Water Air Soil Pollut (2009) 198:317–334
DOI 10.1007/s11270-008-9848-z

Effects of Heavy Metal Contamination (Cr, Cu, Pb, Cd) and Eutrophication on Zooplankton in the Lower Basin of the Salado River (Argentina)

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Received: 24 October 2007 / Accepted: 6 September 2008 / Published online: 28 November 2008

Abstract The effects of heavy metal contamination (Cr, Cu, Pb, Cd) in the lower basin of the Salado River (Argentina) were studied on the zooplanktonic community. The determination of heavy metals in water and sediments was carried out in a previous study. Zooplankton was analyzed quali- and quantitatively. Total density, by-group density (Copepoda, Cladocera and Rotifera), micro and mesozooplankton density, biomass, species richness (*S*), and species diversity (*H*) were studied. The results showed that total density of zooplankton was significantly higher in the river than in the channels and streams ($p<0.001$), with dominance of rotifers but a higher copepod biomass. Calanoida dominated over Ciclopoidea and Harpacticoida. Total species richness was 74, showing the highest values (59 and 56) at the points corresponding to the Salado River at localities Manucho and San Justo (MSR, SJSR) and the lowest ones in North and South channels (NCH, SCH), with 16 and 17 species, respectively), and in the two sampling stations of Las Prusianas stream (LP1, LP2), between 13 and 38 species. The species diversity showed low values (1.8 to 2.3) in channels and streams, and higher values (3.0) in the Salado River, at Manucho and San Justo. Absolute biomass varied in the order SJSR>MSR>LP1>NCH>SCH>LP2, similarly to absolute density, which varied in the order SJSR>MSR>LP1>NCH>SCH>LP2. The comparison of the content of heavy metals in water between the control site (SJSR) and the most contaminated sites showed significant differences with the North and Las Prusianas 1 and 2 channels (ANOVA $p=0.001$; 0.012 and 0.011, respectively) and non-significant differences, although close to the significance level, with the South Channel and Manucho ($p=0.08$; $p=0.059$). The following positive correlations were found: depth with mesozooplankton density, *H* and *S* ($p<0.001$); temperature with microzooplankton density, *H* and *S* ($p<0.004$), and dissolved oxygen with mesozooplankton density, *H* and *S* ($p<0.01$), but not with microzooplankton, indicating a higher tolerance of the organisms belonging to this fraction. A negative correlation was found between biomass of copepods and concentration of Pb and Cu ($p<0.05$ and $p=0.01$, respectively). Rotifers were the most tolerant to heavy metal contamination, followed by copepods and cladocerans. Diversity (*H*) and richness (*S*) were good indicators of stress of contaminated systems. The

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clustering of biological variables and the concentration of heavy metals in water and sediments showed three groups of environments: the first one was the main course of the river, with lower contamination by heavy metals and higher density, biomass, *H* and *S*, which separated clearly from the other two groups of the tributaries, composed by channels and streams. In the tributaries, *r* strategists and a few tolerant species, such as *Eucyclops neumani*, proliferated. The results of this study show that zooplankton responds as good descriptor of water quality, constituting an efficient tool to assess heavy metal contamination.

Keywords Argentina · Heavy metals (Cr, Cu, Pb, Cd) · Tanneries · Zooplankton

1 Introduction

In zooplanktonic populations, changes in demographic parameters can occur as a response to chemical disturbances, although the direct and indirect effects on communities and other trophic levels will depend on the interaction among physical, chemical and biological factors and on the time of exposure to the toxic. The place of "intermediary" that zooplankton occupies in the trophic structure of aquatic ecosystems, with a key role in the transference of energy between primary producers and the other levels of consumers, makes these organisms important indicators of the general functional conditions of aquatic ecosystems.

The responses of zooplankton to anthropic disturbances have been specially studied for the Northern Hemisphere (Winner and Farrell 1976; Marshall 1978; Roch et al. 1985; Baudo 1987; Kerrison et al. 1988; Evans and McNaught 1988; Keller and Yan 1991). However, the information on aquatic environments of the Southern Hemisphere is limited. In Argentina, the records are even scarce, in spite of the increase in the disposal of toxic wastes of industrial origin. The basin of the lower Salado River has serious problems of organic and industrial contamination, mainly related to the disposal of tannery effluents with high content of Cr and other heavy metals coming from galvanoplasties and mechanical repair shops located in the area (Gagneten et al. 2007).

Although the structure and composition of zooplankton of the middle Paraná River basin have been studied for more than four decades, the most contaminated ecosystems of this region have not received a similar attention. In this respect, the information on the relationship between zooplankton and contamination of the Salado River basin, its more important tributary in Argentina, includes some records, particularly on its middle and lower sectors (José de Paggi 1983, 1988; José de Paggi and Paggi 1998; Gagneten and Ceresoli 2004).

In this study, we provide the results of the composition and abundance of zooplankton of water courses belonging to the lower Salado River basin with a different degree of contamination by heavy metals. At six sampling sites (five contaminated ones and a reference one), total density, by-group density (Copepoda, Cladocera and Rotifera), micro and mesozooplankton density, biomass, species richness (*S*), equitability (*E*) and species diversity (*H*) were analyzed to study the possible effects of contamination by heavy metals of water and bottom sediments recorded in previous studies.

2 Study Area

Six sampling sites were selected (Fig. 1): the first one in the North Channel (NCH; 31° 12′ S; 61° 27′ W); the second one in the South Channel (SCH; 31° 20′ S; 61° 24′ W); the third and fourth sites in the Las Prusianas Stream, which, in turn, runs into the Cululú Stream, tributary of the Salado River: Site LP2 (31° 23′ S; 61° 15′ W) and Site LP1 (31° 22′ S; 61° 08′ W); the fifth site at the main course of the Salado River: MSR, close to the locality of Manucho, downstream from the locality of Esperanza (31° 23′ S; 60° 53′ W), and the sixth site, SJSR, in the same river, close to the locality of San Justo (31° 15′ S; 60° 53′ W), located 153 km following the river course, upstream from the mouth of the Las Prusianas Stream. This last site was considered the reference site since it was located upstream from the industrialized area.

2.1 Physical and Chemical Parameters

Table 1 shows a summary of the physical and chemical characteristics of water at each sampling site. The data correspond to the environmental characterization and monitoring of heavy metals in

