

Water quality assessment through biotic indices in a heterogeneous basin (Río Grande, Jujuy, Argentina)

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ABSTRACT. The aim of this work is to evaluate the ecological quality of waters along an environmentally heterogeneous basin, using macroinvertebrate biotic indices (BI). Twelve sampling sites were selected in the Río Grande basin, following an altitudinal gradient across the Puna, Monte and Yungas ecoregions. Sampling was conducted in the dry and rainy seasons, during three consecutive years (2012 to 2014). At each site, physico-chemical parameters were measured and benthic macroinvertebrates were collected. Principal component analysis (PCA) was applied to explore environmental variability in terms of physico-chemical parameters. The water quality of the basin was assessed by means of different biotic indices. Correlation analyses were performed between the biotic indices and the physicochemical parameters. Altitude, NaCl, total dissolved solids (TDS) and electrical conductivity were higher at the Puna and Monte sites, while river discharge, width and depth were higher at the Yungas sites. Biotic indices suggested good water quality. The BMWP (Biomonitoring Working Party) and family richness were the most suitable indices to simultaneously compare the rivers of the different ecoregions. The change in the composition of macroinvertebrate assemblages was observed at an altitude of 1500 m a.s.l., separating arid and humid zones and coinciding with inflection points of conductivity, NaCl and TDS.

[Keywords: biological integrity, aquatic insects, biotic index, river basin, water quality]

RESUMEN. Evaluación de la calidad del agua a lo largo de una cuenca heterogénea (Río Grande, Jujuy, Argentina). El objetivo de este trabajo fue evaluar la calidad ecológica del agua a lo largo de una cuenca heterogénea utilizando índices bióticos basados en macroinvertebrados. Se seleccionaron 12 sitios de muestreo en la cuenca del Río Grande en un gradiente altitudinal a través de las ecorregiones Puna, Monte y Yungas. La colecta de muestras se realizó durante la temporada seca y la de lluvias, a lo largo de tres años consecutivos (2012 a 2014). En cada sitio se midieron los parámetros físico-químicos y se recolectaron macroinvertebrados bentónicos. Para estudiar la variabilidad ambiental en base a parámetros físico-químicos se realizó un análisis de componentes principales. La calidad del agua de la cuenca se evaluó por medio de diferentes índices bióticos. Se correlacionaron los valores de los índices bióticos y los parámetros físico-químicos. La altitud, el NaCl, los sólidos disueltos totales (SDT) y la conductividad presentaron valores mayores en los sitios de la Puna y el Monte, mientras que el caudal, el ancho y la profundidad fueron mayores en los sitios de las Yungas. Los índices bióticos aplicados sugirieron una buena calidad del agua. El BMWP (Biomonitoring Working Party) y la riqueza de familias resultaron los índices más adecuados para comparar simultáneamente los ríos de las diferentes ecorregiones. Se observó el cambio de la composición de los ensambles de macroinvertebrados a una altitud de 1500 m s.n.m., separando las zonas áridas de las zonas húmedas y coincidiendo con puntos de inflexión de la conductividad, el NaCl y los SDT.

[Palabras clave: integridad biológica, insectos acuáticos, índice biótico, cuenca hídrica, calidad del agua]

INTRODUCTION

Aquatic insects and other benthic invertebrates are the most widely used organisms in freshwater biomonitoring (Bonada et al. 2006) due to their high ubiquity and species richness, high susceptibility to disturbance and limited mobility, and because they require little sampling effort (Resh 2008). In Argentina, different assessments of freshwater quality used benthic macroinvertebrates (Vallanía et al. 1996; Miserendino and Pizzolón 1999; César et al. 2000; Rodrigues Capítulo et al.

2001; Pavé and Marchese 2005; Gualdoni et al. 2011; Crettaz-Minaglia et al. 2014; Damborsky and Poi 2015, and others). In northwestern Argentina, numerous studies have focused on aquatic macroinvertebrates (Domínguez and Fernández 1998; Fernández et al. 2002; Fernández et al. 2006; von Ellenrieder 2007; Dos Santos 2011; Nieto et al. 2016; Hankel et al. 2018; Molineri et al. 2020). In general, these investigations have dealt with a single ecoregion such as the Yungas, Chaco, Monte or Puna, adapting one or several biotic indices (BI) to the regional fauna.

