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Water quality index including periphyton chlorophyll-*a* in forested urban watersheds from Tierra del Fuego (Argentina)

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ABSTRACT

The city of Ushuaia, Argentina, has experienced a rapid and unorganized population growth with possible consequences on the dynamics and functioning of urban lotic ecosystems. Urban expansion was not largely accompanied by the development of infrastructure for the provision of services such as drinking water and sewers to all the population. Currently, only 65% of the population is connected to the main sewage system (pre-treatment) and the city does not have a secondary wastewater treatment plant yet. We evaluated the impact of wastewater discharges and non-point contamination on the water quality of the three main water courses that cross the city of Ushuaia: Pipo river (PR), Buena Esperanza stream (BES) and Arroyo Grande river (AGR). We sampled four sites per watershed from spring to autumn in two consecutive years (2018 and 2019): S1 (not urbanized at the upper section), S2 (transitional section), S3 (urbanized at the middle-low section), and S4 (urbanized, close to the outlet). We developed the Fuegian Water Quality Index (F_WQI) as a tool for environmental monitoring in forest streams which includes several indicators of organic inputs (dissolved oxygen, nitrogen-ammonium, total phosphorus, fecal coliform bacteria) and periphyton chlorophyll-*a* (Peri Chl-*a*). The variables were ranked and weighed, and the index was constructed using a mathematical formula. Urbanization negatively impacted the water quality of these ecosystems; the F_WQI showed "very bad" and "bad" water quality categories at S4 in BES and AGR, both sites impacted by wastewater discharges and large percentage of urbanized area. Likewise, in the PR watershed, with less urbanization and sewage discharges, the F_WQI indicated "very good" water quality. The F_WQI summarizes the water quality of forested watersheds in a simple numerical scale, resulting in a useful tool for monitoring studies in temperate rivers and streams. We encourage the inclusion of Peri Chl-*a* in water quality indices, due to its ubiquity in these ecosystems and the fact that it increased the sensitivity of the index.

1. Introduction

Streams and rivers are open ecosystems with dynamic imports and exports of nutrients, energy and water (Karr and Dudley, 1981; Allan and Castillo, 2007). Their characteristics are significantly controlled by lateral, longitudinal and vertical interactions with their surroundings, which imply that these ecosystems are sensitive to perturbations. Changes in land use are considered one of the main global alterations, which in turn modify the structure and functioning of ecosystems (Vitousek et al., 1997; Grimm et al., 2000; Hagazy and Kaloop, 2015). Urbanization is a persistent and quickly-growing type of land use which results in major disturbances to ecological process in streams (Paul and Meyer, 2001).

Studies on urban ecology usually involve the urban development on native ecosystems and the dynamics of urban environments as ecosystems in themselves (Grimm et al., 2000). In fact, there are several studies that mention the term "urban stream syndrome", referring to indicators associated with deterioration of the water chemistry, changes in morphology and hydrology, and others related to urbanization (Walsh et al., 2005; Askarizadeh et al., 2015; Vietz et al., 2016). Some of the associated symptoms are the increase of nitrogen, phosphorus, toxicants and water temperature; changes in water discharge over time, increases

Abbreviations: Peri Chl-*a*, Periphyton chlorophyll-a; F_WQI, Fuegian Water Quality Index.

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to channel width and pool depth, among others (Walsh et al., 2005).

The water quality of urban watersheds has been a topic of great interest, widely studied in different parts of the world (Characklis and Wiesner, 1997; Hatt et al., 2004; Bahar et al., 2008; Kaushal and Belt, 2012). The input of pollutants and sediments is common in urban streams, which increases the concentration of nutrients and alters their biota (Miserendino et al., 2008). Additionally, increased run-off, facilitated by high coverage of impervious surface, as well as changes in landscape conformation, typically leads to higher nutrient loads in streams and rivers (Walsh et al., 2005; Schiff and Benoit, 2007; Fernandes et al., 2019). In water quality studies, the values of specific and standardized variables in local regulations are compared. Although this methodology allows the identification of different sources of contamination and law enforcement (e. g., Environmental Law N°55 in Tierra del Fuego), it does not provide a comprehensive view of temporal and spatial trends in the entire watershed (Debels et al., 2005).