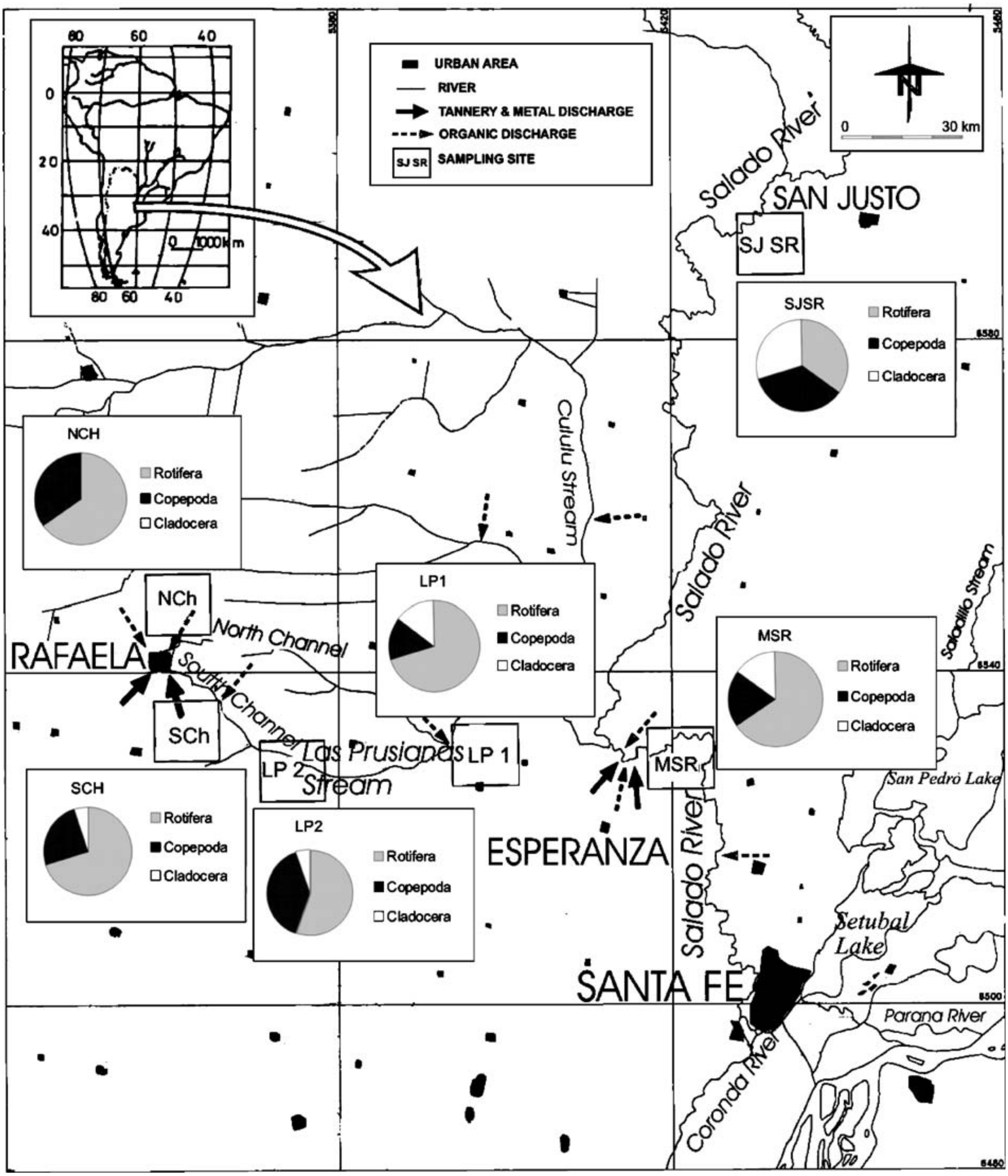


Fig. 1 Map of the Salado river basin showing location of the sampling sites and relative richness of Rotifera, Copepoda and Cladocera recorded at each sampling site

Table 1 General physical and chemical characteristics of water and heavy metals concentrations in water (mg/L) and sediments (μg/g) in each sampling site quoted from Gagneten et al. (2007)

	NCH	SCH	LP 2	LP 1	MSR	SJSR
Ta	23.2±3.8	22.5±2.8	22.7±3.6	18.3±3.1	19.5±3.2	19.6±4.7
Dth	0.41±0.3	0.28±0.05	0.24±0.11	0.64±0.31	3.92±0.34	5.23±0.64
TS	1,267.2±478.4	3,662.7±1531.2	2,158.7±501.3	1,602.2±502.7	2,698±996.8	1,848±354.6
pH	7.7±0.5	7.7±0.4	7.7±0.1	7.7±0.5	7.5±0.9	7.8±0.3
Cnd	1,675±419.3	2,075±189.3	3,650±519.6	5,965±1659.2	3,260±1659.2	2,633.3±378.6
TH	221.3±84.0	502.9±223.6	204.3±56.5	220.4±59.8	160.4±44.1	164.3±20.2
Sal	0.07±0.1	0.1±0.0	0.3±0.3	0.3±0.1	0.2±0.1	0.1±0.0
Sch	14.0 (5.6)	34.0 (12.7)	22.5 (17.6)	50.5 (31.8)	56.0 (39.5)	16.5 (2.12)
DO	7.8±2.8	4.5±1.0	0.79±1.3	1.1±1.8	4.9±2.2	7.7±0.1
CQO	30.8±32.2	38.3±15.9	128.3±59.2	65.5±7.0	85.6±58.6	56.0±13.1
NO_3	45.2±26.4	36.8±27.7	11.0±8.9	17.5±8.9	10.7±6.6	7.3±3.2
NO_2	2.1±1.2	3.5±4.0	0.2±0.1	0.6±0.9	0.15±0.1	0.1±0.0
PO_4^3	5.4±6.0	11.7±7.3	19.7±15.7	14.6±9.0	4.8±1.3	4.4±0.0
Heavy metals in water (μg/L)						
Cr	1.9±0.5	5.3±3.7	3.4±1.9	7.0±4.7	8.3±4.1	4.1±0.7
CrVI	1.2±0.6	1.9±2.1	1.25±0.6	1.3±0.6	1.95±2.2	2.5±0.7
Pb	4.0±0.0	4.7±1.1	4.0±0.0	4.0±0.0	5.1±2.5	4.±0.0
Cu	8.3±2.4	13.3±6.5	9.0±3.0	9.7±3.2	11.3±4.7	8.1±4.9
Cd	0.5±0.0	0.5±0.0	0.5±0.1	0.5±0.0	0.9±0.8	0.6±0.2
Heavy metals in sediments (μg/g)						
Cr	38.2±41.2	45.5±38.5	192.0±119.8	120.9±152.9	260.1±362.6	13.1±8.0
CrVI	1.0±1.2	2.4±2.6	8.0±9.3	2.7±3.4	20.6±39.5	0.7±1.1
Pb	17.4±13.9	24.8±17.3	21.0±12.5	20.5±14.0	7.2±4.6	7.5±8.3
Cu	18.6±5.0	19.3±13.2	19.6±5.5	18.8±8.9	9.5±6.8	10.6±9.8
Cd	0.1±0.0	0.14±0.1	0.1±0.1	0.3±0.4	0.07±0.1	0.1±0.0

Ta Temperature (°C); *Dph* depth (m). *TS* total solids (mg/L); *Cnd* conductivity (μS/cm); *TH* total hardness (mg/L$CaCO_3$); *Sal* salinity (%); *Sch* transparency (Secchi disc, cm); *DO* dissolved oxygen (mg/L O_2); *COD* chemical oxygen demand (mg/L O_2); *NO_3* nitrates (mg/L NO_3); *NO_2* nitrites (mg/L NO_2); *PO_4^3* phosphates (mg/L PO_4^3).

water and bottom sediments of the lower Salado River basin, communicated by Gagneten et al. (2007).

Cr in water exceeded the Canadian guidelines (8.9 μg/L) at SCH (11 μg/L), LP1 (13.6 μg/L) and MSR (13 μg/L). Cr (VI) showed values higher than the Canadian standard but not than the Argentine standard at all sampling sites. Pb was high at SCH and MSR, and Cu in water exceeded the Canadian and Argentine guidelines at all sampling sites, even at the reference site (SJSR). Cd also exceeded the guidelines at the reference site and at MSR. Gallo et al. (2006), in a study of dissolved and particulate heavy metals of the Salado River, recorded values of Cr in water <10–30 μg/L. These authors considered that, taking in account the level of contaminant discharge, although not estimated in that study, the average could be much higher during low waters.

In sediments, we found high values of Cr in Las Prusianas and Manucho in autumn (350 and 800 μg/g Cr, respectively). On the other hand, very low values were found in San Justo in all seasons. Cr (VI) showed high values in spring in Las Prusianas (21.5 μg/g) and in Manucho (79.8 μg/g, mean 20.6 μg/g). Pb was more variable, with low values in spring at all sampling sites, but very high in autumn, winter and summer in Las Prusianas and the South Channel (maximum 40.3 μg/g in autumn, mean 24.88 μg/g). In general, Cu showed higher values in the channels and Las Prusianas. Relatively high values of Cu were found at all sampling sites, even at the control site (21.9 μg/g). Highest values were recorded in the South Channel and Las Prusianas (means 19.3 and 19.6 μg/g, respectively). Cd values were mostly low, except in Las Prusianas in autumn (mean 0.26 μg/g).

