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Integrative Assessment of Sediment Quality in the São Francisco River (Mina Gerais, Brazil)

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Abstract: The São Francisco River (one of the most important South American rivers) has many contamination sources, but just a few environmental assessments have been conducted. A weight-of-evidence approach identified the pollution sources (industrial activities, mineral processing, fisheries, and tourism) in the river and the city of Três Marias based on two different lines of evidence: the structure of the benthic community (biological monitoring working party score system, abundance of taxa, number of individuals, Margalef species richness, Pielou evenness, and Shannon–Wiener diversity) and the physicochemical determination of sediments (%fines, TOC, nitrate, ammonium, ammonia, ammoniacal nitrogen, metalloids, and SEM/AVSs). The results show that the wastewater treatment plant was the most important source of pollution. A factory was also detected as a source of contamination, with related adverse effects having been measured downstream. Other sources of contamination and stress were detected in the studied area. The macro-benthic identification study identified three different sentinel species (*Tanytarsus* sp., *Crytochironomus* sp., and *Polypedilum* sp.) for future monitoring assessments of the sediment quality in riverine areas. Thus, an improvement in the management of river effluents and more measures focused on cutting contaminant emissions from the waste treatment plant are recommended.

Keywords: sediment assessment; weight of evidence; SIMPER analysis; biological monitoring working party

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1. Introduction

The São Francisco River (SFR) is one of the most important Brazilian and South American rivers; with a 1800 km length, its source is in Serra da Canastra (Mina Gerais), and it drains 641,000 km^2 across different states (Bahía, Pernambuco, Sergipe, and Alagoas) until it reaches the Atlantic Ocean. It also acts as a geographical barrier. Some other important rivers are effluents, such as the Paraopeba River, das Velhas River, or Verde Grande River. This river plays an important economic role due to electricity generation and transportation of goods; nevertheless, some other activities strongly influence it, such as zinc metallurgy, sewage discharges, and agriculture, among others [1]. A few environmental assessments have been performed in this area [1–6].

Sediments are an essential component of aquatic ecosystems and should be considered a resource of high ecological and socio-economic value that requires appropriate protection and management [7]. Sediments in a river could act as a sink of pollutants, releasing them into the waters when the sediment accumulation capacity is saturated [8].

Ecological monitoring using benthic community structure analysis provides information on different changes related to species composition, abundance, and diversity; all these indicators show alterations in the communities in the studied areas [9–11]. This kind of study provides realistic information about the environmental health of aquatic ecosystems. The macro-benthic fauna plays a fundamental role in sediment processes, providing an important measure of the response of a community to environmental perturbation [12–16] and exhibiting the greatest potential for monitoring conditions at a particular site [17]. Biotic indices are useful tools in decision-making processes [18], and those indices synthesize complex scientific data. The structure of the macro-benthic community was analyzed using univariate measures (biological monitoring working party score system, the abundance of taxa, the number of individuals, Margalef species richness, Pielou evenness, and Shannon–Wiener diversity). The biological monitoring working party (BMWP) score system is very useful for water quality management due to its adaptability. However, the use of biotic indices has limitations, since natural variability can be difficult to establish. Therefore, reliable environmental assessment studies must integrate as much information as possible.

Another line of evidence (LOE) was selected in order to establish the cause of potential alteration and/or pollution. This LOE is based on the physicochemical determination of sediments: grain size (fines), organic carbon content (TOC), chemical nitrogen (nitrate, ammonium, ammonia, and ammoniacal nitrogen), metal concentrations in sediments (Al, As, Co, Cr, Cu, Fe, Ni, Pb, Zn, and Hg), and simultaneously extracted metals–acid volatile sulfides (SEM-AVSs). The USEPA [19] establishes some benchmarks for metal(loid)s Cd, Cu, Pb, Ni, Ag, and Zn in freshwater sediments. Ref. [3] classified the toxicity of the sediments according to benthic organisms in total organic carbon fraction (f_{toc}), but also additional elements or factors affecting the area, such as chemical nitrogen or other metals. Therefore, chemical nitrogen was analyzed mainly due to the sewage discharge and agricultural pollution in the Barreiro Grande stream. Furthermore, the concentrations of Al, As, and Fe were measured due to the proximity of the VM-TM refining factory.

The main objective of this study was to establish the sediment quality in this river, addressing the main causes of the potential alteration and/or pollution with an integrative assessment. It was conducted by linking the results of the LOEs of sediment contamination and the descriptive parameters of macro-benthic community. Furthermore, the pollution sources in the upper-middle region of the São Francisco River were located and identified using a weight-of-evidence (WOE) approach based on the integration of both lines of evidence in order to define a management plan approach to determine the best remedial actions for improving the ecological status of the river.

