

The great majority of metal species that enter the aquatic systems are associated with suspended particulate matter because they are adsorbed onto organic and inorganic particles with several origins. Suspended organic particulate matter usually exhibits a large variety of organic groups such as carboxylic

<sup>\*</sup>Corresponding author. E-mail: <u>santos\_felipe1@yahoo.com.br</u>

acids, amines, amides, phenols and thiol or sulfhydryl groups (-SH), which can act as adsorption sites for metal species [12, 13]. The inorganic suspended particulate matter is composed of several classes of substances. However, the hydrous oxides of iron and manganese are of special importance. Because the surfaces of these hydrous oxides are covered with hydroxyl groups, which can act as Brønsted acids (proton donors), these surfaces in their deprotonated forms can also interact with metal species in solution [14].

Another parameter of great importance that is currently under investigation is the sediment particle size. Because smaller particles present a higher surface area and also can travel longer distances than larger particles, the smaller particles have the potential to contaminate locations more distant from the emitting source [15].

To understand the risk posed by metal species in surface sediments, numerous speciation methods have been proposed to evaluate the forms and phases with which metals are associated. In the last decade, sequential extraction methods have been applied to this type of investigation. Recently, the speciation method developed by the Community Bureau of Reference (BCR sequential extraction) began to be used extensively to assess the bioavailability and pollution risk potential of metal species in water samples [16-19].

In addition to the concentrations of metal

species, the quality of water and sediments can also be evaluated relative to the organisms that exist in the water. Thus, many investigations have been performed using benthic organisms as bioindicators because the presence or disappearance of these organisms can reflect eventual environmental changes occurring in the ecosystem [20-22].

The goal of this work was to monitoring and investigates the quality of sediment and water of Paraná River, during a period of three years, at several sampling points to understand the influence of an effluent plume released by a local stabilization pond. Parameters such as pH and dissolved oxygen in the water were investigated. A sequential extraction procedure was applied to the surface of sediment samples to determine the availability of metal species, and some benthic organisms were used as water quality indicator parameters.

# 2. MATERIAL AND METHODS

# Characterization of the study area

The Paraná River is one of the most important rivers in Brazil because two of the biggest Brazilian hydroelectric power plants are installed along its course: The *Itaipú* and *Ilha Solteira* Hydroelectric Power Plants. The *Ilha Solteira* Hydroelectric Power Plant is situated in Ilha Solteira City, São Paulo State, Brazil, and the *Ilha Solteira* Hydroelectric Power Plant is located near the study area, as shown in Figure 1.



**Figure 1.** Image of studied area, including the Ilha Solteira Hydroelectric Power Plant (A), sewage releasing point (2), stabilization ponds (B) and Ilha Solteira City (C). Source: Google Earth.

The Paraná River is the main tributary of La Plata Bay with a flow of approximately 16,000 m<sup>3</sup> s<sup>-1</sup>. This river is a natural divisor between Brazil and Paraguay and between Paraguay and Argentina. Along its course to Argentina, the Paraná River runs through

regions with growing population densities as well as agricultural areas, which are the two major causes of the decrease in water quality.

Ilha Solteira is a small city with very few industries, implying that fishing activity is very

important to the local population. The fishermen from Ilha Solteira are responsible for the local and regional fish supply, so the monitoring of water and sediment quality is very important to ensure good development of aquatic organisms and animals.

### Sampling and sample analysis

The representative sediment samples from the surface were collected in the first 10 cm layer at five sites 1 mile apart from each other, and the depth of the sampling points ranged between 4 and 5 meters. At each point 5 sediment samples (with ~1 Kg each one) were collected and mixed in order to homogenize and get a laboratory sample. A Petersen dredge with an area of 0.052 m<sup>2</sup> was used. Sediment samples were treated differently according to the purpose of the analysis. To investigate the benthic organisms, the samples were immediately sieved in a nylon sieve (0.250 mm mesh) and conditioned in a 4 % formaldehyde solution. In the laboratory, a counterstaining technique using a rose Bengal (12.0 mg L<sup>-1</sup>) solution was applied to the sediment samples, and organisms identification was performed according to methods described in the literature [20-22]. To analyze the metal species, the collected sediment samples were transferred into plastic bags (without any conditioning substance) and at the laboratory it were dried at 105 °C, homogenized and fractionated according to size. The sampling procedure were repeated during a period of three years (samples were collected in January/May/September) in order to investigated and understand the changes occurred in the environment. This way all results were expressed as a medium of three years of investigation.