3 Materials and Methods

The monitoring of environmental parameters and the zooplankton sampling were carried out seasonally (autumn, winter, spring and summer) during 2002. Five zooplankton samples (replicates) were taken at each sampling site with a 20 L Schindler–Patalas trap of 45 μm mesh size, fixed and stained *in situ* using a formaldehyde (5%) and erythrosine solution.

Environmental parameters measured in water*, in situ,* using a Horiba U10 equipment, were: temperature (°C), pH, salinity (%), conductivity (μS/cm) and dissolved oxygen (mg/L O_2). Moreover, depth (m), transparency (Secchi disc, cm), total solids (mg/L), total hardness (mg/L $CaCO_3$), chemical oxygen demand (mg O_2/L), nitrates (mg/L NO_3^-), nitrites (mg/L NO_2−) and phosphates (mg/L PO_4^{3-}) were also measured.

Metal concentrations of river water samples were obtained according to Martin et al., in Environmental Protection Agency (EPA) (1994). One replicate of river water from each site was preserved after sampling by adding concentrated nitric acid (pH<2). For the determination of total recoverable metals a 100 mL aliquot from each well mixed acid preserver sample was transferred to a 250 mL Griffin beaker. Two milliliters of (1+1) nitric acid and 1 mL of (1+1) hydrochloric acid were added. The bakers were placed on the hot plate for solution evaporations. The volume of the samples aliquots was reduced to about 20 mL, heated at 85°C, covering the beakers lips with watch glasses to reduce additional evaporation and heated to gently reflux for 30 min. When the beakers were cool, the sample solutions were quantitatively transferred to 50 mL volumetric flasks, made to volume with reagent water, stopped and mixed.

For the determination of total recoverable analytes in sediments, the sample was mixed thoroughly and a portion (>20 g) was transferred to a tared weighing dish; then, the sample was weighed and the wet weight recorded. The samples were dried to constant weight at 60°C and the dry weight was recorded for calculation of percent solids. To achieve homogeneity, the dried samples were sieved using a five-mesh polypropylene sieve, grinded in a mortar and pestled. From the dried, grounded material a representative 1.0±0.01 g aliquot (W) of each sample was weighed accurately and transferred to 250 mL Phillips beakers for acid extraction; then, 4 mL of (1+1) HNO_3 and 10 mL of (1+4) HCl were added. The beakers lips were covered with watch glasses. The beakers were placed on a hot plate for reflux extraction of the analytes. The samples were heated to gently reflux for 30 min. Then, the samples were allowed to cool and we quantitatively transferred the extracts to 100 mL volumetric flasks. We diluted to volume with reagent water, stopped and mixed. Metal concentrations were measured according to Creed et al. in Environmental Protection Agency (EPA) (1994) with a Perkin Elmer atomic absorption spectrophotometer (model PE 5000) equipped with a graphite furnace, using a standard addition technique for calibration. The detection limits for heavy metals in water were >1; >4; >3 and >0.5 μg/L for Cr, Pb, Cu and Cd, respectively. Cr (VI) in water and sediments was analyzed by UV–visible spectroscopy. Operational precision of the analytical methods was 1 μg/L.

3.1 Zooplankton Analysis

The quali-quantitative analysis of samples was carried out by counting the organisms with a stereoscopic microscope (Nikon SMZ 10), in a 5 cm^3 Bogorov chamber for mesozooplankton (adult copepoda and cladocerans) and with a binocular composed microscope (Olympus CX31) in 1 cm^3 Segdwick–Rafter chambers for microzooplankton (rotifers and copepod nauplii). Five aliquots of 5 mL for mesozooplankton and five aliquots of 1 mL for microzooplankton were analyzed.

As can be observed through the lists of organisms, the separation into microzooplankton and mesozooplankton can be somewhat different to the same categories used in studies referred to zooplankton of the Northern Hemisphere. However, it becomes a more adjusted picture of the nature of zooplankton in the Neotropical region which in general lacks of large species as dominant in zooplankton community, particularly the scarcity or absence of the big specimens of *Daphnia* (Fernando et al. 1987)

The taxonomic identification was carried out through the use of specific keys (Paggi 1995; Reid 1985 and José de Paggi 1995). The organisms were identified at the species level except for some rotifers, in which the determinations were done at the genus level due to the difficulties to recognize them because of the scarce material or because of the contraction of specimens (e.g. *Asplanchna* sp.; *Cephalodella* sp.; *Epiphanes* sp.; *Synchaeta* sp. and Bdelloidea spp.).

The attributes of the community selected as variables of response were: total density (ind/L) and by-group density (Copepoda, Cladocera and Rotifera), micro and mesozooplankton density and dry weight biomass (μg/L), approaching the values of the different species of cladocerans, copepods and rotifers according to the tables and constants of Dumont et al. (1975). Species diversity and its components of richness (*S*) and equitability (*E*) were also calculated through the Shannon–Weaver index (Omori and Ikeda 1984):

$$H = -\sum_{i=1}^{S} Pi \bullet \ln Pi$$

where *H*: specific diversity, *Pi*: number of individuals of species *i*, and *S*: total number of present species.

Equitability (*E*) was quantified expressing specific diversity *H* as a proportion of the maximum value of *H* if all individuals were uniformly distributed among species (Begon et al. 1988):

$$E = \frac{\sum_{i=1}^{S} Pi \bullet \ln Pi}{S}$$

where:

Pi No. of individuals of species *i.*
S species richness

The possible differences among species density of the sampling sites were analyzed through one-way ANOVA with a significance level of $p \leq 0.05$. In the diversity analysis, only data from adult individuals that could be identified taxonomically at the species level were used.

The similarity analysis was used for the comparison of the composition of ensembles through the measurement of the Euclidian Distance:

$$d(i,j) = \sqrt{|x_{i1} - x_{j1}|^2 + |x_{i2} - x_{j2}|^2 + \ldots + |x_{ip} - x_{jp}|^2}$$

and expressed through a dendrogram following the Unweighted pair-group method with arithmetic mean (UPGMA), which showed the highest cophenetic correlation ($r = 0.903$, $p < 0.05$) using the program InfoStat (2004).

4 Results

4.1 Zooplankton Structure

4.1.1 Abundance

Table 2 shows the list of copepod, cladoceran, and rotifer species recorded, as well as the mean abundance of each species at each sampling site.

Total density of organisms was higher at the reference site (Salado River at San Justo, 0.86 ind/L) than at the more contaminated sites (0.31; 0.07; 0.03; 0.61 and 0.62 at the North Channel, South Channel, Las Prusianas 2, Las Prusianas 1 and Manucho, respectively; Fig. 2).

Copepods dominated the community in numbers. However, adult copepods were poorly represented quanti and qualitatively. The dominance observed at LP1, MSR and SJSR is due to the great proliferation of larvae and juveniles (nauplii and copepodites). Nauplii reached densities of 6.9, 1.9 and 3.0 ind/L at LP1 (average deviated by a maximum of 20.7 ind/L in autumn), MSR and SJSR, respectively. In general terms, adult crustaceans were not as numerous as rotifers; the presence of cladocerans was very low or null at NCH, SCH and LP2. The most frequent genera were *Bosmina*, *Ceriodaphnia*, and *Moina*. The most abundant species at LP1, MSR and SJSR were *Moina minuta*, *Bosmina hagmani*, *Diaphanosoma spinulosum* and *Macrothrix squamosa.*

Among copepods, the most frequent genera were *Eucyclops* and *Metacyclops*, being *Acanthocyclops* represented in a lower proportion. The most frequent and abundant species was *Eucyclops neumani*, which was recorded in all environments and with a relatively high abundance, except at LP1. Other species that appeared very sporadically were: *Microcyclops anceps* and *Paracyclops fimbriatus.*

When comparing the partial dominance of the three orders of copepods, we found a dominance of ciclopoids at the first four sampling sites, although they never exceeded 0.2 ind/L. At Manucho, some calanoidea and harpacticoidea were also recorded with a similar density (0.05–0.06 ind/L). At San Justo, the three orders were well represented, with dominance of calanoidea (0.53 ind/L) over harpacticoidea and ciclopoidea (0.28 and 0.21 ind/L, respectively).