2. Materials and Methods

2.1. Study Area

The area of study (Figure 1) was located between the Três Marias reservoir (SF1), and around 300 m downstream of the effluent discharge point a refining factory (ZM2). Therefore, the region is affected by anoxic water entering from the reservoir, the zinc-refining factory, and human alterations, such as sewage effluents from the Barreiro Grande stream and other diffuse sources. The upper sampling station (SF1) was located on the left bank of the SFR, and upstream of the factory and the confluence with the Barreiro Grande stream. This station was selected as control in the study. Downstream, the BG4 station was located in the Barreiro Grande stream 10 m upstream of the SFR confluence and was selected to determine the sediment characterization before the confluence with the SFR and the effects of the Três Marias wastewater treatment plant (WWTP). In the distance, the SF2 station was located on the right bank of the SFR, just opposite the security dam, in order to determine the sediment quality in the proximity area of the dam. On the right bank of the SFR, the SF3 station was located 100 m downstream of the effluent discharge site of the factory, to assess the direct impact of the factory on the SFR sediments. Upstream, the ZM2 station was located in the mixing zone of water from the SFR, just on the right bank of the SFR, 300 m downstream of the effluent discharge site of the factory and 20 m downstream of the 'Consciência' stream confluence. This station was selected to study the potential contamination associated with factory activity.

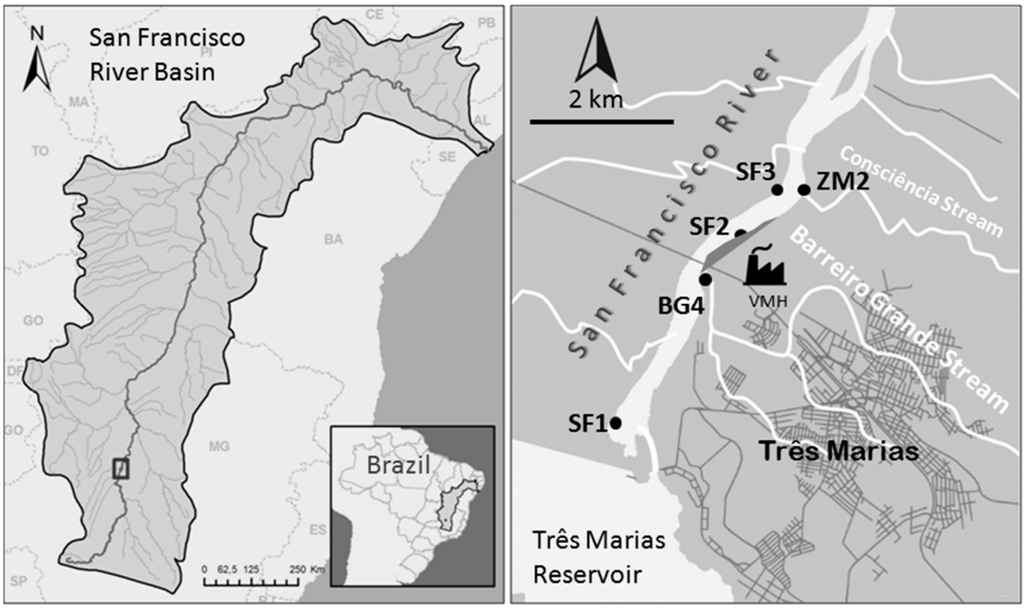

Figure 1. Map showing São Francisco River basin extension (Brazil) and study area with sampling stations (SF1, BG4, SF2, SF3, and ZM2). Some of the sources of contaminants, such as a factory, the city of Tres Marias, and the security dam (in dark grey), are also included.

2.2. Sampling Survey and Characterization

Sediment samples were collected with a Petersen grab of 5 L in capacity at a 7 cm depth at the sampling stations (SF1, BG4, SF2, SF3, and ZM2) (Figure 1). The samples were transported to the laboratory, where they were analyzed for physicochemical characterization. Samples for benthic analysis were separately collected; benthic animals were retrieved with the help of a 0.25 mm sieve and fixed in formalin (4%).