The particle size fraction equal to 0.063 mm was used in the sequential extraction procedure. Because contaminants are usually concentrated in the clay-silt fraction, this fraction is nearly equivalent to the material carried in suspension, and sieving does not alter the contaminant concentration [15]. The organic carbon was determined by mass loss upon ignition at 550 °C for 12 hours using 1.0 g of sieved sediment sample weighed in a porcelain capsule [1,23].

The sequential extraction procedure was performed according to the method of Minello *et al.* (2010). The solutions and steps involved in the metal extraction using 1.0 g of sieved sediment sample are summarized in Table 1. The mixture (solvent extractor and sediment) were separated using a centrifuge (FANEN, model Excelsa II). All reagents used in this step were of analytical grade.

Steps Involved	Solutions/Concentration	Extraction time (h)	Associated phase
Step 1	$0.11 \text{ mol } L^{-1}$ acetic acid	16	Carbonate
Step 2	5.0 mol L <sup>-1</sup> hydroxylammonium chloride	16	Iron and manganese hydrous oxide
	5 mL of hydrogen peroxide (30 %)	~ 1	-
Step 3	5 mL of hydrogen peroxide (30 %)	~ 1	Sulfides and organic matter
	1.0 mol L <sup>-1</sup> ammonium acetate, pH 2 adjusted with nitric acid	16	

**Table 1.** Solutions and steps involved in the sequential extraction procedure applied to sediment samples from

 Paraná River, Ilha Solteira, São Paulo State, Brazil.

To determine the concentrations of the metal ions under investigation, 0.5 g of each sieved sample was accurately weighed and put into a glass tube with 15 mL of concentrated HCl (J.T. Baker) and 5 mL of a HNO<sub>3</sub> (J.T. Baker) solution. The mixture remained unheated for 12 hours. The samples were then heated to 100°C for 60 minutes. Every 15 minutes, 1 mL of H<sub>2</sub>O<sub>2</sub> (30 %) was added. After mineralization, the sample was filtered, and the final volume was adjusted to 25 mL in a volumetric flask. The procedure was also applied to a standard sediment reference material (Lake sediment IAEA-SL-1 and LKSD-3). A Shimadzu Model AA-6800 atomic absorption spectrometer equipped with hollow cathode lamps for analytes and a deuterium lamp for background correction was used. The spectrometer monochromator was adjusted to 324.7, 213.9, 248.3 and 279.5 nm for the analysis of copper, zinc, iron and manganese, respectively. The standard metal solutions were prepared daily after stepwise dilution from a 1000 mg  $L^{-1}$  stock solution (Merck-Germany) in ultrapure water (Direct-Q, Millipore) and acidified with nitric acid to pH 2.

The parameters related to water quality

(conductivity, pH, dissolved oxygen (DO)) were measured in situ over a period of 12 months (from January 2011 to January 2012) in the region immediately above the area where the sediment samples were collected using a pre-calibrated multiparameter probe, YSI 6600 V2-4 model.

# 3. RESULTS AND DISCUSSION

Water analysis

The parameters pH, dissolved oxygen and conductivity were selected because the pH and dissolved oxygen are closely related to metal mobilization in sediment. This mobilization may occur through ion exchange or redox reactions. Because the conductivity reflects the direct presence of dissolved ions, the extent of the plume produced by the sewage outfall could be assessed by the analysis of the conductivity values. The results of the analysis of these parameters are presented in Table 2 as an average of 12 measurements.

**Table 2.** Parameters related to water quality obtained using a multiparameter probe and organic matter content in surface sediment from Paraná River, Ilha Solteira, Brazil. The results are present as an average of 12 measurements.

	Parameters		Sampling points			
	1	2	3	4	5	
рН	$7.4\pm0.7$	$7.38\pm0.5$	$7.09\pm0.6$	$7.36\pm0.6$	$7.45\pm0.5$	
Cond. ( $\mu$ S cm <sup>-1</sup> )	35.0 ± 5	52.2 ± 15	49.4 ± 13	38.1 ± 10	34.0 ± 13	
OD (mg L <sup>-1</sup> )	$6.36 \pm 2.1$	4.55 ± 1.3	$2.94\pm0.6$	$4.24\pm0.4$	$5.40 \pm 0.9$	
OM (%)	$1.34\pm0.2$	$5.55\pm0.4$	$4.80\pm0.7$	$3.15\pm0.5$	$2.36\pm0.4$	

1 =Blank point; 2 = Sewage outfall or emissary; OM = Organic matter.