Mesozooplankton was only well represented at San Justo, being scarce at Manucho and very scarce or

Table 2 Density (ind/L) of species recorded at sampling sites

Taxa	NCH	SCH	LP 2	LP 1	MSR	SJSR
Copepoda						
Cyclopoida						
Acanthocyclops robustus	–	0.067	–	–	0.100	0.156
Eucyclops neumani	0.483	0.150	0.100	–	1.133	1.844
Metacyclops laticornis	1.000	–	–	0.783	–	–
Metacyclops mendocinus	–	–	–	–	0.283	–
Microcyclops anceps	0.133	–	–	–	–	–
Paracyclops fimbriatus	–	–	–	–	0.067	0.044
Paracyclops sp.	–	–	–	–	0.033	–
Calanoida						
Argyrodiaptomus falcifer	–	–	–	–	–	0.089
Notodiaptomus incompositus	–	–	–	–	0.117	0.533
Harpacticoida						
Attheyella sancarliensis	–	–	–	–	0.067	0.333
Copepodites	0.033	–	0.017	0.117	0.500	2.489
Nauplii	0.417	0.467	0.117	6.911	1.950	3.000
Cladocera						
Alona aff rectangula	–	0.033	–	–	0.017	0.022
Bosmina hagmani	–	–	–	0.017	0.017	0.156
B. huaronensis	–	–	0.05	0.017	0.017	0.044
Ceriodaphnia cornuta	–	–	0.05	–	0.300	–
C. reticulata	–	–	–	0.050	0.050	1.178
Daphnia gessneri	–	–	–	–	–	0.022
D. spinulata	–	–	–	0.067	–	–
Diaphanosoma spinulosum	–	–	–	0.017	0.183	–
Ilyocryptus spinifer	–	–	–	0.017	0.050	0.067
Leydigia louisi	–	–	–	0.050	0.050	–
Macrothrix goeldi	–	–	–	0.033	0.083	0.377
M. squamosa	–	–	–	0.050	0.117	0.378
Moina micrura	–	–	–	0.133	–	0.111
M. minuta	–	–	–	0.050	0.083	0.556
Moinodaphnia macleayi	–	–	–	0.033	0.050	0.089
Pleuroxus similis	–	–	–	0.033	0.050	0.111
Simocephalus sp.	–	–	–	0.033	0.067	–
Rotifera						
Anuraeopsis fissa	0.183	–		–	–	–
Asplanchna sp.	0.017	0.201	–	0.050	0.267	0.333
Brachionus alhstromi	–	–	–	–	0.258	0.029
B. angularis	–	–	–	0.017	0.042	–
B. austrogenitus	–	0.017	–	–	0.150	0.039
B. bidentata	–	–	–	–	0.001	0.002
B. budapestinensis	–	–	–	–	0.050	0.017
B. calyciflorus	–	–	–	–	–	0.065
B. caudatus insuetus	–	–	0.001	–	0.471	0.056
B. caudatus provectus	–	–	–	0.017	0.126	0.033
B. caudatus vulgatus	–	–	–	–	0.075	0.037
B. plicatilis	–	–	0.001	0.017	0.040	0.036
B. quadridentatus	–	0.050	0.050	0.002	0.178	0.088
B. urceolaris	–	–	–	0.033	0.004	0.012
B. variabilis	–	–	–	–	0.019	0.004
Cephalodella sp.	–	0.017	0.033	–	–	0.008
Epiphanes clavulata	0.467	0.017	–	0.117	0.068	–

Table 2 (continued)

Taxa	NCH	SCH	LP 2	LP 1	MSR	SJSR
Epiphanes sp.	–	0.083	–	0.033	0.067	0.006
Euchlanis dilatata	0.150	0.083	–	–	–	–
Filinia longiseta	–	–	–	–	0.017	0.017
F. terminalis	–	–	–	–	0.018	0.017
Hexarthra intermedia	–	–	0.033	–	0.017	0.001
Keratella americana	–	–	–	0.217	2.951	0.033
K. cochlearis	–	–	0.002	0.067	2.471	0.067
K. tropica	–	–	–	–	1.100	0.036
Lecane curvicornis	–	–	–	0.100	0.216	0.221
L. bulla	–	–	–	–	0.100	0.002
L. elsa	–	–	–	0.183	0.083	–
L. hastata	–	–	–	–	0.133	0.001
L. lunaris	0.050	0.017	–	0.083	0.017	0.004
L. papuana	0.117	–	–	–	0.150	0.004
L. pyriformis	0.117	–	–	0.117	0.283	0.056
Lepadella acuminata	0.033	–	0.004	0.167	0.092	0.002
Lepadella rhomboides	–	0.050	–	0.067	0.083	–
Monostyla lunaris	0.017	0.033	0.017	0.050	–	0.009
Mytilina mucronata	–	–	–	0.133	–	0.029
Notholca acuminata	–	–	–	0.117	0.017	0.001
Platyas quadricornis	–	–	–	–	0.033	–
Polyarthra vulgaris	–	–	–	–	0.051	3.500
Phylodina sp.	4.319	0.017	–	0.017	–	0.047
Synchaeta sp.	–	–	–	–	0.004	0.002
Testudinella patina	–	–	–	–	–	0.008
Trichocerca bicristata	–	–	–	–	0.200	0.009
Trichotria tetractis	0.117	0.034	–	–	–	0.001
Bdelloideo sp. 1	–	0.053	0.168	0.083	0.852	–
Bdelloideo sp. 2	0.536	0.285	0.001	0.850	0.605	0.009
Bdelloideo sp. 3	0.650	–	–	0.400	0.654	0.002

Means. (–): absence

null in the tributaries (Fig. 3). Microzooplankton reached comparatively high values at Las Prusianas 1 (3.48 ind/L), caused by the abundance of nauplii, and was lower in the Salado River (1.10 and 1.55 ind/L) at Manucho and San Justo.

The high microzooplankton values are also explained by the abundance of rotifers, which were the best represented group quali and quantitatively.

The best represented rotifer genera in relation to the number of species were *Brachionus* (10 species), *Lecane* (seven species) and *Keratella* (three species). The most numerous species of the genus *Brachionus*, or with a more constant presence, were *B. quadridentatus, B. calyciflorus*, *B. plicatilis* and *B. caudatus*. The latter, most of all abundant and frequent in the Salado River, was represented by different "varieties": *insuetus*, *provectus* and *vulgatus*. *B. austrogenitus* and *B. alhstromi* were also frequent at Manucho and San Justo. The most numerous and frequent species of the genus *Lecane* were *L. lunaris* and *L. pyriformis*, and *K. americana* and *K. cochlearis* prevailed from the genus *Keratella.* The genus *Polyarthra* was recorded in the Salado River, with *P. vulgaris* showing a high density at San Justo (3.5 ind/L). Bdelloid rotifers (among them, *Phylodina* sp.) were also frequent and abundant. Among the rotifer species of higher frequency, although represented with low density values, we can mention *Monostyla lunaris*, *Lepadella acuminata*, *Asplanchna* sp. and *Epiphanes* spp.