Grain size distribution (% fines) and total organic carbon (TOC) were analyzed using the ABNT NBR 7181/82 procedure [20]. This standard involves the drying, sieving, and sedimentation of the fines. Briefly, 500 g of each sample was dried at 50 °C for 48 h. The dried material was disaggregated in a mortar and sequentially sieved with 0.85 mm, 0.5 mm, 0.25 mm, 0.125 mm, and 0.063 mm meshes. The 0.063 mm fraction was introduced in 1000 mL beakers, and the sedimentation of fines was determined by pipetting/weighing the sediment over time.

Chemical nitrogen forms (nitrate, ammonium, ammonia, and ammoniacal nitrogen) were also measured in sediment samples to analyze diffuse contamination, a common example of which is the leaching of nitrogen compounds from fertilized agricultural lands. Nitrate, ammonium, ammonia, and ammoniacal nitrogen were determined in sediment interstitial waters. Sediment samples were centrifuged at 11,000 rpm and 15 °C for 15 min using an Eppendorf centrifuge. Afterward, the supernatant was filtered with a 0.6 µm pore fiberglass filter (Whatman) and analyzed with ion-exchange chromatography using a DIONEX DX-80 ion analyzer [21].

The methodology used for the analysis of secondary sulfides from sediments followed the procedures of the AVS-SEM technique [22]. In short, a sample of whole sediments (moist) was weighed and introduced into the reaction flask along with 100 mL of Milli-Q water. The flask was sealed, and the suspension was stirred for ultrapure nitrogen flow. Then, 20 mL of previously aeriated HCl (6M) was introduced with nitrogen. The AVSs were purged by the carrier gas (N_2) through the glass tubes that were immersed within the receiving flasks for the time of 30 min. The remaining suspension of the balloon (SEM) was filtered for the determination of metals (Cu, Cd, Pb, Ni, and Zn).

Metal and metalloid concentrations (Al, As, Co, Cr, Cu, Fe, Ni, Pb, Zn, and Hg) were determined using inductively coupled plasma–atomic emission spectroscopy (ICP-AES; Jobin-Ybon Ultima2) using a modification of the USEPA 3050B methodology [23]. The modification was performed to prevent the evaporation loss of the volatile trace elements (Hg, As, and Se). Standards were prepared with SCP SCIENCE standards. Reference certificated material SEM-1640 NIST for freshwater samples and inter-laboratory standard IRMM-N3 were also analyzed. The detection levels were calculated considering half of the signal and the standard deviation of a blank calculated 10 times.

2.3. Benthic Community Analysis

A total of 25 sediment samples were collected with a 0.0928 m^2 Petersen grab. Five replicate samples were taken at each station for benthic faunal analysis. After collection, samples were sieved in situ with a 0.025 mm mesh, and the retained organisms were transported to the laboratory in buffered formalin (4%). There, these samples were washed and transferred to 70% isopropyl alcohol. Then, they were sorted and identified to the lowest taxonomic level.

We used the biological monitoring working party (BMWP) score to study the tolerance of the benthic community. It is based on the principle that different aquatic invertebrates have different tolerance to pollutants, especially organic (i.e., nutrient enrichment that can affect the availability of dissolved oxygen). The presence of mayflies or stoneflies indicated the best water quality, with a score of 10, while worms indicated the worse water quality, with a score of 1 [24].

Moreover, the Shannon–Wiener diversity (H′), species richness (d′ [25,26]), Pielou evenness (J′ [27]), total abundance (A), and abundance of taxa (S) classic indices were calculated using the PRIMER-E (Plymouth Routines in Multivariate Ecological Research, v6) [28,29] suite of computer programs developed at Plymouth Marine Laboratory, UK.

2.4. Statistical Approach

Univariate analyses were based on the community descriptive parameters of the benthic fauna and the physicochemical measurements, which were calculated for each replicate sample and summarized for each site. The results were expressed as arithmetic averages ± standard deviations. In order to determine whether there were significant differences among stations, one-way analysis of variance (ANOVA) of each benthic univariate measure and physicochemical analysis result was used, followed by the multiple comparisons of Dunnett's test. The significance was set at $\alpha < 0.05$. Univariate statistical analyses were performed using SPSS 17.0.

Data were previously checked and/or transformed to ensure normality and heteroscedasticity and conduct the ANOVA test. For multivariate analyses, species abundance was transformed with the fourth root prior to performing non-metric multidimensional scaling (MDS) and similarity percentage analysis (SIMPER). Relative similarities in the abundance or biomass of benthic fauna among treatments (plus the reference samples or D0) were graphically analyzed using MDS with the Bray–Curtis similarity index [29]. The reliability of the MDS representations of the assemblage similarities was assessed with their stress values; stress < 0.01 was considered acceptable [28]. Clustering was performed using a hierarchical, agglomerative method using group average sorting, the results of which are displayed in a dendrogram. SIMPER analysis of abundance (cut-off percentage: 90%) was used to identify the taxa with the greatest contribution to differences among samples [28]. The significance of differences among stations was tested using the randomization/permutation test ANOSIM [30]. All analyses in MDS, SIMPER, and BIOENV were performed using PRIMER 6.1.6.