According to Table 2, as the plume pollution rises at sampling point 2 (the sewage outfall), the plume exhibits the highest conductivity value (52.2 µS cm<sup>-1</sup>). At sampling points 3 to 5, the conductivity values decrease due to dilution as the flow increases until the conductivity value is 34.0  $\mu$ S cm<sup>-1</sup> at point 5, a value close to the value that was obtained for the blank at sampling point 1, where there is no influence from the sewage outfall. The pH values did not undergo major changes over the series of sampling points, which reflects the good capacity of the system to neutralize acids and bases. The pH parameter is of great importance because at low pH levels, a higher concentration of H<sup>+</sup> ions can mobilize metal species that exist on the surface or in particles through ion exchange reactions. The dissolved oxygen levels also show the influence of the sewage outfall over the series of sampling points. Table 2 shows that the dissolved oxygen concentration at sampling point 3 exhibits the lowest value, 2.94 mg L<sup>-1</sup>. This site represents the peak of oxygen consumption by the oxidation of organic matter and other compounds such as ammonia. At subsequent points, the values for the dissolved oxygen

concentration increase, again reaching 5.40 at sampling point 5.

#### Sediment analysis

After sediment sampling, each sediment sample was dried, and the organic matter content was determined in the bulk samples through loss upon ignition at 550 °C for 12 h. The organic matter content is an important parameter because this fraction is composed of several types of substances such as amines, carboxylic acids and sulfur-containing compounds, all of which can bind metals. The results obtained are listed in Table 2. According to these results, sampling points 2 and 3 show the highest organic matter content due to the proximity of the sewage-emitting source.

Another important parameter related to sediment characterization is the size fraction because finer particles exhibit greater reactivity. Each sediment sample was fractionated into several particle sizes, ranging from 1000 to  $45 \mu m$ .

Sampling point 3 shows the highest percentage (18%) of particles with diameters smaller than 100  $\mu$ m. The blank sampling point presented negligible amounts of particles smaller than 100  $\mu$ m, so the majority of the smaller particles found in SP3 are derived from the sewage outfall, which, therefore, has a high organic content. These results corroborate the results presented in Table 2 for the organic matter content. The study of particle size distribution is important not only for sediment transport modeling but also because there is a strong correlation between concentration of contaminants and fine-grained particles [1, 5, 15, 24, 25].

According to the literature, the fraction with particle size < 75 and  $> 63 \mu m$  is recommended for contaminant study once it is concentrated in the claysilt fraction [15, 25]. The clay-silt fraction is nearly equivalent to the material carried in suspension, and the sieving process does not alter the contaminant concentration. Aliquots of 1 g of sediment from each sampling point were subjected to total mineralization followed by metal species determination using flame atomic absorption spectrometry (FAAS). The results obtained are shown in Figure 2.



**Figure 2.** Total concentration of copper, zinc (A), iron and manganese (B) after sample mineralization with *aqua regia*.

Analysis of Figure 2 shows that the SP3 sample contained the highest concentration of all investigated metal species: 47.5, 67.4, 4557.6 and 376.6 mg kg<sup>-1</sup>, for copper, zinc, iron and manganese, respectively. Although the results do not indicate that the sediment is contaminated, compared with quality guideline values in Table 3, the values for the copper concentration at sampling point 3 between the TEL and

PEL values reflect some concern because this sampling point seems to be a sedimentation zone for the sewage outfall suspended particles. Aliquots of sediment reference material (IAEA-SL-1 and LKSD-3) were subjected to the same mineralization procedure applied to the samples, and the values obtained and the values supplied by the manufacturer are also listed in Table 3.

Table 3. Comparison of guidelines provided by the CCN	ME (Canadian Council of Ministers of the Environment)
and measured values obtained for sampling point 3.	