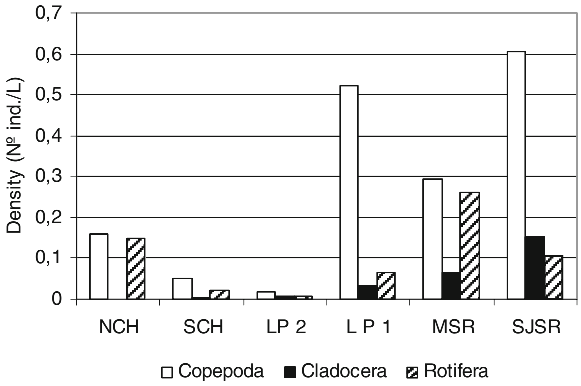


Fig. 2 By-group density (Copepoda, Cladocera and Rotifera) (ind/L) in each sampling site

4.1.2 Biomass

Absolute biomass was 17 μg/L for copepods (9.41, 4.24, 2.92 and 9.42 μg/L for Ciclopoida, Calanoida, Harpacticoida and copepodites+nauplii, respectively), 4.2 μg/L for cladocerans, and 0.4 μg/L for rotifers.

Figure 4 shows that, at Manucho and San Justo, zooplankton was constituted by the three main zooplanktonic groups: copepods, cladocerans and rotifers, with high values of biomass. Biomass of copepods was high and constant (near 3 μg/L) at SJSR. It was somewhat lower at MSR. Biomass of Copepoda, concentrated in the river and at LP 1, was distributed as follows: 55% Cyclopoida, 25% Calanoida and 17% Harpacticoida. In decreasing order of importance, cladocerans showed biomass values between a minimum of 0.3 (LP1) and a maximum of 1.6 (SJSR), being absent at NCH. They were followed by rotifers, with comparatively lower values of biomass (0.01 at LP2 and 0.2 at NCH).

Absolute biomass varied in the order: SJSR>MSR>LP1>SCH>NCH>LP2 with 11.1; 4.9; 2.7; 1.5, 1.2 and 1.1 μg/L, respectively.

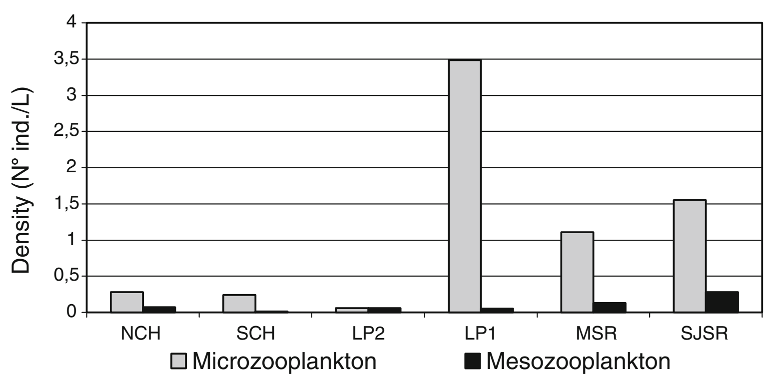
4.1.3 Species Richness, Diversity and Equitability

Figure 5 show total species richness and species diversity, recorded at the six sampling sites. A total of 74 species (Table 2) were recorded, from which 13.5% corresponded to copepods, 22.9% to cladocerans, and 63.5% to rotifers.

At MSR, a total of 59 species were recorded, while 56 species were recorded at SJSR, 38 at LP1, 17 at SCH, 16 at NCH, and 13 species at LP2. Therefore, species richness decreased among the sampling sites in the following order: MSR>SJSR>LP1>SCH>NCH>LP2. In function of richness, two environmental groups can be formed: the tributaries, with lower species richness (between 13 and 36 species), and the main river course at MSR and SJSR, with almost twice the number of species (between 56 and 59 species).

The dominant group was rotifers, which were present at all sampling sites. In the river (MSR and SJSR), 99% of all rotifer species were represented. At LP1, 50% of species were represented; at LP2, 22%, and only 24% at NCH and SCH, with some species being exclusively from these environments. Such is the case of *Anuraeopsis fissa* and *Euchlanis dilatata.* The second group were copepods, with low species richness [1–2 species in the tributaries and somewhat higher (6–7 species) in the river], while cladocerans contributed significantly to the community only at the reference site (RSSJ), where they showed a more uniform abundance (Fig. 1).

Fig. 3 Density (ind/L) of microzooplankton (Rotifera +nauplii larvae) and mesozooplankton (Copepoda+ Cladocera) in each sampling site



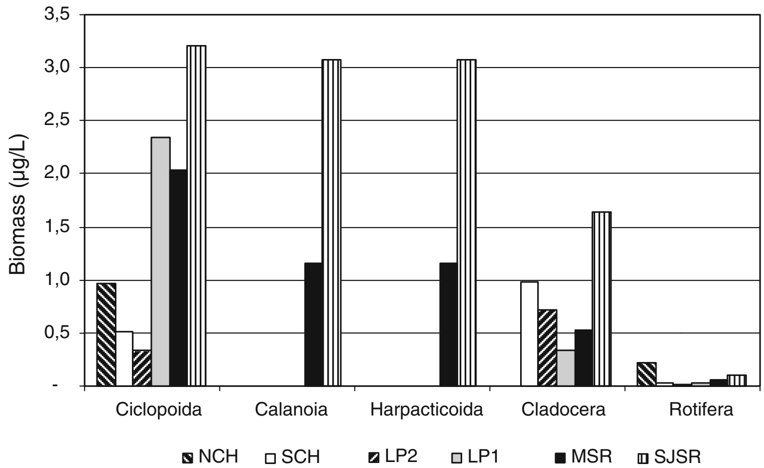


Fig. 4 Biomass (µg/L) of Cyclopoida, Calanoida, Harpacticoida, Cladocera and Rotifera in each sampling site

Figure 1 shows the relative richness of Rotifera, Copepoda and Cladocera when considering the 20 most frequent species recorded at each sampling site. In the direction of the basin current, i.e., from NCH to MSR, and in relation to RSSJ, the absence of cladocerans was observed at NCH, with absolute dominance of rotifers and scarce copepods. This situation was maintained at the other contaminated sites, but the presence of cladocerans increased progressively towards the river, at Manucho (MSR). A similar proportion of the three groups was found in the river, at San Justo (SJSR). Species diversity showed low values (0.35 to 1.56) in the tributaries and higher values in the Salado River, at Manucho (3.0) and San Justo (3.16). Equitability varied between 0.03 (in LP1) and 0.06 (in SJSR).

5 Discussion

5.1 Community Structure

Mean total density of organisms was significantly higher at the reference site (San Justo=2.73 ind/L) than at the contaminated sites (0.31; 0.07; 0.03; 0.62 and 0.63 at the North Channel, South Channel, Las Prusianas 2, Las Prusianas 1 and Manucho, $p<0.002$; 0.0002; 0.0001; 0.0013 and 0.0015, respectively).

In general, total zooplankton abundance was low, if compared with other aquatic environments of the region belonging to the Paraná River basin, including the Salado River. Values of zooplanktonic density recorded by other authors show that it is higher in lenitic than in lotic water bodies, in secondary courses

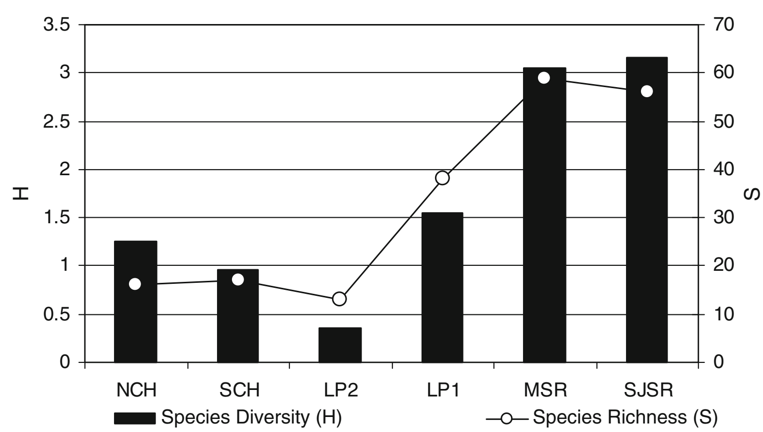


Fig. 5 Species richness (S) and species diversity (H) recorded in each sampling site