There is an extensive literature on Water Quality Indices (WQI). Horton (1965) and Brown et al. (1970) were pioneers in developing the indices to assess contamination in aquatic ecosystems. Since then, many different methods for the calculation of WQI have arisen (Smith, 1989; Khan et al., 2003; Sarkar and Abbasi, 2006). Indices employ many of the same variables (e. g., dissolved oxygen, temperature, ammonium), but they differ in the way the parameter values are integrated and interpreted. The use of WQI in contamination studies is a very valuable tool, since it summarizes in a simple numerical scale the degree of pollution for a given water resource. It allows comparisons of the watersheds water quality in space and time, which helps for instance, to evaluate the progress of monitoring programs and simplifies the communication of the results (Tyagi et al., 2013). Also, many studies use biological indicators to perform water quality evaluation since biological communities reflect a combination of current and past watershed conditions (Lobo et al., 2004; Munné and Prat, 2009; Gelis et al., 2020). The use of periphyton chlorophyll-*a* for biomonitoring studies has a long tradition (Biggs, 1989; Lowe et al., 1986; Arini et al., 2012; Pandey et al., 2014) but there are few examples in which this variable is included in the calculation of a water quality index (e.g. Zandbergen and Hall, 1998).

Ushuaia city, located in the province of Tierra del Fuego, Argentina, has undergone major transformations in its natural landscape (Orzanco, 1999). In the last 20 years, the population doubled and had a higher growth rate than the rest of the cities of the Patagonian region (Gessaga and Frias, 2010). The main reason for this population increase has been the promulgation of the Argentinian National Law N° 19.640 of industrial promotion and customs benefits, which motivated immigration from the central and northern provinces of Argentina. These facts led to substantial changes in land use, mainly due to the need of space for urban and industrial settlement. Urban expansion was not largely accompanied by the development of the necessary infrastructure for the provision of services such as drinking water and sewers to all the population. Up to date, the city does not have a sewage treatment system, which is reflected in the degradation of fluvial ecosystems that cross the city and in the coast of the Beagle Channel as it receive the input of sewage effluents (Amin et al., 2011).

There is very little information about the water quality of lotic ecosystems in Ushuaia City, Argentina (Diodato, 2013; Zagarola et al., 2017). The objectives of this study are twofold. First, our aim is to evaluate the impact of wastewater discharges and non-point sources of contamination on the water quality of the three main water courses that flow through the city of Ushuaia. Second, we developed the Fuegian Water Quality Index (F_WQI) as a tool for environmental monitoring in forested urban watersheds. The F_WQI aims to integrate in a simple numeric scale the chemical, physical and biological characteristics of forested streams. We sampled three water courses from the headwaters to the outlet, before and after crossing the city of Ushuaia to study spatial variation. In addition, this work was developed during two consecutive years to evaluate better encompass temporal variation. We expected to find reduced water quality in sampling sites downstream the city, as they would be more affected by the input of wastewater and run-off associated with urbanization. We explored this idea with a PCA analysis for each sampling year. We predicted the F_WQI to be lower in the sites nearby the city of Ushuaia.

2. Materials and methods

2.1. Study area

The study area (Fig. 1) is located in the city of Ushuaia, southwest of the Argentinian region of Tierra de Fuego Island (54.5° S, 68.2° W). The landscape is composed of large mountain ranges and glacial modeling valleys. Deciduous forests are dominated by *Nothofagus pumilio. N. pumilo* also coexists in mixed forests with the perennial species *N. betuloides*, alternated with high Andean vegetation communities and peat bogs at lower elevations (Moore, 1983; Henn et al., 2016). The climate of the area is characterized by an annual mean temperature of 5.9 °C and annual precipitations of ca. 580 mm in Ushuaia, with ca. 20% in the form of snow (Rodríguez et al., 2020). The water courses in the study area have a snowy-rainy regime and are also glacial fed, particularly between January and March (Iturraspe and Urciolo, 2000).

Our study was carried out in three forested water courses that cross the city and flow into Beagle Channel: Pipo river (PR), Buena Esperanza stream (BES) and Arroyo Grande river (AGR). These main water courses have different drainage areas and in their upstream sections are sources of drinking water to the local population. The area of BES watershed is smaller than AGR and PR watersheds; it is less than a quarter of the other two, while the PR is slightly larger than AGR (Table 1). In relation to the total area, the identified percentage of urbanization is higher at BES than in the other two watersheds (Table 1).

We calculated the area of each watershed and its percentage of urbanization through the QGIS 3.10. software. First, each watershed was modeled as polygons, using the “Rwatershed” tool. Then, we calculated the area of each polygon with the raster calculator tool. The percentage of urbanized area was calculated from the overlapping layers: the area of intersection between the satellite image of the city (updated, extracted from Google Earth) and the total area of each watershed.