Metal species		CCME	Exp	Experimental values			
	TEL	PEL					
Cu(II) (mg kg <sup>-1</sup> )	35.7	197.0		47.5 ± 1.37			
Zn(II) (mg kg <sup>-1</sup> )	123.0	315.0		$67.4 \pm 2.9$			
Mn(II) (mg kg <sup>-1</sup> )				$376.58 \pm 9.85$			
Fe(II) (mg kg <sup>-1</sup> )			4	$4557.6 \pm 147.8$			
Certified reference material							
IAEA-SL-1 Found LKSD-3 Found							
Copper (mg kg <sup>-1</sup> )	24-36	$24.8 \pm 0.4$	34	$34.1 \pm 0.7$			
Zinc (mg kg <sup>-1</sup> )	213-233	$212.54 \pm 14.3$	139	$144.7 \pm 5.2$			
$Mn(II) (mg kg^{-1})$	3300-3620	$3427 \pm 122.7$	1220	$1253.2 \pm 41.7$			
Fe(II) (mg kg <sup>-1</sup> )	65700-69100	$67225 \pm 1857.6$	3.5 %				

As the results show, no significant difference was found between the experimental and

manufacturer-provided values. Although the results indicate that the river sediment is not contaminated, the total concentration obtained through the complete sample mineralization does not provide a true idea about the percentage of the metal that is bioavailable and that can readily be assimilated by organisms.

To investigate the bioavailable metal fraction, the sediment samples were subjected to a Bureau

Community of Reference (BCR) sequential extraction procedure that consists of using different extracting solutions. The mixture (sediment sample and extracting solution) was stirred in an end-over-end stirrer, and after each extraction step, the extracting solution was stored in polyethylene flasks under refrigeration until analysis. The results obtained for the BCR sequential extraction procedure for copper and zinc are depicted in Figure 3 and Figure 4, respectively.



Figure 3. Distribution of copper in sediment fractions obtained through BCR sequential extraction at all sampling points.



Figure 4. Distribution of zinc in sediment fractions obtained through BCR sequential extraction at all sampling points.

From the graphic representation of the results obtained for the sequential extraction procedure performed for all sampling points, the copper distribution pattern from SP2 to SP5 is observed to be quite different from the pattern presented by SP1, where there is no influence from the sewage outfall. These results demonstrate that the sewage outfall has been modifying the quality and composition of water and sediment downstream. The same pattern was observed for zinc in the sediment fractions at all sampling points, as seen in Figure 4. In the BCR sequential extraction procedure, the metals in Step 1 are those that precipitated or coprecipitated with carbonate, and because carbonates are susceptible to changes in pH, this fraction is more mobile and, hence, more available for uptake by living organisms [26]. Figures 3 and 4 show that this fraction represents approximately 10 % of the total copper and zinc at SP1 (blank) and approximately 40 % of the total copper and zinc at SP2 (sewage outfall) and SP3 (accumulation zone), clearly reflecting the influence of the sewage outfall. SP4 and SP5 also present higher

values than SP1 relative to step 1 for copper and zinc, even when the metal concentrations show a decreasing pattern from SP3 to SP5.

The reducible fraction, obtained in Step 2, represents those metals bound to iron and manganese oxides or hydrous oxides existing in their hydrated forms with their surfaces covered by OH groups. These oxides also act as metal carriers transporting them over long distances [27]. Hydroxyl groups can act as Brønsted acids (proton donors), and the surface becomes negatively charged. Metals can then be coordinated with negatively charged oxygen atoms and, as protonation or deprotonation reactions are involved, implying that pH is also an important parameter that is directly related to metal availability. At lower pH values (high H<sup>+</sup> concentration), copper and zinc that are adsorbed onto the iron and manganese hydrous oxide surface can be displaced and mobilized into the water column. The results of this extraction step (SP1 ~15 % of copper and zinc; SP2 and SP3 ~ 30 % of copper and zinc and SP4 and SP5 < 30 % of copper and zinc) corroborate the results presented in Figure 3, where the iron and manganese concentrations have their highest values at SP3: 4557.3 and 376.5 mg kg<sup>-1</sup>, respectively. Step 3 represents the oxidizable fraction, where metals are associated with sulfides and organic matter such as humic substances through covalent bonds (sharing of electron pairs) between nitrogen, sulfur and other atoms existing in organic chains [13]. This fraction therefore represents a less labile metal species compared to those obtained with step 1 and 2 and, relative to copper, the fraction is equivalent to 8 to 33 %, with a distribution as follows: SP1<SP2<SP3<SP4<SP5. This distribution indicates that copper may be transported over long distances in complexes with organic matter. Relative to zinc, the content of zinc at all sampling points is near the values

found at SP1 (blank), which was approximately 20 %. Despite of this the metal existing in this fraction also deserve special attention once metal bound to organic matter and/or sulfides also can be mobilized to water column depending on the environmental conditions [10,28].