than in the main course of the rivers, and also in low waters than in high waters. In this respect, José de Paggi and Paggi (1998) recorded 750 ind/L in a pond that receives permanent nutrient and organic matter inputs, related to the Salado River by overflows. On the other hand, Paggi and José de Paggi (1990) recorded between 10 and 148 ind/L in the main course of the Paraná River, but communicated that values of 10–1,100 ind/L are frequent for littoral zooplankton (Paggi and José de Paggi, 1974), 3.74–17.1 ind/L for the main course, and 25.25 ind/L for a secondary course of the Paraná River (José de Paggi 1988). Values between 148 and 400 ind/L were obtained in the middle sector of the Paraná River (José de Paggi 1984), or between 33.67 and 327.2 ind/L in another study in the same sector (José de Paggi 1983). On the other hand, José de Paggi (1981) communicated a density between 1 and 10 ind/L for secondary courses of the middle Paraná River during floods and of 100–500 ind/L during low waters. The same study indicated a mean density of 100 ind/L for the Salado River. In this river, the density increases considerably in areas with organic enrichment, with means between 166.8 and 498.9 ind/L, and maximum values of 984 ind/L (José de Paggi and Paggi 1998) in the area of influence of the Paraná River alluvial valley.

Moreover, the higher plankton abundance found in the Salado River at Manucho and San Justo can be related with some physicochemical parameters and the trophic condition of the system.

Physicochemical parameters setting up conditions that could be limiting for zooplankton are depth and temperature (Gillooly 2000; Hobæk et al. 2002; José de Paggi and Paggi 2007). Depth, which could be interpreted as representative of the flow, significantly higher in the main sector of the Salado River, showed positive correlations with mesozooplankton density (r=0.67, p=0.0001), diversity and richness (r=0.63, p=0.0001 and r=0.70, p=0.001, respectively). Temperature was positively related with microzooplankton density (r=0.39, p=0.05), diversity and richness (r=0.56, p=0.004 and r=0.56, p=0.0004, respectively), probably due to the high population growth rate of rotifers, dominating microzooplankton.

Salinity and conductivity, both indicators of the quantity of dissolved solids, would seem to have a negative effect on abundance and richness of zooplankton of the Salado River basin if compared with those of the Paraná River system, with very low salinity and conductivity values.

However, there were no significant variations in conductivity and salinity among sites, so correlations with abundance, biomass, richness and diversity were not significant. Hardness and pH, which are environmental factors associated, to a great extent, to those mentioned previously, were neither important in the structuring of the community, since they did not show a significant relationship with the biological parameters. This pattern would be related to the mentioned environmental context of the Salado River: The lower basin of the Salado River is a system with high levels of salinity within a system of low salinity (the Paraná River), imposing limiting conditions for the establishment of populations belonging to the Paraná River main system, adapted to conditions of low salinity.

In this line of evidence, Depetris and Pasquini (1976) reported a mean dissolved salt concentration of 79.8 mg/L and communicated that conductivity of the Parana River measured between 1981 and 1984 at the city of Paraná (located in front of Santa Fe city) varied between 30 and 110 mS/cm (Depetris and Pasquini 2007). Gallo et al. (2006) reported results in the same area of our study and in a similar period of our study (between June 2000 and November 2001): These authors performed five field survey samplings and registered high organic matter and high microbial counts, as well as significant amounts of salt in the basin soils and consequently high concentrations of total dissolved solids (as high as 12,000 μS/cm in middle water levels) in the Salado River. In a more recent work et al. (2008b) reported that the Salado River is characterized by a high conductivity, with a maximum value of 3,100 mS/cm.

DO showed a positive relationship with total density (r=0.42, p=0.04) and with mesozooplankton (r=0.50, p=0.01), but not with microzooplankton (r=0.26, 0.20). This would be indicating that microzooplankton would be more tolerating to DO deficiency than the highest fraction. It is well known that rotifers stands a wide range of oxygen concentrations (0.6–13.3 mg O_2/L) (Mikschi 1989; Duggan et al. 2001; Pecorari et al. 2006). And, on the other hand, that the highest oxygen content allowed the better development of mesozooplankton recorded at Manucho and San Justo. DO also showed a significant relationship with diversity and richness (r=0.39, p=0.05 and r=0.39, p=0.058, respectively), both at high levels at Manucho and San Justo.

A previous study (Gagneten and Ceresoli 2004), carried out in the tributaries of the Salado River, also showed that the concentration of dissolved oxygen was very low (1.6 mg O_2/L) at SCH, in agreement with the higher BOD (45.8). Organic matter values were high (between 200 and 256 mg/L), although not very different among the sampling sites. BOD showed very high values at SCH and high values at Las Prusianas, corresponding to poly and mesosaprobious environments, respectively, according to Margalef (1983).

In the tributaries, the highest input of heavy metals, plus the oxygen deficit at LP, could be the causes of the scarce development of the analyzed species association. In spite of the high concentration of organic matter, that could mean higher resources available for zooplankton, specially for filtering organisms (Gulati and Demott 1997; Gliwicz 1990; Nogrady et al. 1993; Ravera 2001), the density at NCH, SCH, LP2 and LP1 was very low, which suggests the probable effects of heavy metals. Moreover, specific diversity was lower at the two environments with the highest trophic levels (SCH and LP2), probably due to a combined effect of the increase of resistant or tolerant species and the decrease of the most sensitive ones (Ravera 1996).

Figure 6 summarizes the clustering of all biological variables (total and by-group density, micro and mesozooplankton density, biomass, species richness and diversity) along with the concentration of heavy metals in water and sediments. The main steam of the river showed high density, richness, and biomass both in Manucho and in San Justo, the site originally taken as reference site. The river in Manucho and San Justo showed bigger Euclidean distance, and separation from channels and streams. On the other hand, Las Prusianas with its two sampling sites close to Esperanza City, separated from the North and South channels of Rafaela City. In summary, the results of this study show that the highest density, richness, biomass and equitability of zooplankton were found in the main course of the river, at Manucho and San Justo. That is to say, at Manucho and San Justo, the community found better conditions for its development.

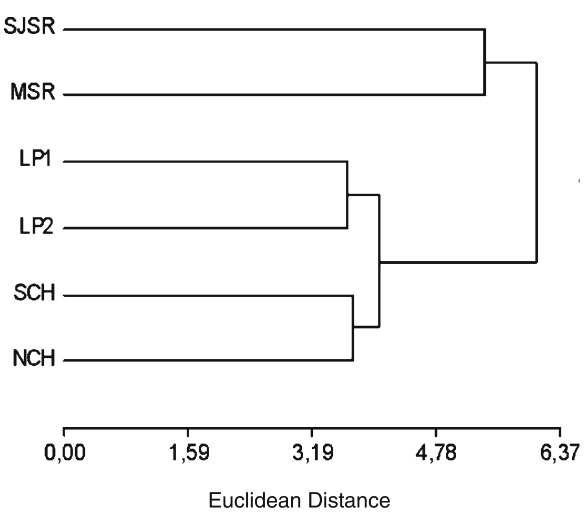


Fig. 6 Cladogram (UPGMA method) of biological parameters and heavy metals concentration in water and sediments

Other data on the enrichment with heavy metals of the system studied, were provided by Gallo et al. (2006): total Cr showed huge variation (that the authors explained as intermittent inputs of the metal load from anthropogenic source) varying from an average concentration of 30 μg/l to <10 μg/l in the Cululu River (a tributary of Las Prusianas stream). In the Salado River they recorded 18.8 and 12.1 mg/L. In the discharge point, near Esperanza City, (which correspond to MSR in our survey) they recorded a level of Cr as high as 4,573 mg Cr/L. Moreover, the authors points out that the discharge, being direct, showed a great impact, mainly during low water levels in the Salado River.