3. Results

3.1. Physicochemical Analyses

The summarized results of the physicochemical parameters analyzed are shown in Table 1. The BG4 station registered the greatest concentrations of compounds (nitrate, ammonium, ammonia, and ammoniacal nitrogen). However, the greatest element concentrations of Fe and Cr were found at SF1; those of Cu, Pb, and Zn, at SF3; and those of Co and Ni, at SF2. The ZM2 station displayed the highest concentrations of Al and As. The concentrations of metals Pb, Zn, Hg, and Cu showed a smooth gradient that increased from station SF1 as follows: SF1 < SF2 < SF3 < ZM2. Station ZM2 was located in the area of mixing between the SFR and the heavily factory-influenced stream, which could justify the high values of Al and As.

Table 1. Summarized results of physicochemical parameters analyzed in sediments from the San Francisco River (SFR): percentage of fines, total organic content (TOC), nitrogen species (nitrate, ammonium, ammonia, and ammoniacal nitrogen), SEM-AVSs, and metal(loid) concentrations (Al, As, Co, Cr, Cu, Fe, Ni, Pb, Zn, and Hg).

	SF1	**BG4**	**SF2**	**SF3**	**ZM2**
Fines (%)	7.37	50.16	26.28	37.82	27.81
TOC (μg/g)	20,970	10,590	11,960	12,430	17,590
Nitrate (μg/L)	83.46	617.32	50.87	24.52	35.28
Ammonium (μg/L)	1511	8760	1210	1432	1424
Ammonia (μg/L)	33.99	35.18	4.86	8.65	8.59
Ammoniacal nitrogen (μg/L)	1545	8795	1215	1441	1432
SEM-AVS (μmol/g)	58.7	3687	2720	2947	96.1
Al (μg/g)	2197	3993	2596	4558	32,767
As (μg/g)	5.51	1.36	1.48	2.56	8.83
Co (μg/g)	6.83	3.48	8.36	8.27	5.91
Cr (μg/g)	15.78	10.55	14.63	11.99	12.50
Cu (μg/g)	4.76	14.50	21.19	43.64	16.41
Fe (μg/g)	22,017	13,059	8030	14154	12,307
Ni (μg/g)	5.15	3.56	5.64	4.56	4.71
Pb (μg/g)	4.28	14.5	36.7	132	15.3
Zn (μg/g)	25.1	958	1239	3676	639
Hg (μg/g)	0.02	0.02	0.09	0.17	0.12

Due to the placement of BG4, situated on the Barreiro Grande stream, where the Três Marias water treatment plant discharges, the nitrogen species concentrations (nitrate, ammonium, ammonia, and ammoniacal nitrogen), well known as domestic waste indicators, were higher than the values obtained at the other stations. On the other hand, the sediments from the SF1 sampling point, although not directly influenced by the factory, received many contaminants from all over the drainage basin, since this sampling point was located under the Três Marías reservoir. According to the results, we could establish a decreasing pattern related to domestic waste contamination as follows: BG4 > SF1 > SF3 ≈ ZM2 > SF2.

3.2. Benthic Community Structure

Concerning benthic community structure analysis, a total of 3915 individuals were identified in the benthic fauna in sediments from the five sampling stations and along the river. They were classified into 60 species (1 hirudinean, 12 oligochaetes, 1 crustacean, 43 insects, 3 mollusks, and 1 nematode), and 4 species were not fully identified (17 *hirudinean* annelids, 14 *conchostracan* crustaceans, 2 *dryiopidae* beetles, and 63 individuals classified as *nematode*). Figure 2 shows a relative abundance of different taxonomic groups (hirudinea, oligochaeta, mollusca, and nematoda) observed at each sampling station. Therefore, the benthic community at BG4 clearly differed from the rest of the stations; it was dominated by nematodes (around 68%), unlike the communities from the rest of the stations, where insects were the dominant group (60–77%). Relative nematode abundance varied from 5% (at the SF1 station) to 0% (at the SF3 and ZM2 stations), and relative insect abundance was 2% at the BG4 station.