The residual fraction, which was obtained through mineralization with aqua regia, represents the metals retained within the crystal lattices of minerals and inside crystallized oxides and, therefore, is not considered bioavailable. Through analysis of Figures 3 and 4, the concentration of copper and zinc in this fraction is observed to be higher at SP1 (66 and 48.2 %, respectively) and drastically decreased at SP2 (8.05 and 11.8 %, respectively) and SP3 (4.6 and 12.0 %, respectively) as a result of sediment composition modified by the sewage outfall. At SP4 and SP5, the metal content increases to approximately 20 %, and as the distance from the pollution source increases, there is a tendency of the values to approach those observed at SP1.

To evaluate the environmental hazards posed by these metals, the individual contamination factor  $(C_f^i)$  for copper and zinc was calculated according to the following equation:

$$C_{f}^{i} = \frac{c_{1} + c_{2} + c_{3}}{c_{res}}$$
(1)

where  $c_1$ ,  $c_2$  and  $c_3$  represent the metal concentration in each extraction step, and  $c_{res}$ represents the metal concentration in the residual phase. According to equation 1, the individual contamination factor was calculated for SP3 (the highest concentration of copper and zinc compared to all sampling sites), considering the metal concentration fractions presented in Table 4.

	Copper (mg kg <sup>-1</sup> )	Zinc (mg kg <sup>-1</sup> )
Step 1	$20.69 \pm 1.07$	$31.61 \pm 1.19$
Step 2	$13.04 \pm 0.43$	$18.34 \pm 0.24$
Step 3	$12.91\pm0.52$	$11.08 \pm 0.31$
Aqua regia	$2.75 \pm 0.04$	$8.19\pm0.09$
Sum of extraction steps	$49.37 \pm 1.26$	$69.22 \pm 1.25$

Table 4. Results obtained for the sequential extraction procedure applied to SP3.

According to table 4, the calculated contamination factors were 16.96 and 7.45 for copper and zinc, respectively, and the global contamination factor (sum of individual factors) was 24.41. The contamination factor for copper is very close to the

contamination factor obtained for another sampling location upstream of the hydroelectric power plant [1].

Water quality assessed through benthic organisms

The benthic invertebrate assemblage found in sediments and substrates of streams reflects an important aspect of the biological condition of the stream because these communities can become very sensitive to various stressors that may occur during their life [20]. These stressors can be related to physical and chemical water parameters such as water flow, turbidity, pH, oxygen and the presence of toxic substances. To understand the effect of sewage outfall upon benthic organisms, the benthic organisms were investigated at all sampling points, SP1 to SP5, and the results concerning their incidence and density are presented in Table 5.

Tabla 4	5 1	Macroinv	ortohrata	doncitios	$(ind/m^2)$	obtained	over th	a compline	reitar
Table .	<b>5.</b> I	viacioniv	encorate	uensities	(mu/m)	obtained	over ui	e sampning	, snes.

Таха	SP1	SP2	SP3	SP4	SP5
OLIGOCHAETA	0	0	0	0	0
Tubificidae	0	29	337	91	0
Aullodrilus limnobius	0	22	28	19	78
GASTROPODA	0	0	0	0	0
Mesogastropoda	0	0	0	0	0
Aylacostoma guaranitica	85	0	81	152	98
Melanoides tuberculata	0	0	0	0	0
Pomacea	0	0	0	0	0
Basommatophora	0	0	0	0	0
Biomphalaria peregrina	38	0	0	29	0
BIVALVIA	0	0	0	0	0
Corbiculidae	0	0	0	0	0
Corbicula fluminea	89	0	438	77	62
DIPTERA	0	0	0	0	0
Chironomidae	0	0	0	0	0
Tanypodinae	65	0	0	0	53
Tanytarsus	0	0	0	0	0
ODONATA	0	0	0	0	0
Progomphus	0	0	0	38	0
HIRUDINEA	0	2342	0	0	0
TOTAL	85	2409	884	406	291

According to Table 5, the influence of sewage outfall at SP2 over benthic organisms is dramatically harmful; only two groups of such benthic organisms were found. These results indicate that the organisms found are resistant to the stressors posed by the sewage outfall. These stressors consist of a decrease in the dissolved oxygen concentration, high turbidity, high temperatures and high organic matter content. Nevertheless, the variety of benthic families increases from SP2 to SP5, indicating a possible improvement in environmental quality.