The Andes are characterized by the altitudinal succession of ecological zones. Andean basins can include three or more ecoregions as contrasting as deserts and rainforests, which are reflected in the composition and diversity of aquatic communities challenging the use of BI.

The Río Grande basin, from the Quebrada de Humahuaca (UNESCO World Heritage 2003) is born in a high altitude desert (Puna) and crosses xerophilous shrublands (Monte), foggy grasslands, mountain rain forests (Yungas) and dry forests (Chaco). Despite the importance of this basin, the most populated of the province of Jujuy, its macroinvertebrate diversity has been little studied, and ecological quality assessments are limited to one survey carried out using BI based on aquatic organisms from a short section around the foothills (Fernández et al. 2006). Given the high environmental heterogeneity within the Río Grande basin, we set out to evaluate the functioning of various biotic indices by studying macroinvertebrate communities along the main channel of the Río Grande and in relevant tributaries of its basin.

MATERIALS AND METHODS

Study area

The Río Grande basin in the province of Jujuy has an extension of ~240 km, an average width of 50 km and drains a region of 7250 km², with channels totaling about 1900 km. Its main collector is the Río Grande, which crosses the entire province (Figure 1) in a north-south direction (854-4840 m a.s.l.), passing through the Puna, Monte and Yungas ecoregions, and with several population settlements along the course (Chayle et al. 2001).

The Puna ecoregion is located above 3000 m a.s.l., its relief is more or less flat crossed by some mountain ranges. The climate is cold and dry with a high daily thermal amplitude. Precipitation concentrates in summer (100-200 mm/year), and the soils, of little development and vegetal coverage, are very susceptible to erosion. The vegetation is of the steppe type, adapted to extreme arid environmental conditions, and is mainly composed of dwarf shrubs (Malizia et al. 2010). The Monte de Sierras and Bolsones sector extends as a north-south strip, east of the Andes mountain range.

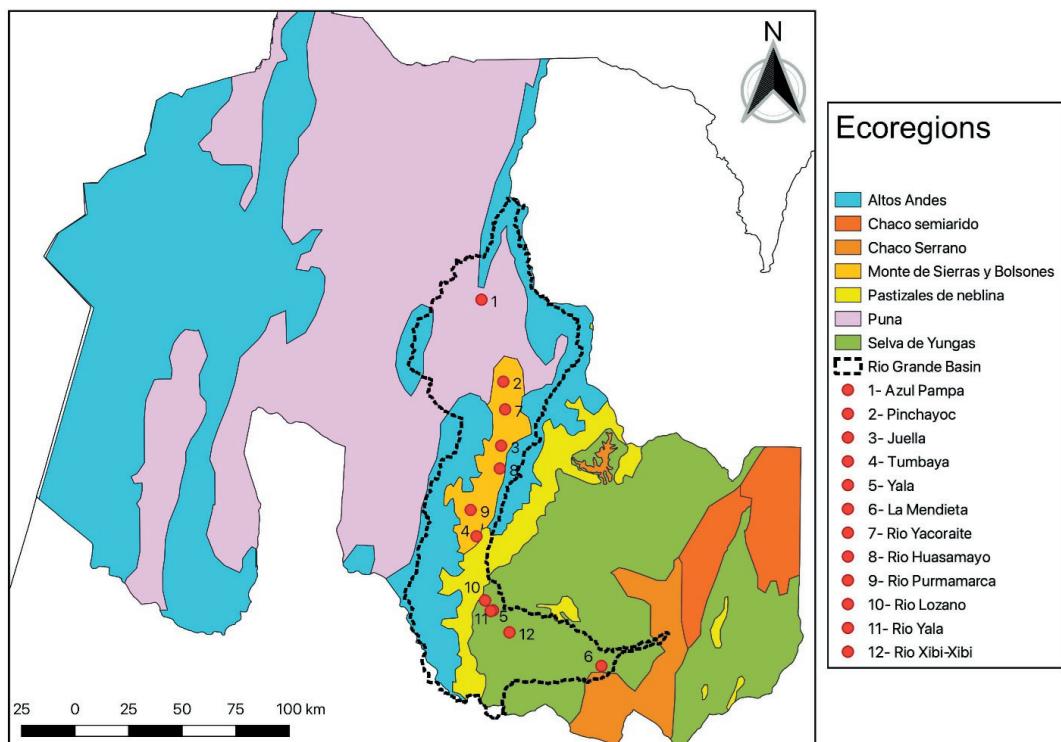


Figure 1. Map of the province of Jujuy with site locations. Río Grande basin is marked with a dotted line. Colors indicate ecoregions.