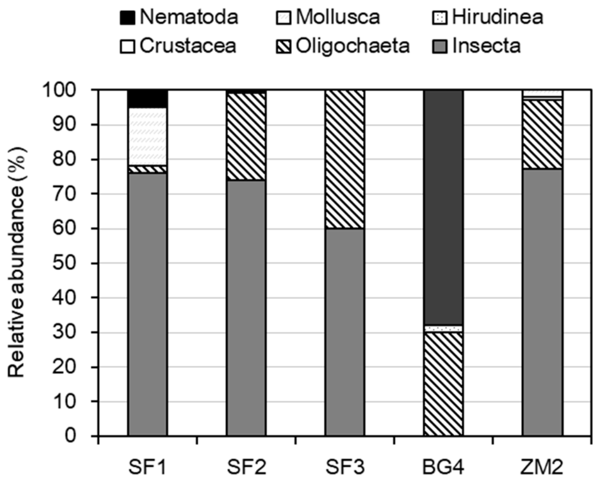

Figure 2. Relative abundance of different taxonomic groups (Hirudinea, Crustacea, Insecta, Oligochaeta, Mollusca, and Nematoda) identified in sediments from each sampling station at the São Francisco River (SF1, SF2, SF3, BG4, and ZM2).

The univariate metrics of the community structure for all the sampling stations are shown in Table 2. According to the BMWP results, all stations showed a bad ecological status. The BMWP score showed significant ($p < 0.05$) differences among sampling sites (BG4 and SF2), close to the affluent, and the control site (SF1). As expected, the SF1 site presented the best BMWP results, which were still considered 'bad' but just at the limit between degraded and regular. Conversely, BG4 showed the lowest BMWP score, defined by the abundance, number of individuals, richness, and diversity values, and the SF2 station showed the lowest Pielou evenness values. The benthic community structure at the SF2 station showed the highest Pielou evenness, while BG4 showed the lowest values.

Table 2. Mean and standard deviation (mean ± SD) values of different biotic indices (abundance of taxa—S′; number of individuals—N′; Margalef species richness—d′; Pielou evenness—J′; Shannon–Wiener diversity—H′; and BMWP score) per square meter at each sampling station.

Index	SF1		BG4		SF2		SF3		ZM2	
S′	3.60	±3.65	0.60	±0.89	6.20	±1.30	6.40	±1.14	4.60	±2.70
N′	185	±245	94.3	±206	896	±667	366	±144	1459	±2588
d′	0.51	±0.62	0.08	±0.12	0.79	±0.20	0.93	±0.21	0.57	±0.28
J′	0.82	±0.33	0.88	±0.00	0.65	±0.14	0.72	±0.12	0.69	±0.25
H′	0.79	±0.84	0.12	±0.27	1.16	±0.24	1.33	±0.32	0.87	±0.28
BMWP	45.7	±2.5	23.3 *	±1.75	25.6 *	±2.14	28.13	±5.02	27.87	±3.29

Asterisks (*) indicate significant differences ($p < 0.05$) compared with the control site (SF1).

Figure 3 shows the results of clustering the different stations using the Bray–Curtis index for abundance similarities. Cluster analysis distinguished three main groups of sites (BG4, SF1, and the rest of the sites), which were considered to represent different communities on the basis of their geometric means. As mentioned above, BG4 was clearly different from the other stations, and the control site (SF1) was different from the other stations but closer than BG4. The SF2, SF3, and ZM2 stations showed similar communities among them.

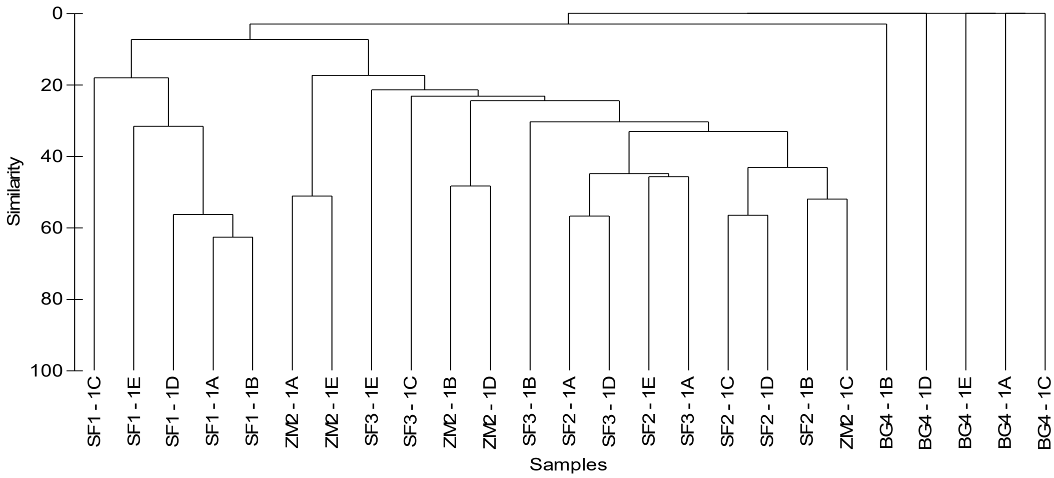

Figure 3. Bray–Curtis similarity dendrogram based on abundance, showing the classification of each station replicates.