Similar to our records of heavy metals and more recent ones in the same study area were reported by Marchese et al. (2008b): The aim of the study was to analyze Cr concentrations in water and bottom sediments in the main channel of the Salado River (tributary of Middle Parana River) and its floodplain. The main changes caused by human activities and hydrological disturbances on benthic invertebrate structure were also analyzed. Cr sediment concentrations varied between 44.2 and 209 mg Cr/g (dw) reaching the highest values in the wetland floodplain. Total water Cr values obtained in this study have been always higher than guidelines given by the Subsecretaria of Recursos Hídricos of Argentina for the aquatic biota (<2.5 mg/L). On the other hand, the authors conclude that the reference area (at San Justo) also shows, although at lower levels, as in our study, signs of contamination exceeding the guidelines given by CEPA (2002) (37.3 mg/g) according to results obtained on Cr concentrations in sediments, where live the benthic invertebrates. On the other hand, Gagneten et al. (2007) reported a value as high as 800 μg/g at MSR.

Moreover, the high level of leather production did not diminish since the samples our study were taken, rather it increased (6,000 leathers/day) and with the exception of certain improvement in the treatment of the effluents, no measures of environmental sanitation neither fiscal policies of bigger control of discharges of effluents were applied in the region.

5.2 Sensitivity of Cladocerans, Copepods and Rotifers

A pattern observed in relation to the prevalence of rotifers, cladocerans or copepods in regional studies is that rotifers are dominant in lenitic and lotic environments, but relative abundance of crustaceans increases in quiet waters (Paggi and José de Paggi 1990). However, an important structuring factor of the community and of the population abundance is the deterioration of the chemical quality of the water. Gama-Flores et al. (2006) suggested that, although the competition among zooplankton populations is a natural phenomenon, in which cladocerans are usually competitively superior, anthropogenic factors, such as the disposal of industrial effluents, can modify this interaction. In this study, the limiting factor for crustacean development—especially cladocerans—in the tributaries, with lower depth and flow than in the river main course, was the high degree of contamination by heavy metals recorded in water and sediments. Heavy metals could explain the low occurrence of copepods and the almost absence of cladocerans in the tributaries and their higher representativeness in the river, in spite of the higher turbulence.

Our data indicate that metals did not affect similarly the different components of zooplankton: cladocerans were the most affected by heavy metals, while copepods and rotifers were the most resistant ones. It is well known that cladocerans are particularly sensitive to the toxic substances, being frequently utilized in normalized toxicity tests. Industrial effluents constitute a potential threat to the maintenance of the diversity of this group (Paggi 2004). In a similar way, in laboratory assays, heavy metals affected cladocerans in a higher degree than copepods. The latter are relatively tolerant to the toxic action of heavy metals, and also adults are more resistant than copepodites and nauplii (Roch et al. 1985).

Available data of contaminated ponds indicate that the residence time of heavy metals in the epilimnion can be high: Coale and Flegal (1989) recorded a residence time of 660, 23, 20, and 4 days for Cu, Zn, Cd and Pb, respectively. Fleming and Trevors (1989) indicated a mean concentration of only 3.0 μg/L Cu for uncontaminated aquatic systems of the world, but this value was exceeded at all sampling sites in this study. Pb was mainly found in the sediments, as was indicated by WHO (1989), that stated that in contaminated aquatic systems almost all Pb is strongly related to sediments. In relation to Cr, the mean value in freshwater worldwide is 0.00018 ppm (Rollinson 1973), exceeded many times in this study.

Cr values up to 21 times higher than permitted standards were recorded in sediments, being high at NCH, SCH, LP2, LP1 and MSR, corresponding to current and historical contamination Gagneten et al. (2007). In addition to the known importance of organic matter in the process of capturing of heavy metals in the water column and posterior deposit in the sediments, the biota can also act sequestering suspended heavy metals. It is possible that zooplankton would be an important component of this process due to their great surface–volume relationship by unit of mass and their high metabolic rate (Ravera 2001). In an enrichment experiment with heavy metals in limnocorrals, Kerrison et al. (1988) found that Cu remained more time in the water column widely associated to suspended particulate material, while Cd remained more time in solution. These authors showed that copepods were more tolerant to Cu and more sensitive to Cd, while rotifers showed an inverse sensitivity.

5.3 Biomass

This study showed that, although rotifers were numerically dominant, the highest biomass was provided by copepods. In uncontaminated systems, crustaceans have a key role in the community structure. In this respect, Fussman (1996) indicated that ciclopid and calanoid copepods decreased rotifer abundance, in addition to competing, in many cases, for the algal resource; on the other hand, cladocerans decreased rotifer populations by trophic competition and mechanical interference. However, our data indicate that contamination is a more significant structuring factor than the traditionally considered biological factors. In many cases, contamination can determine the prevalence of more tolerant populations. Thus, in our study the community became dominated by *r* strategist or "opportunistic" organisms, represented by rotifers and nauplii.

Absolute biomass decreased in the order Copepoda> Cladocera>Rotifera. The highest biomass was concentrated in the river. Among the sampling sites, absolute biomass varied in the order: SJSR>MSR>LP1>SCH> NCH>LP2 with 11.1; 4.9; 2.7; 1.5; 1.1 and 1.0 μg/L, respectively. Biomass followed a similar distribution than absolute density, which varied in the order: SJSR> MSR>LP1>NCH>SCH>LP2 with 0.86; 0.63; 0.62; 0.31; 0.07 and 0.03 ind/L, respectively.

In the Middle Paraná River, Paggi and José de Paggi (1990) recorded higher biomass values: between 8 and 420 μg/L dry weight, with a mean of 95 μg/L. For secondary courses of the Paraná River, José de Paggi (1981) recorded between 0.26 and 58.8 μg/L dw. Although the percentage attributed to crustaceans was higher, rotifers were generally dominant.

The results of this study indicate that biomass was much lower compared to less contaminated systems of the region. This pattern is contrary to that recorded for density, which was related, as already mentioned, to the establishment and proliferation of small size species.

The correlation between density and biomass was r=0.84 (p=0.0001); being r=0.55 (p=0.005) with the macro fraction and r=0.94 (p=0.0001) with the micro fraction, i.e. the main responsible for the relationship between biomass and density was microzooplankton. Likewise, Roch et al. (1985) found an inverse correlation (r=−0.759) between zooplanktonic biomass and concentration of heavy metals in Canadian lakes and rivers.

Correlations between heavy metals and biological parameters were significant when considering copepods, cladocerans and rotifers separately. We recorded a significant negative correlation between copepod biomass and concentration of Cu and Pb in sediments (r=0.50; p=0.01 and r=0.40; p=0.05, respectively). Other significant correlation was found between rotifer density and Cr in sediments (r=0.46; $p<$ 0.024). This indicates that this line of evidence (decrease in zooplankton biomass at higher heavy metal concentration) is a good indicator of contaminated aquatic systems.

Moreover, in the cases were a negative correlation among metals in water and biological parameters of zooplankton could not be found, this could be explained by the evolutionary adaptation that populations have reached to be able to survive in such environments. The disposal of heavy metals to the system is produced since decades ago (at least since 1887), enough time for the development of thousands of generations of planktonic organisms with a very short generation time.

The higher biomass of copepods recorded in this study was related to their greater size, but also to the higher tolerance shown by copepods in relation to cladocerans. The former would also have a greater capacity to accumulate heavy metals. There are some studies that demonstrate this fact, although the information on bioaccumulation of heavy metals in zooplankton is scarce, due, to a great extent, to the operative difficulties related to the quantity of biomass necessary to carry out the determinations. In a study on marine zooplankton, Pempkowiak et al. (2006) found that the concentration of Cu, Pb, Co, Ni, Cr, Mn, Zn, Fe and Al, but not of Cd, was higher in copepods than in cladocerans. In experimental studies, Yan et al. (2004) found an important degree of resistance of copepods to heavy metals. Likewise, Gulati et al. (1988) found that *Daphnia* spp. were the most affected ones, while copepods, especially ciclopids, showed to be the most resistant to Cd exposure.