Figura 1. Mapa de la provincia de Jujuy con la ubicación de los sitios. La cuenca del Río Grande está marcada con la línea punteada. Los colores indican ecorregiones.

The climate is characterized by its aridity with rainfall restricted to summer (<500 mm/year), and has large daily and seasonal thermal amplitude. The soils are predominantly sandy, poor in organic matter, and saline. The predominant type of vegetation is the tall shrub steppe (1-3 m height), with the presence of arboreal cacti (Malizia et al. 2010). The Yungas ecoregion occurs in numerous mountainous sectors linked to the foothills of the Andes (400 to 3000 m a. s. l.). The climate is hot and humid to sub-humid with precipitation (>1000 mm/year) concentrated in summer. The soils are comparatively more developed with abundant organic matter. The strong altitudinal gradient is reflected in different plant formations: Pedemontane Jungle (400-700 m a. s. l.), Montane Jungle (700-1500 m a. s. l.), Montane Forest (1500-3000 m a. s. l.) and Grasslands altitude (>3000m a. s. l.). This study includes sites located in Montane Jungle and Montane Forest (Burkart et al. 1999; Malizia et al. 2010).

The Río Grande basin sustains different economic activities. For example, in the Puna and the Monte there are crops of fruits and vegetables on the terraces and alluvial cones, while in the Yungas ecoregion, sugar cane and tobacco are produced (Chayle et al. 2001). There is also extensive cattle ranching of sheep and llamas in the Puna, goats in the Monte, and bovines, horses and pigs in the Yungas. Moreover, there are numerous mining operations in the area, with extraction of iron, lead, silver, zinc, borates and construction stones (Dente and Martínez, nd.).

Sampling

Four sampling campaigns were carried out on the main channel (sites 1 to 6) (Table 1),

in dry and rainy periods of 2012 and 2013, whereas in the six tributaries, two samplings were performed in 2014, covering also the two hydrological periods (Table 1). The samplings were carried out under stable environmental conditions, that is, avoiding abrupt increases in flow and/or post storm conditions. At each date and sampling site water velocity, depth, discharge, dissolved oxygen (mg/L), electric conductivity (EC) ($\mu\text{S}/\text{cm}$), temperature ($^{\circ}\text{C}$), pH, NaCl (ppm) and total dissolved solids (ppm) were recorded using a multi-parametric probe (Oakton PCD650). The channel granulometry was estimated along three transects (10 m) with observations every 0.5 m, identifying the dominant and subdominant substrates [block (>256 mm), stone (64-256 mm), coarse gravel (32 to 64 mm), fine gravel (16 to 32 mm), gravel (2 to 16 mm), sand (0.06 to 2 mm) and silt-clay (<0.06 mm) (modified from Cummins 1962)].

In the six sites of the main channel (Río Grande), invertebrate quantitative samples were taken in rapids or streams [3 replicates using a Surber net (0.09 m^2 , pore 250 μm)] in dry and wet periods of 2012 and 2013 (n=72 samples). At each of these sites, qualitative samples were collected with a D net (pore 250 μm , n=24 samples), standardizing by time (15 minutes) and covering all the microhabitats present in each site (e.g., margins, pools, submerged vegetation, etc.). In the six tributaries, qualitative samples of invertebrates were taken with a D net in the dry and wet periods of 2014 (n=12 samples). The samples were preserved with alcohol 96%, and separated under a direct microscope (Nikon SMZ 800). Then, the invertebrates were preserved in alcohol 70% and identified at the lowest possible taxonomic level using available keys (Domínguez et al. 2006; Domínguez

Table 1. Sampling sites in the Río Grande basin.

Tabla 1. Sitios de muestreo en la cuenca del Río Grande.