The SIMPER analysis showed how the structure of the benthic community contributed to the differences among stations, indicating the main taxa and their contribution to the dissimilarity. The abundance of the macrofauna showed the highest dissimilarity (100%) between the SF3 and BG4 stations, while the least dissimilar (69.38%) stations were SF2 and SF3. Moreover, BG4 showed large differences (or dissimilarities) from the rest of the stations: BG4-ZM2 (99.52%), BG4-SF1 (99.33%), and BG4-SF2 (98.77%). The abundance of species of *Tanytarsus* spp., *Cryptochironomus* spp., and *Polypedilum* spp. was responsible for the dissimilarities between the most different stations (SF3-BG4 and ZM2-BG4), showing dissimilarities of around 8.37–13.33 (*Tanytarsus* spp.), 6.44–11.8 (*Cryptochironomus* spp.), and 5.25–11.63 (*Polypedilum* spp.). Pairwise comparisons derived from the ANOSIM test on species abundance data showed that the stations were significantly different from each other ($p < 0.01$).

3.3. Linking Physicochemical and Benthic Community Results

The relation between benthic fauna abundance and physicochemical variables was studied using the BIOENV procedure (Table 3). This statistical analysis selects environmental variables 'best explaining' community patterns by maximizing the correlations among their respective similarity matrices. This approach is advantageous, since it identifies not only the best correlation but also the best combination of them.

Table 3. BIOENV correlations (Spearman rank correlation coefficients, r_s) between benthic community structure and environmental variables (n).

n	Variables	r_s
5	Nitrate + ammonium + ammonia + Al + Zn	0.915
5	Nitrate + ammonia + ammoniacal nitrogen + Al + Zn	0.915
5	Ammonium + ammonia + ammoniacal nitrogen + Al + Zn	0.915
1	Nitrate	0.879
2	Nitrate + SEM-AVSs	0.879
3	Nitrate + ammonium + SEM-AVSs	0.879
2	Nitrate + ammonium	0.879
2	Nitrate + ammoniacal nitrogen	0.867
5	Nitrate + ammonium + ammonia + ammoniacal nitrogen + SEM-AVSs	0.867

The results of the BIOENV analysis showed that 91.5% of the variance in the different stations was due to the nitrate, ammonium, ammonia, Al, and Zn contents; the nitrate, ammonia, ammoniacal nitrogen, Al, and Zn contents; or the ammonium, ammonia, ammoniacal nitrogen, Al, and Zn contents. This analysis always showed Al and Zn as the contaminant metals of concern in the analyzed macro-benthic community. This variance was lower (87.9%) when one (nitrate), two (nitrate and SEM-AVSs), or three variables were considered (nitrate, ammonium, and SEM-AVS; or nitrate, ammoniacal nitrogen, and SEM-AVS). These results associate the nitrogen species with those measured changes in the macro-benthic community structure.

4. Discussion

This study identified the degradation in the surrounding area of the SFR course using the chemical measurement of sediments and macro-benthic community structure. This area, located between the Três Marias reservoir and around 300 m downstream of the effluent discharge point of the refining factory, showed several sources of contamination. In this kind of situation with multiple stressors, it is necessary to assess the overall sediment quality, and the use of multiple lines of evidence with an integrative approach is recommended [31–36]. The use of these multiple lines of evidence integrated with a weight-of-evidence approach (WOE) links the results obtained in each LOE, in this case, physicochemical parameters and benthic community structure indexes. Metal contamination has been extensively studied, but metal bioavailability and its effects are not commonly studied in this region [1–3,37,38]. The environmental state of the SFR was evaluated in this study using different lines-of-evidence (LOEs), which were integrated using the BIOENV procedure.