5.4 Richness

In general terms, lower species richness was found in the tributaries, according to the increase in the contaminant discharge, the trophic condition of the system and the dominance of *Eucyclops neumani.* This species showed to be very tolerant, so that it could be used as indicator species for water quality in the region.

Among the indicators used by ecotoxicologists to evaluate the effects of contaminants, the reduction in species richness is probably the most consistent and less argued response. Rapport et al. (1985) included the decrease in species richness as a general indicator of the ecosystemic distress syndrome.

In the environments analyzed in this study, cladoceran species, although always showed low densities, were comparatively numerous, most of all in the river, due to the better conditions already mentioned, offered for plankton proliferation, but they could not develop adequately in its tributaries SCH, LP2 and NCH with 1, 2, and 0 species, respectively. This indicates that Cladocera was the less tolerant group to contamination by heavy metals.

In contrast with what was observed for density, richness values were not low (maximum=59 spp.) if compared to those obtained for uncontaminated environments of the region such as the Paraná River, where José de Paggi (1981) recorded 76 species, from which 71% corresponded to rotifers, 20% to copepods and only 9% to cladocerans.

In this study, richness and density showed a high positive correlation (r=0.88, p=0.0001). Species richness was not low at NCH, SCH, LP 1 and 2, since there was an increase in rotifer taxa. That is to say, there was replacement of crustaceans by rotifers (zooplankton richness–rotifer density correlation r=0.40, p=0.0012; zooplankton richness–copepod density correlation r=0.53, p<<0.0001; zooplankton richness–cladoceran density correlation r=0.39, p=0.0007; zooplankton richness–copepod richness correlation r=0.76, p=0.0001; zooplankton richness–cladoceran richness correlation r=0.51, p=0.001; and zooplankton richness–rotifer richness correlation r=0.88, p=0.0001).

The prevalence of rotifers in contaminated environments is widely documented in the literature (Havens and Hanazato 1993; among others) and implies important changes in the community structure, which becomes dominated by smaller size organisms. In this sense, Pecorari et al. (2006) after the analysis of the zooplankton of the shore of a shallow lake that receives urban drainage, recorded high frequency of Bdelloid rotifers and significantly higher abundance of organisms. Rotifers frequently occur in treatment ponds since they resist low oxygen concentrations and feed on bacteria and microflagellates. Changes in the structure and dominance of communities indicate stress situations (Rapport and Whitford 1999). Therefore, the observed characteristics of the community may be considered as effects of contamination.

In experimental conditions, Havens (1994) found that Cu affected freshwater plankton, decreasing species richness/diversity, shortening the trophic chains and webs, reducing their complexity and decreasing the efficiency in the use of the resources.

5.5 Diversity

A marked decrease in the specific diversity is considered by some authors (Margalef 1983; Clements and Newman 2002; among many others) as a good indicator of contamination. These last authors also point out that, due to the computational simplicity to calculate the diversity index, it becomes the best way to obtain ecologically relevant information. In this study, H varied between 1.8 and 2.3 in the most contaminated tributaries and was 3.0 at the two sampling stations corresponding to the river main course, Manucho and San Justo. As mentioned previously, at NCH, SCH and LP, only three cladoceran records were found, at only three occasions, and with a very low abundance.

Obviously, the few species that were able to remain, would have reached mechanisms of resistance to contamination conditions. Such is the case of the larval stages of copepods and adults of *E. neumani*, of which males and females were recorded with relative frequency. The Shannon diversity index varied according to the contamination gradient. This would allow the differentiation of levels of contamination from diversity values, and indicates that H is also a good indicator of stress in contaminated systems. Diversity was related to zooplankton richness (r=0.93, p=0.0001) and density (r=0.95, p=0.0001).

In summary, the analysis of all recorded parameters allows to suppose that the high concentration of heavy metals, especially Cr and Cu, would prevent the establishment and development of stable populations in the river tributaries. On the contrary, the concentration of heavy metals was a little lower in the Salado River, at Manucho and San Justo, which, added to other limnological aspects, has determined that populations found better conditions for their development. That is to say, the low flow of the tributaries would have a scarce effect of dilution on contaminants and, therefore, an important impact on the biota, while the contrary was observed in the Salado River, at Manucho and San Justo, where the flow is much higher.

Although it is much argued if organic contamination affects positively or negatively the abundance of zooplankton in lotic environments, there is a wide agreement on that contamination by heavy metals has a marked negative effect on this parameter. In this respect, the high degree of eutrophication of the system, evidenced by the environmental parameters analyzed, was added to the disturbing effect of contamination. Suspended solids, nutrient and metal loads, and COD indicated the system degradation. At SCH, the organic matter input from the tanneries, as rests of skins, hairs and hoofs, could also be recorded (personal observation).

6 Conclusions

The results of this study show that heavy metals in the lower Salado River basin could act facilitating the proliferation of *r* strategists, decreasing the biomass, the species richness and diversity and favoring the occurrence of a few tolerant species and the decrease of the most sensitive ones. These effects would be related mainly with the action of Cr and Cu and with the high degree of eutrophication of the system.

In comparison with less contaminated systems, the biomass was much lower. This indicates the establishment of *r* strategist species, such as rotifers, of small size, very short generational time and high reproductive rate. Another evidence line from this study was the decline of zooplankton biomass at a higher concentration of heavy metals. This indicates that this parameter was also a good indicator of the system contamination.

Rotifers were the most tolerant ones; copepods were the second group, while cladocerans contributed significantly to the community only at SJSR, where there was also a higher equitability. Cladocerans did not show to be tolerant to the toxic action of heavy metals.

Due to the high tolerance demonstrated by *Eucyclops neumani* it is proposed as indicator species of water quality.

It was also possible to identify different levels of contamination from the diversity values: H was a good indicator of stress of contaminated systems.

Richness (S) allowed the separation of the studied environments into two groups: the tributaries, with low species richness, and the river, with higher species richness.

The weak correlation among metals in water and biological parameters of zooplankton could be explained by the evolutionary adaptation that populations have reached to be able to survive in highly contaminated environments. The disposal of heavy metals to the system is produced since decades ago (at least since 1887), enough time for the development of thousands of generations of planktonic organisms with a very short generation time. More recent data on heavy metal contamination of the region provided by other authors confirm the vigency of the contamination reported in this work.

The results of this study show the importance of zooplankton for the biological characterization of the environment in areas (Fig. 6). We can conclude that zooplankton responds as good descriptor of water quality, constituting an efficient tool together with environmental parameters.

The river offered better conditions for the development of the community (higher flow and amount of dissolved oxygen) than the tributaries. This allows the establishment of important populations at Manucho, one of the contaminated sites, and San Justo, the site initially taken as the reference site.

Our results agree, in general terms, with what was proposed by Clements and Newman (2002) in relation to the importance of biomonitoring at the community level. This approach let us to conclude that species differ in their sensitivity degree to anthropogenic contaminants, producing structural and functional changes at the contaminated sites.

Considering the results obtained in this study, we can also conclude that contamination of the system imposed extreme conditions that lead to its gradual degradation affecting zooplankton negatively. The decrease in specific richness and diversity observed at the stations nearest the cities of Rafaela and Esperanza, i.e. the points of tanneries effluent discharges, was related with the increase in the concentration of heavy metals and the high degree of eutrophication of the system.

This study completes the physicochemical study of the lower Salado River basin and relates the contamination by heavy metals recorded in water and sediments with the deterioration of the zooplanktonic community. We agree with Ravera (2001) in that it is not only useful but essential to gather different lines of evidence (e.g. chemical and biological monitoring) to determine if there is contamination in a given area.

Acknowledgements This research was supported by grants from the Universidad Nacional del Litoral, Argentina (Projects CAI+D 2002 No. 14/99 and CAI+D 2006 No. 21/122).

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