Main channel	Code	Lat-lon	Altitude (m a. s. l.)	Tributaries	Code	Lat-lon	Altitude (m a. s. l.)
Azul Pampa	AP	22°58'18" S 65°27'01" W	3542	Huasamayo	HU	23°35'36" S 65°22'29" W	2816
Pinchayoc	PI	23°16'24" S 65°21'44" W	2870	Yacoraite	YC	23°22'30" S 65°21'16" W	2771
Juella	JU	23°30'34" S 65°22'11" W	2560	Purmamarca	PU	23°44'47" S 65°29'26" W	2298
Tumbaya	TU	23°50'36" S 65°28'00" W	2112	Lozano	LO	24°04'46" S 65°25'53" W	1640
Yala	YA	24°06'57" S 65°23'58" W	1440	Yala	YL	24°07'06" S 65°24'31" W	1468
La Mendieta	LM	24°19'07" S 64°57'48" W	733	Xibi-Xibi	XX	24°11'48" S 65°19'59" W	1314

and Fernández 2009; Michat et al. 2008; Isa Miranda and Rueda Martín 2014).

Data analysis

Two separate principal component analyses (PCA) were run to evaluate the environmental variability based on physico-chemical characteristics of the main collector and tributary sites. Data was standardized through the 'z transformation' algorithm, which scales to mean=0. RStudio software version 1.1.456 was used. Biological data was incorporated to a matrix, including the total abundances for each site and date. The following biotic indices were calculated: BMWP (Biomonitoring Working Party) and ASPT (average score per taxon) following Domínguez and Fernández (1998), %Chironomidae/total and FR (family richness), EPT (number of species of Ephemeroptera, Plecoptera and Trichoptera), %EPT (% of EPT on total abundance) (Carrera and Fierro 2001), EIPT (number of species of Elmidae, Plecoptera and Trichoptera) (von Ellenrieder 2007), IBY (Biotic Index of the Yungas) (Dos Santos et al. 2011) and ABI (Andean Biotic Index) (Ríos-Touma et al. 2014). The states 'impacted' and 'not impacted' were applied to

evaluate the ecological condition of the rivers, according to Dos Santos et al. (2011).

Spearman correlation analysis was applied to study the relationship between the first principal component of each PCA and the biotic indices, and the relationship between some of the BI (EPT, EIPT, IBY and %Chironomidae/total) and physico-chemical parameters. Correlation coefficients ($r=\rho$) greater than 0.5 ($P<0.05$) were considered.

RESULTS

Physico-chemical characterization along the main channel

The six sites located on the main channel presented different characteristics (Table S1). The EC and parameters associated with it (i.e., TDS, NaCl) took higher values in the highest sites (in the Puna and the Monte), and lower values in the lowest site 'La Mendieta'. Dissolved oxygen (DO) values were close to saturation (7.3 to 9.9 mg/L), except in the 'Azul Pampa' site during the wet season (4.6 mg/L). Differences in flow between the two hydrological periods were low.