We sampled 12 sites, four belonging to each watershed: S1 (reference, not urbanized at the upper section), S2 (transitional section), S3 (urbanized at the middle-low section), and S4 (urbanized, close to the outlet) (Fig. 1). Due to that watersheds are located in mountainous topography, there is a marked altitude gradient between S1 and S4 (Table 2).

2.2. Field sampling and analytical procedures

Water and periphyton samples were collected and *in situ* measurements were carried out from austral spring to autumn in 2018 and 2019. The samplings were made during those seasons to maximize differences among sites provided higher productivity. The first-year sampling was performed on January, March and April 2018 every 30–40 days. The second-year sampling was performed on November 2018, January and March 2019, with approximately the same frequency as in the previous year. At each sampling site, water velocity, depth and wet width were taken in three transects located at ten meters intervals; water velocity and depth were measured with a flow meter (Global Water FP111) and the wet width was measured with a measuring tape from one side to the other of the course in contact with water. Water discharge (Q) was calculated from these data by using the velocity-area method following Gordon et al. (2004). During the first year, water discharge at lower sites in AGR was not possible to measure because of logistic constraints.

Dissolved oxygen (DO) and water temperature were recorded *in situ* using a Lutron DO 5510 oximeter, turbidity was measured with a Lutron portable turbidimeter TN3024 while conductivity and pH were registered with a HANNA HI 98129 portable sensor. Water samples were collected in PVC bottles (previously rinsed with 2% HCl and milliQ

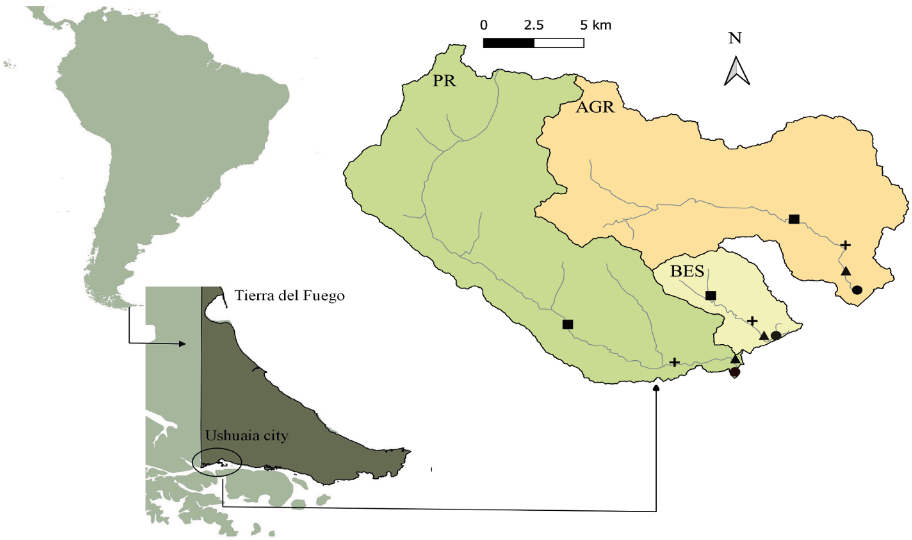

Fig. 1. Map of the study area (Ushuaia city) located in South America, province of Tierra del Fuego, Argentina showing the studied watersheds and sampling sites: Pipo river (PR – green area), Buena Esperanza stream (BES – beige area) and Arroyo Grande river (AGR – orange area). Sampling sites: S1 (reference, not urbanized at the upper sections – square), S2 (transitional, semi-urbanized – cross), S3 (urbanized – triangle), S4 (urbanized, close to the outlet – circle). (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

Table 1
Morphometric variables estimated in the three studied watersheds: Pipo river (PR), Buena Esperanza stream (BES) and Arroyo Grande river (AGR).

Water course	Watershed Urbanization (%)	Watershed Size (km^2)
PR	2.6	159.0
BES	27.4	23.0
AGR	6.6	133.2

Table 2
Geographical coordinate (in GMS) and altitude over sea level for each sampling site. Pipo river (PR), Buena Esperanza stream (BES), Arroyo Grande river (AGR). Each geographic coordinate begins with the same degrees south and west (58° S, 62° W); for simplicity, they were placed in the heading of the column (*).