According to Table 5, the increasing number of organisms from the Oligochaeta family observed from SP2 to SP3 is a good indication that the water quality has been reduced. Oligochaetes are common in most freshwater habitats and are the group of invertebrates most often enlisted for bioindication purposes because their reproduction is enhanced in environments with a high organic content [30,31]. The increase in organic

matter has generally been associated with a decrease in the concentration of dissolved oxygen because part of the oxygen is used in the oxidation of the organic matter itself. Because some species can tolerate anoxic conditions, the presence of organisms from the Oligochaeta family at the SP3 sampling site was expected. This point represents a gathering of some characteristics such as a low concentration of dissolved oxygen and a high organic content due to the sewage outfall. Figure 5 presents the relationship between dissolved oxygen, organic content and Oligochaetes expressed as a percentage of the maximum value found for each. A high correlation between available dissolved oxygen, organic matter and the number of individuals found is readily observed.

As the organic matter content decreases from SP2 to SP5, the Oligochaetes follow the same behavior, and the high turbidity may be related to the small number of individuals found at SP2 (the sewage

outfall). Another family that deserves special attention is the Hirudinea (abundantly found at SP2), a class of leeches morphologically adapted for obtaining food consisting chiefly of the blood of fishes or other animals, including other invertebrate organisms, because many of these species of leeches are predators [30]. This behavior may explain the low variety in species and the high number of Hirudinea found at SP2. Nevertheless, the presence of Hirudinea may not be related to the water quality because Hirudinea is a species that tolerates a wide variety of conditions.



**Figure 5.** Relationship between benthic groups found and proximity to the sewage outfall.

#### 4. CONCLUSION

Although the values found for total copper and zinc concentration in sediment do not show that the water body is contaminated, constant monitoring is necessary because heavy metal species are accumulated in the sediment fraction that is more labile (associated with carbonate and hydrous oxides of iron and manganese). On the basis of the analysis of the benthic organisms, the decrease in water quality downstream of the sewage outfall was established, and SP3 was found to be a sedimentation zone where a great tendency of suspended matter deposition was observed through the results of organic matter content. The existence of the sewage outfall makes SP2 a special place with an abundance of food for fish and some invertebrate organisms. Because Ilha Solteira City is an important fish supplier, its water quality is of key importance to local human health.

#### 5. ACKNOWLEDMENTS

The author thanks CNPq for the fellowships granted to Pasquali D.P. (Proc. 501563/2010-5) and for

financial support (Proc. 479872/2010-4) and FAPESP for the fellowships granted to A. O. Jorgetto (Proc. 2011/14944-5) and G. R. de Castro (Proc. 2012/21795-9).