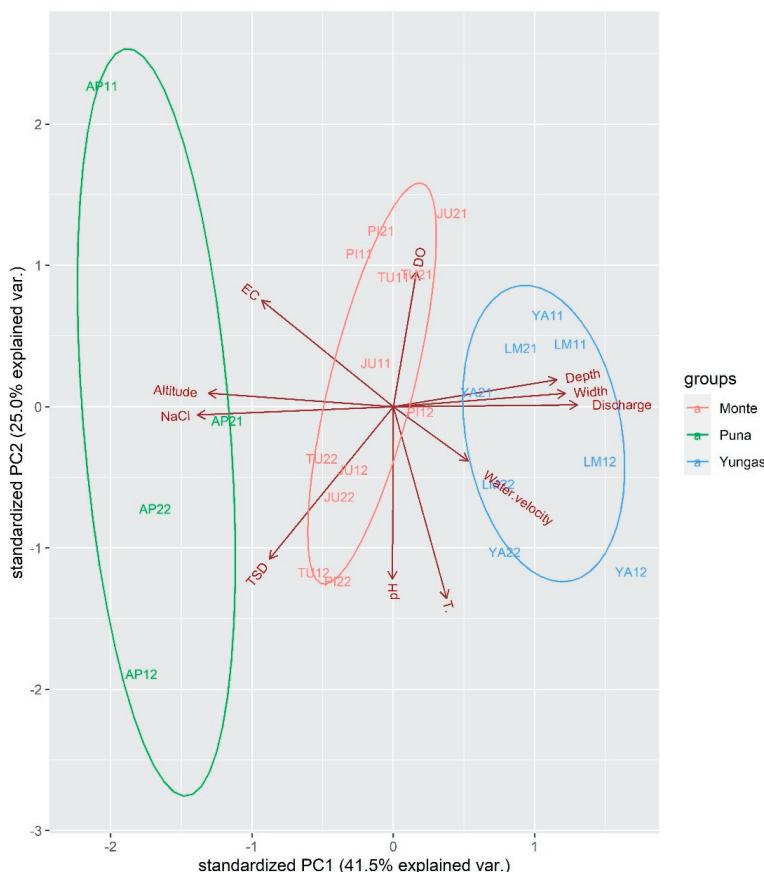


Figure 2. Principal component analysis. Biplot of the environmental variables of the six sites studied in the main channel of Río Grande basin during two hydrological periods (2012-2013).

Figura 2. Análisis de componentes principales. Biplot de las variables ambientales de los seis sitios estudiados sobre el canal principal de la cuenca del Río Grande, durante dos períodos hidrológicos (2012-2013).

The two principal components of the PCA (Figure 2, Table S4) explained 66% of the total variability [axis 1 (41%), axis 2 (25%)]. In the first axis, the sites were ordered following the altitudinal gradient, from the lower sites in the Yungas at the right side of the plot, followed by those in the Monte, and at the left the high altitude Puna sites. Discharge, width of the wet channel and mean depth were positively associated with the Yungas sites, while NaCl and altitude were positively related to the Monte and Puna sites. The second axis discriminated the wet and dry periods. Samples of the dry season characterized by higher values of DO and conductivity, in opposition to wet season samples which displayed higher values of temperature, pH, and TDS.

Physico-chemical characterization of the tributaries

In the six tributaries sampled, the environmental variables showed similar values to those recorded in the main channel, although the TDS did not display such a clear variation with altitude (Table S2). Dissolved

oxygen generally was close to saturation. Discharge did not show significant spatial or temporal variation.

The two principal components of the PCA (Figure 3, Table S4) explained 68% of the total variability [axis 1 (51%), axis 2 (17%)]. In the first axis, the sites were ordered following the altitudinal gradient, the lower sites of the Yungas were clustered at the right side of the plot, and the high altitude Monte sites at the left. The width of the wet channel, the discharge and the mean depth were positively correlated with the Yungas rivers; while EC, NaCl, TDS and altitude were positively correlated with the Monte rivers. The second axis discriminated the wet and dry periods; dry season samples were characterized by higher values of DO, in opposition to wet season samples which displayed higher values of temperature.

Biological indices

A total of 211723 individuals belonging to 133 taxa were collected, identified and counted. Most of them corresponded to Insecta (66%) and the remaining were distributed in