Water course	Site	Geographical coordinate (54° S; 68° W) *	Altitude over sea (m)
PR	S1	48′ 56″ S; 28′ 42″ W	120
	S2	49′ 56″ S; 23′ 56″ W	68
	S3	49′ 52″ S; 20′ 50″ W	23
	S4	50′ 12″ S; 20′ 52″ W	6
BES	S1	47′ 55″ S; 22′ 15″ W	353
	S2	48′ 38″ S; 20′ 22″ W	68
	S3	49′ 5.9″ S; 19′ 45″ W	13
	S4	49′ 4.4″ S; 19′ 17″ W	5
AGR	S1	45′ 33." S; 18′ 23″ W	184
	S2	46′ 20″ S; 16′ 7.8″ W	114
	S3	47′ 3.2″ S; 16′ 1.0″ W	82
	S4	47′ 36″ S; 15′ 18″ W	5

water) and were kept under dark conditions until chemical analyses. In the laboratory, the concentration of dissolved inorganic nutrients, total nitrogen (TN), total phosphorus (TP) and suspended solids (SS) were assessed. Water samples for the study of dissolved inorganic nutrients were filtered through Whatman GF/F filters (0.7 µm nominal pore size). Samples for nitrogen-ammonium ($N\text{-}NH_4$) were analyzed within the day or the following day with the phenol–hypochlorite method (APHA, 2005). A volume of the filtered samples was frozen (−20 °C) and later analyzed as follows: phosphate ($P\text{-}PO_4$) with the ascorbic acid method, nitrogen-nitrite ($N\text{-}NO_2$) with the diazotization method, and nitrogen-nitrate ($N\text{-}NO_3$) with the same method as $N\text{-}NO_2$ with a previous cadmium reduction (APHA, 2005). For $P\text{-}PO_4$, $N\text{-}NO_2$ and $N\text{-}NO_3$ determinations, a Hach DR/2700 spectrophotometer was used along with Hach reagents (Hach Company, CO, USA). For total nutrients (TN and TP), water samples were digested according to Valderrama (1981) and measured as $N\text{-}NO_3$ and $P\text{-}PO_4$, respectively following the same methodology indicated for dissolved nutrients. Suspended solids were collected by filtration on pre-dried Whatman GF/C filters (1 µm nominal pore size), dried to constant weight at 105 °C, and weighed to the nearest 0.1 mg (APHA, 2005).

2.3. Periphyton assessment

At each sampling site, several stones from the watershed were taken from inside a 20x20 cm quadrant. The quadrant was randomly thrown in triplicate in the stream section (30 m) and the stones were transported in cold and dark conditions in plastic bags to the laboratory. Stones area was estimated in the laboratory by wrapping each one with aluminum foil, weighing the foil, and compared with a standard area. The materials attached to the stones were scraped with a brush and then suspended in a known volume of distilled water which was filtered through Whatman GF/F filters (0.7 µm nominal pore size) and the filter stored at −20 °C in the freezer until analysis. The periphyton chlorophyll-*a* (Peri Chl-*a*) was determined following hot ethanol extraction (60–70 °C) and absorbance measurements at 665 and 750 nm before and after acidification with 0.1 N HCl (Jespersen and Christoffersen, 1987).

2.4. Coliform bacteria

To estimate bacterial numbers, water samples were collected in sterile flasks and preserved at 4 °C until analysis within 24 h. The Most Probable Number (MPN/100 ml) technique was employed to estimate Total and Fecal Coliform (TC and FC) (APHA, 2005) using 24 h Colilert test (IDEXX Laboratories, USA). In the second sampling year, to minimize costs, we only analyzed these variables in March, which was the time-period when the positive results were concentrated the previous year.

2.5. Water quality index calculation

To evaluate the impact of wastewater discharge and non-point sources of contamination on the river/stream water quality, we developed the Fuegian Water Quality Index (F_WQI) according to the equation used by Berón (1984):

$$F\_WQI = \frac{\sum_{i=1}^{n} Qi}{\sum_{i=1}^{n} Wi} \times 10$$

where, Q_i: weighted value of the variable i, (Table 3), W_i = relative weight of the variable i, and n = number of variables (in this case n = 5).

To select the variables to be included in the F_WQI, we took as a

Table 3
Tables of classification for each variable selected to construct the F_WQI: Dissolved oxygen (DO), ammonia ($N\text{-}NH_4$), Total phosphorus (TP), periphyton chlorophyll-a (Peri Chl-*a*) and fecal coliforms (FC). The first column shows the range of each variable (concentration or number of bacteria) and the second column show the value of weighted classification (Q_i). The last column indicates the references used as a guide.