Benthic infaunal communities, as living components of sediments, represent the integrated response of the biological effects of pollutant contents on sediment samples. The results obtained using the classical descriptive parameters show the lowest values of Margalef richness (0.08) and Shannon–Wiener diversity (0.12), as well as the BMWP score system (23.34), at the BG4 station. Moreover, this station also showed the highest values of Pielou evenness (0.88). Furthermore, this station showed a significant ($p < 0.05$) difference in the macro-benthic community structure results compared with those obtained for the other stations. Thus, the highest descriptive parameters were observed at different stations, showing, at SF3, the highest values of the abundance of taxa (6.40), Margalef richness (0.93), and Shannon–Wiener diversity (1.33). Significant differences ($p < 0.05$) were only found in the BMWP score results obtained at the BG4 and SF2 stations, which can be observed in the MDS analysis (Figure 3). Furthermore, the SIMPER analysis showed two of three species (*Polypedilum* sp. and *Tanytarsus* sp.) that were classified as sensitive by [39], and the *Tanitarsus formosanus* species, which was classified as sensitive [40–42]. Additionally, the other marked species (*Cryptochironomus* sp.), according to [43,44], typically prefers well-oxygenated sandy substrates and is generally intolerant to polluted conditions. [*Polypedilum trigonous* species showed a positive relationship with most of the organic pollutants, such as TOM, ammonium, and nitrateas well as trace metals Zn, Cu, and Mn [45]. Ref. [41] reported that *Polypedilum* sp. was very tolerant to metal contamination.

Despite the use of biotic indices having some limitations, Ref. [3] used the BMWP score system in this area with good results. Moreover, according to this index, the analyzed area of the SFR showed a 'low' environmental quality classification, except for the station located upstream of the factory, which showed a 'regular' environmental quality classification. Based on these results, it can be considered that the factory exerts negative effects that are reflected in downstream stations. However, the BG4 station, which showed the worst quality classification, was located near the Três Marias WWTP and far away from the influence of the mining factory. The community at this station seemed to be significantly affected ($p < 0.05$) by this plant because of the discharge of urban waste, presenting a very different specific composition (abounding nematode and oligochaete species). Indeed, these species are well-known pollution bioindicators, and WWTPs can deposit large amounts of organic matter and nutrients into receiving waterways, leading to eutrophication [46] and temporary oxygen deficits [47].

The BMWP score system itself, or a modified version of this index, has been previously applied in other countries in order to study the water quality of rivers. The results in [48] show significant differences between quite unpolluted areas and anthropogenically impacted sites along the Nysa Klodzka River (Poland), as well as high correlations with some chemical variables. The modified version of the IBMWP (Iberian biomonitoring working party) score was successfully applied by [49] to 50 Iberian rivers, showing strong correlations between this index and different physicochemical parameters. Both studies show results and relationships similar to those reported in the present work.

Sediment quality assessment has demonstrated the importance of sediment characterization in terms of organic matter, grain size, metals [50–53], and SEM-AVS-SEM [54] in determining the environmental quality conditions, mainly in heavily populated and industrialized regions. The results here obtained related to sediment characterization show three main different groups of findings: (i) station SF1 presented the lowest values of fines, SEM-AVSs, Al, Cu, Pb, Zn, and Hg, but the highest values of TOC, Cr, and Fe; (ii) station BG4 showed the lowest values of TOC, As, Co, Cr, and Ni, but the highest values of fines, nitrate, ammonium, ammonia, ammoniacal nitrogen, SEM-AVSs, and Al; (iii) the other two stations, SF3 and SF2, showed the highest Cu, Pb, Zn, and Hg concentrations, and the highest Co and Ni concentrations, respectively. As reported by [55], the nitrate values found in the Hindon River, which is also polluted by wastewater, were in the range of 96–245 μg/L, i.e., lower values than those found in sediments from the BG4 station. Many rivers around the world are suffering contamination by industrial, agricultural, and domestic activities. The case study on the Axios/Vardar River in Southeastern Europe exposed the results of the water quality in an area surrounding a metal factory that receives domestic wastewater and agricultural runoff. In this case, the maximum values of nitrates and ammonium were 0.22 mg/L and 1.13 mg/L, respectively, which are again lower than those found in this study. The metal concentrations in that river were Pb (13.126 μg/L), Zn (49 μg/L), Cd (0.72 mg/L), and Cr (64 μg/L), indicating that it was fairly less polluted than the river studied here (SFR).

Taking into account the SEM-AVS results, all the sampling stations showed potential alterations in benthic organisms [19], showing SEM-AVS values higher than 0. Station BG4, had the highest value (3687.98), also had the highest alteration values of the calculated quality indices based on the benthic community structure (BMWP, S′, N′, d′, H′, and J′), as mentioned above. Moreover, according to the benchmarks of [3], the toxicity of our sediment samples was classified as unlikely (SF1 and ZM2 stations), uncertain (SF2 and SF3 stations), or likely (BG4 station).