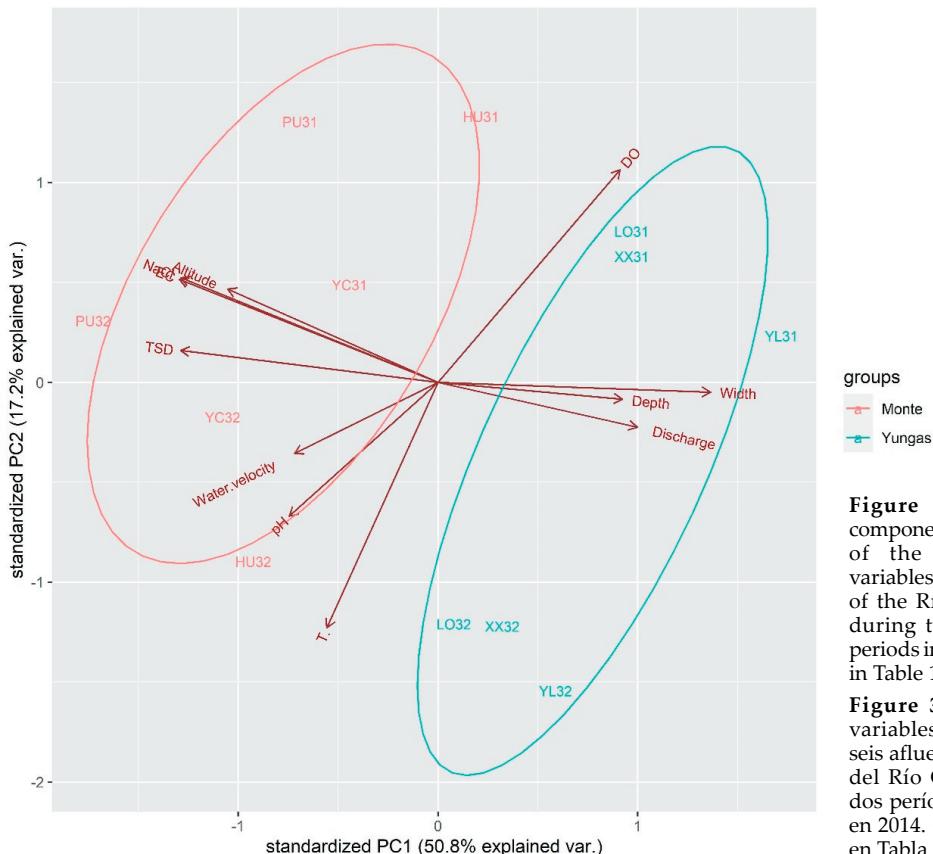


Figure 3. Principal component analysis. Biplot of the environmental variables of six tributaries of the Río Grande basin, during two hydrological periods in 2014. Sites codes in Table 1.

Figure 3. Biplot de las variables ambientales de seis afluentes de la cuenca del Río Grande, durante dos períodos hidrológicos en 2014. Códigos de sitios en Tabla 1.

Crustacea, Acari, Gastropoda, Collembola, Oligochaeta, Hirudinea and Tardigrada (Table S3). The BMWP, ABI and FR indices indicated good water condition in most of the sites evaluated, excepting those of the Yacoraite River. The ASPT indicated that all the sites were impacted, except the Lozano and Yala rivers (Table 2). The EPT and EIPT indices classified as 'not impacted' the sites located in the Yungas, while the IBY indicated contamination in sites of the Río Grande within the Yungas, but not in the tributaries of

this ecoregion. The %EPT was high (generally, >50%) and the %Chironomidae was low in the Yungas sites (mean 20%), while in the Monte and the Puna averaged 49%.

The axis 1 of the PCA performed including data from tributaries, correlated positively with all the biotic indices, except with the Chironomidae/total. Axis 1, corresponding to the Río Grande PCA, correlated positively with the EPT, EIPT and ASPT indices (Table 3).

Table 2. Biotic index scores 'at each sampling site (site code in Table 1). *: impacted.

Tabla 2. Valores de los índices bióticos en cada sitio de muestreo (código de cada sitio en la Tabla 1). *: impactado.

Site and date ¹	EPT	%EPT	EIPT	IBY-4	%Chiro/total	BMWP	ASPT	ABI	FR
AP11	3*	2.3	4*	2*	44	76	4*	74	18
AP12	7*	57.2	5	2*	20.7	115	4*	114	28
PI11	5*	0.2	4*	2*	12.1	73	4*	72	19
PI12	6*	1.2	3*	2*	53.9	122	4*	118	30
JU11	3*	0.2	4*	2*	92.1	69	4*	67	17
JU12	2*	10.9	3*	2*	76.4	69	4*	66	17
TU11	3*	9.2	4*	2*	66.6	62*	4*	60*	15
TU12	5*	21.9	5	2*	59.1	109	4*	106	26
YA11	9	54.0	4*	2*	36.9	112	5*	106	22
YA12	12	87.8	6	2*	8.5	81	5*	79	15
LM11	11	39.9	3*	1*	52.8	71	5*	64*	15
LM12	12	92.6	7	1*	4.9	80	5*	77	16
AP21	3*	0.2	4*	2*	82.8	81	4*	81	21
AP22	5*	3.1	4*	2*	35.6	96	4*	98	25
PI21	3*	0.8	3*	2*	42.3	85	4*	82	22
PI22	4*	2.5	4*	2*	49.5	76	4*	79	21
JU21	4*	1.8	4*	2*	77.4	85	4*	85	22
JU22	4*	17.7	3*	1*	69.1	105	4*	99	27
TU21	3*	0.3	3*	2*	16.1	81	4*	81	21
TU22	5*	8.0	5	2*	72.5	101	4*	100	25
YA21	12	34.7	7	3	37.9	122	5*	110	23
YA22	10	73.7	7	2*	12	92	4*	85	21
LM21	11	60.9	8	3	30.8	98	5*	95	19
LM22	13	63.6	7	1*	25.4	85	5*	82	19
YC31	3*	4.4	2*	2*	50	61*	4*	59*	14*
YC32	5*	4.1	5	2*	45.9	59*	4*	57*	14*
HU31	10	28.2	8	3	51.3	129	5*	127	25
HU32	14	24.5	8	2*	21	101	5*	98	21
PU31	6*	3.0	4*	2*	43.3	105	4*	103	24
PU32	6*	8.6	4*	2*	6.3	87	5*	85	19
LO31	17	69.1	11	4	3.9	197	6*	185	33
LO32	16	54.0	10	4	8.1	169	7	151	25
YL31	19	56.7	12	4	3.7	211	7	199	32
YL32	17	54.1	10	4	5.3	184	6*	168	30
XX31	18	46.7	13	2*	13.1	198	5*	187	37
XX32	12	33.3	8	3	35.6	154	6*	146	27