#### 6. REFERENCES AND NOTES

- Minello, M. C.; Paçó, A. L.; Martines, M. A. U.; Caetano, L.; Santos, A.; Padilha, P. M.; Castro, G. R. *J. Environ. Sci. Heal. A.* 2009, 44, 861. [CrossRef]
- [2] Manahan, S. E.; Environmental Science and Technology CRC Press/Lewis publish: New York, 2000.
- [3] Scanes, P. Monitoring environmental impact of ocean disposal of sewage, Lamparelli, C.C.; Ortiz, J.P., eds.; Submarine Outfalls: Design, Compliance and Environmental Monitoring. Secretaria do Meio ambiente de São Paulo, 2007, cap. 3.
- [4] Teodoro, A. C.; Duleba, W.; Gubitoso, S.; Prada, S. M.; Lamparelli, C. C.; Bevilacqua, J. E. Mar. Pollut. Bull. 2010, 60, 536. [CrossRef]
- [5] Boyd, G. R.; Reemtsma, H.; Grimm, D. A.; Mitra, S. Sci. Total Environ. 2003, 311, 135. [CrossRef]
- [6] Gauthier, C.; Campbell, P. G. C.; Couture, P. Ecotoxicol. Environ. Saf. 2009, 72, 2066. [CrossRef]
- [7] Hirsch, R.; Ternes, T.; Haberer, K.; Kratz, K-L. Sci. Total Environ. 1999, 225, 109. [CrossRef]
- [8] Ternes, T. A.; Meisenheimer, M.; Mcdowell, D.; Sacher, F.; Brauch, H. -J.; Haist-Gulde, B.; Preuss, G.; Wilme, U.; Zulei-Seibert, N. *Environ. Sci. Technol.* 2002, *36*, 3855. [CrossRef]
- Joksimovic, D.; Tomic, L.; Stankovic, A. R.; Jovic, M.; Stankovic, S. *Food Chem.* 2011, 127, 632. [CrossRef]
- [10] De Jonge, M.; Teuchies, J.; Meire, P.; Blust, R.; Bervoets, L. Water Research 2012, 46, 2205. [CrossRef]
- [11] Peng, J.; Song, Y.; Yuan, P.; Cui, X.; Qiu, G. J. Hazard. Mater. 2009, 161, 633. [CrossRef]
- [12] Castro, G. R.; Padilha, C. C. F.; Rocha, J. C.; Valente, J. P. S; Florentino, A. O.; Padilha, P. M. *Ecletica Quim.* 2005, 30, 45. [CrossRef]
- [13] Romão, L. P. C.; Castro, G. R.; Rosa, A. H.; Rocha, J. C.; Padilha, P. M.; Silva, H. C. Anal. Bioanal. Chem. 2003, 375, 1097. [Link]
- [14] Stumm, W. Chemistry of the solid-water interface: processes at the mineral-water and particle-water interface in natural systems. John Wiley & Sons: New York, 1992.
- [15] Mudroch, A.; Azcue, J. M.; Mudroch, P. Manual of physico-chemical analysis of aquatic sediments. CRC Press/Lewis publish: New York, 1997.
- [16] Quevauviller, P.; Ure, A.; Muntau, H.; Griepink, B. Int. J. Environ. Anal. Chem. 1993, 51, 129. [CrossRef]
- [17] Tessier, A.; Campbell, P. G. C. Anal. Chem. 1979, 51, 844. [CrossRef]
- [18] Ure, M.; Thomas, R.; Littlejohn, D. Int. J. Environ. Anal. Chem. 1993, 51, 65. [CrossRef]
- [19] Yang, Z.; Wang, Y.; Shen, Z.; Niu, J.; Tang, Z. J. Hazard. Mater. 2009, 166, 1186. [CrossRef]
- [20] Lopretto, E. C.; Tell, G.; Ecosistemas de aguas continentales: metodologias para su estudio. Ediciones Sur: La Plata, 1995.

- [21] Merritt, R. W.; Cummins, K. W. An introduction to the aquatic insects of North America, 3<sup>a</sup> ed. Kendall/Hunt Publ: Dubuque, **1996**.
- [22] Pennak, R. W.; Freshwater invertebrates of the United States, second ed. John Wiley & Sons: New York, 1978.
- [23] Segura, R.; Arancibia, V.; Zúñiga, M. C.; Pastén P. J. Geochem. Explor. 2006, 91, 71. [CrossRef]
- [24] Minello, M. C. S.; Paçó, A. L.; Castro, R. S. D.; Caetano, L.; Padilha, P. M.; Ferreira, G.; Martines, M. A. U.; Castro, G. R. Fresenius Environ. Bull. 2010, 19, 2210.
- [25] Förstner, U.; Salomons, W. Environ. Technol. Lett. 1980, 1, 494. [CrossRef]
- [26] Kazi, T. G.; Jamali, M. K.; Kazi, G. H.; Arain, M. B.; Afridi, H. I. Anal. Bioanal. Chem. 2005, 383, 304. [CrossRef]

- [27] El Samrani, A. G.; Lartiges, B. S.; Ghanbaja, J.; Yvon, J.; Kohler, A. Water Research. 2004, 38, 2063. [CrossRef]
- [28] Zhuang, Y.; Allen, H. E.; Fu, G. Environ. *Toxicol. Chem.* 1994, 13, 717. [CrossRef]
- [29] Gupta, M.; Paliwal, A. Adv. Bio Res. 2010, 1, 71. [Link]
- [30] Klemm, D. J.; Biota of freshwater ecosystems. Identification Manual n°. 8: Freshwater Leeches (Annelida:Hirudinea) of North America. EPA Project: 18050 ELD 1972.
- [31] Slepukhina, T. D. Hidrobiologia. 1984, 115, 183. [CrossRef]