The highest concentrations of nitrogen species were analyzed in sediments from station BG4, which was located downstream of the Três Marias WWTP. These results are in concordance with those reported by [56]. These authors concluded that wastewater contains significant amounts of ammonium, as only a small proportion is oxidized by conventional treatment plants [57]. Furthermore, as exposed by [58], sediments that are rich in organic matter and microbial biomass host the greatest number of denitrification processes, since they require sub-oxic conditions and a source of nitrate (from external inputs or the nitrification of ammonium).

The link between cause (sediment contamination) and effect (macro-benthic community structure) was determined using BIOENV analysis. The obtained results indicate that a large proportion of the among-site variance in the abundance of benthic fauna along the studied river course was strongly associated with the measured environmental variables, particularly, nitrogen chemical species and metals Al and Zn. Changes in benthic fauna abundance and diversity were related to biogeochemical transformations, such as nitrification and denitrification, and thus to the impacts of stress and disturbance on microbial communities. Much research on bioindicators in aquatic systems has been limited to heterotrophic bacteria in relation to the decomposition of dissolved organic matter and as a measure of sewage pollution [59,60]. In the same way, BIOENV analysis indicated changes related to Al and Zn contamination. These metals were at their highest concentrations at the BG4 and SF3 stations. In this sense, these adverse effects may be provoked by other sources of contaminants not measured in this study (e.g., by the influx of oxygen-depleted water or sewer discharge), as exposed by [61].

The biology/ecology status clearly reflects the environmental health of a river. However, apart from contamination [18], there are other problems associated with the basin. Water scarcity in the SFR basin has been detected as a significant base-flow reduction [62]. Further, a recent study on the SFR basin [63] assessed the influence of dams on eco-hydrological conditions, and the socio-economic impact due to the transfer of water from the SFR [64]. The study also highlighted that the intensification of natural resources provokes impacts on land and water resources, as also shown by the current study. Therefore, hydrological management plans should include environmental monitoring programs for river quantity and quality, especially because the Brazilian government is promoting the expansion of irrigated agriculture [63] and due to the increasing population and other associated overexploitation problems. Therefore, the SFR basin presents several problems that are tackled by the United Nations Sustainable Development Goals, such as clean water, poverty, health and wellness, and the distribution of equality [65].

Based on the obtained results, different approaches to remediation and/or mitigation should be addressed in this area. The integration of the two LOEs used in this study suggests that the environmental risk present in the area is associated with different sources of contamination (WWTP, metal factory, and reservoir). The monitoring of the risk identified in the area studied (between the reservoir and the metal factory) is highly recommended.

5. Conclusions

This study characterized the contamination in the middle area of the San Francisco River (SFR) by linking contamination and macro-benthic community structure results of the sediments studied. The main stressor identified and characterized in the area was associated with the activities of the WWTP and, to a lower extent, with the mining factory. ZM2, which was located in the mixed zone between the SFR and the stream produced by the metal factory, showed the highest metal concentrations. BG4, situated on the Barreiro Grande stream where the Três Marias water treatment plant discharges, showed the highest nitrogen species concentrations, but also presented a particular specific community composition (abounding nematode and oligochaete species) related to the discharge of urban waste.

The study identified the macro-benthic community structure as a valuable tool for monitoring the environmental quality of riverine sediments. Thus, we determined three different species (*Tanytarsus* sp., *Crytochironomus* sp., and *Polypedilum* sp.) as useful bio-indicator species for monitoring plans.

The links between cause (sediment contamination) and effect (community structure) showed that a large proportion of the variance in the abundance of benthic fauna along the studied river course was strongly associated with the measured contaminants, especially the nitrogen chemical species, and Al and Zn, related to the WWTP and the mining factory, respectively. The monitoring plan for the area needs to be designed using variables similar to those used in this study. Potential new LOEs such as sediment toxicity could be considered in the WOE approach.

This kind of study (integrative assessment) could allow the identification of the sources of contamination, the localization of the areas affected by pollution, and the characterization of sediment quality to be conducted. Furthermore, integration helps in the identification of the risk associated with different human activities and consequent pollution. Finally, it could be an input to the potential decision to perform remedial and/or mitigating actions to improve the environmental quality of the river.

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writing—review and editing, visualization, supervision, project administration, and funding acquisition. All authors have read and agreed to the published version of the manuscript.

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