¹ First two letters indicate site (see Table 1), first number indicates year (1=2012, 2=2013, 3=2014), second number indicates season (1=dry, 2=wet).

Table 3. Spearman's correlation analysis between biotic indices and values of axis 1 of the principal component analysis. (1)=PCA corresponding to the Río Grande, (2)=PCA corresponding to the tributaries of the Río Grande basin. *: significant correlations ($r \geq 0.5$, $P' < 0.05$).

Tabla 3. Análisis de correlación de Spearman entre los índices bióticos y los valores del eje 1 de los análisis de componentes principales. (1)=ACP correspondiente al Río Grande, (2)=ACP correspondiente a los tributarios de la cuenca del Río Grande. *: correlaciones significativas ($r \geq 0.5$, $P' < 0.05$).

	PC1(1)	PC1(2)
EPT	$r=0.6$, $P=0.001^*$	$r=0.8$, $P=0.001^*$
EIPT	$r=0.5$, $P=0.02^*$	$r=0.8$, $P=0.0007466^*$
IBY-4	$r=-0.1$, $P=0.5$	$r=0.6$, $P=0.02^*$
%Chiro/total	$r=-0.4$, $P=0.04$	$r=-0.5$, $P=0.1$
BMWP	$r=-0.02$, $P=0.9$	$r=0.9$, $P=2.2e-16^*$
ASPT	$r=0.7$, $P=6.608e-05^*$	$r=0.6$, $P=0.03066^*$
ABI	$r=-0.1$, $P=0.6$	$r=0.9$, $P=2.2e-16^*$
FR	$r=-0.42$, $P=0.03$	$r=0.9$, $P=3.524e-05^*$

The EPT and EIPT indices correlated negatively with NaCl, TDS, EC and altitude (which characterized the sites of the Puna and the Monte ecoregions). IBY was negatively correlated with CE and TDS (Table 4). The abundance of the community decreased above 1500 $\mu\text{S}/\text{cm}$ of EC, 1500 m a.s.l., 300 ppm NaCl and 300 ppm TDS (Figure S1-3). The %Chironomidae showed positive correlations with EC and NaCl (Table 4, Figure S4). The correlations between EPT, EIPT, IBY and %Chironomidae indices with other parameters (i.e., discharge, width, depth, temperature, pH and dissolved oxygen) were not significant.

Table 4. Spearman's correlation analysis between ETP, EIPT, ABY-4 and %Chironomidae/total indices and physicochemical parameters of Río Grande basin evaluated during the 2012-2014 period. *: significant correlation ($r \geq 0.5$, $P' < 0.05$).

Tabla 4. Análisis de correlación de Spearman entre los índices ETP, EIPT, ABY-4 y %Chironomidae/total y los parámetros fisiocoquímicos de la cuenca del Río Grande evaluados durante el período 2012-2014. *: correlación significativa ($r \geq 0.5$, $P' < 0.05$).

	EPT	EIPT	IBY-4	%Chiro/total
Discharge	$r=0.0$, $P=0.9$	$r=-0.0$, $P=0.7$	$r=-0.2$, $P=0.2$	$r=-0.1$, $P=0.6$
Width	$r=-0.9$, $P=0.6$	$r=-0.2$, $P=0.3$	$r=-0.4$, $P=0.03$	$r=-0.1$, $P=0.5$
Depth	$r=0.1$, $P=0.6$	$r=0.0$, $P=0.8$	$r=0.0$, $P=0.9$	$r=0.0$, $P=0.9$
NaCl	$r=-0.8$, $P=1.6e-08^*$	$r=-0.7$, $P=6.6e-07^*$	$r=-0.4$, $P=0.01$	$r=0.5$, $P=0.001^*$
TSD	$r=-0.5$, $P=0.02^*$	$r=-0.5$, $P=0.005^*$	$r=-0.5$, $P=0.002^*$	$r=0.3$, $P=0.1$
Altitude	$r=0.6$, $P=6.7e-05^*$	$r=0.6$, $P=0.0002^*$	$r=0.1$, $P=0.6$	$r=0.4$, $P=0.01$
EC	$r=-0.7$, $P=1.9e-07^*$	$r=-0.7$, $P=9.3e-07^*$	$r=-0.5$, $P=0.003^*$	$r=0.5$, $P=0.001^*$
DO	$r=-0.0$, $P=0.8$	$r=0.0002$, $P=0.1$	$r=0.2$, $P=0.3$	$r=0.0$, $P=0.8$
T°	$r=0.1$, $P=0.3$	$r=0.2$, $P=0.2$	$r=-0.2$, $P=0.3$	$r=-0.3$, $P=0.06$
pH	$r=0.0$, $P=0.9$	$r=0.0$, $P=0.9$	$r=-0.2$, $P=0.3$	$r=0.2$, $P=0.2$

DISCUSSION

The physico-chemical parameters of the Río Grande basin showed natural gradients following the altitudinal range. The salinity (NaCl, TDS and EC) showed higher values in high sites and lower values downstream, contrasting with previously reported patterns (i.e., increase in salinity downstream) in other mountain basins (Medina et al. 1997; Zelarayán Medina and Salas 2014). This is due to the fact that the high altitude sites are in an area of 'low slope and high evapotranspiration (Puna and Monte). The high precipitation of the Yungas increase the discharge decreasing the relative ionic load of the waters downslope.

The electrical conductivity values 'of the sites sampled in the Río Grande basin located in the Puna and the Monte were higher than those reported by Nieto et al. (2016), Hankel et al. (2018) and Pero et al. (2019) for different rivers of arid zones of northwestern Argentina. The other sites located in Yungas showed values 'of EC and TDS similar to those reported previously (Fernández et al. 2002; Colla et al. 2013; Zelarayán Medina and Salas 2014; Pero et al. 2019), while the main channel showed somewhat higher values.

The fact that differences in the discharge were negligible between the dry and rainy seasons is not surprising since our sampling coincided with the beginning of the rainy season (late spring) when the rainfall volume was still low to significantly affect the flow. A similar situation was observed by Fernández et al.

(2002) in the Lules river, located in the Yungas of the province of Tucumán. In the dry period, higher DO levels were detected, probably related to the low winter temperatures.

According to our results, the general state of the Río Grande basin is good, although just a single point was sampled below the capital city of San Salvador de Jujuy. However, Fernández et al. (2006) warned about the contamination in the main channel of the Río Grande caused by the activities of this city. The impact detected by several BI at one of our high altitude sites (Yacoraite River) could be related to mining activity as there is evidence of presence of toxic metals in the tailing dams of Mina el Aguilar (Kirschbaum et al. 2012).

Regarding the ASPT, which classified almost all the sites as impacted, we believe that the cutoff value proposed by Dos Santos et al. (2011) to separate impacted from non-impacted sites is very strict, and should be re-evaluated. The ABI (for high-altitude rivers), IBY, EPT and EIPT indices (with cutoff

values 'proposed only for the Yungas) cannot be applied throughout the entire basin due to their restricted scopes. Hankel et al. (2018), suggested that the IBY, EPT and EIPT indices are inadequate to evaluate arid zones, since they characterize as contaminated sites with naturally poor communities that are highly adapted to extreme environmental conditions. Only the BMWP and FR indices could be used in the entire basin, and their performance coincided with the physico-chemical variables and the performance of the other BI. For this reason, we suggest the application of BMWP and FR, which are suitable for monitoring water quality along heterogeneous basins in terms of altitude and ecoregions, like the Río Grande basin.

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