Variable	Range	Q_i	References
DO (mg L^{-1})	0–2	0	Cude (2001)
	2–4	2	
	4–6	4	
	6–8	6	
	8–9	8	
	> 9	10	
$N\text{-}NH_4$ (mg L^{-1})	0 – 0.001	30	Cude (2001)
	0.001–0.01	24	Miserendino et al. (2008); (2011)
	0.01–0.02	18	
	0.02–1.0	12	
	1.0 – 3.0	6	
	3.0 – 5.0	3	
	> 5.0	0	
TP (mg L^{-1})	0 – 0.025	10	Dodds et al. (1998; 2004; 2006); Omernik (1977);
	0.025–0.049	8	Van Nieuwenhuyse and Jones (1996)
	0.049 – 0.09	6	
	0.09–0.25	4	
	0.25–0.75	2	
	> 0.75	0	
Peri Chl-*a* (mg m^{-2})	0–1,7	20	Biggs (1996);
	1.7–2.5	18	Dodds et al. (1998; 2006); Nordin (1985)
	2.5–5.0	15	
	5.0–10.0	10	
	10.0–21.0	5	
	21.0–84.0	3	
	> 84.0	0	
FC (NMP/100 ml)	0	30	Cude (2001); Environmental Law 55 (Tierra del Fuego province)
	0–10	24	
	10–450	18	
	450–1000	12	
	1600–3000	6	
	3000–5000	3	
	> 5000	0	

reference those variables that are mostly related to organic contamination. Thus, the variables included are: DO, $N\text{-}NH_4$, TP, Peri Chl-*a* and FC. All these, showed the wider range of variation (CV) in relation to the rest of the measured variables in this study, with the exception of DO (Appendix A1).

The relative weight (W_i) defines the importance of each variable in the F_WQI; the variables with the highest weight were FC and $N\text{-}NH_4$ ($W_i = 3$) because they are direct indicators of the effect of domestic wastewater provided their concentration increase in watercourses with the input of domestic waste. Even though $N\text{-}NH_4$ may be associated with natural and urban runoff carrying decomposed organic matter such as plants leaves and organic litter, and the presence of coliform bacteria may be related to the feces of the animals present in the study area (for example, horses, dogs, beavers), we expected a larger contribution of ammonium and fecal coliforms from domestic origin as there is no sewage treatment plant. We gave a weight of 2 to Peri Chl-*a* ($W_i = 2$) as this community responds to nutrients increase. Finally, for DO and TP we assigned the lower weight ($W_i = 1$). The concentration of DO, which is a useful metric in contamination studies, has little variation in our study area because of the low environmental temperature, being oversaturated most of the time (Appendices B.1 and B.3). As watersheds in the area are very low in nutrients (Rodríguez et al., 2020), we expected an increased concentration of TP due to external loadings. TP and Peri Chl-*a* are in general highly correlated (Van Nieuwenhuyse and Jones, 1996); therefore, we gave to TP a lower weight, to avoid redundancy in the information as Peri Chl-*a* weight was defined as 2.

For each variable included in the index, we constructed a classification table, with the purpose of obtaining weighted classification values (Q_i), used to transform the different units and dimensions of the variables to a common scale (Table 3). According to Berón (1984), the range of variation of each classification table is relative to the W_i assigned to each variable (for example: $W_i = 1$, the classification table ranged from 0 to 10; with $W_i = 3$, ranged from 0 to 30). We assigned to each concentration range a value of weighted classification (Q_i) taking into account an extensive literature review (Table 3).

The sum of all weights must be equal to 10. Further, in case of not having the value of any given variable, an estimate of the F_WQI can be obtained by dividing the total of the classification values (Q_i) by the sum of the relative weights of the remaining variables.

The water quality was evaluated by calculation of the F_WQI for each sampling site and date. This index does not have units; it ranges from 0 to 100 with the following categories: 0–25 *Very Bad*, 26–50 *Bad*, 51–60 *Medium* 61–75 *Good*, 76–90 *Very Good*, 91–100 *Excellent*. These ranges were adapted for the F_WQI taking into account the National Sanitation Foundation (NSF, http://bcn.boulder.co.us/basin/watershed/wqi_nsf.html) earlier developed by Brown et al. (1970).

2.6. Data analysis

We used a Generalized Linear Mixed Model (GLMM) to compare variables in space with "year" as a random effect for "site" and "month" as a fixed effect (Zuur et al., 2009). We analyzed both sampling years together ($n = 6$) to obtain more statistical power, since each year of sampling had 3 samples per site. These analyses were performed comparing the different sections of each river/stream (S1, S2, S3, and S4). Prior to analysis, we tested the homoscedasticity of the variance and the normality of the data. Although normality was not fulfilled (only DO achieved normality), the selected statistical test allows us to adjust the probability function that fits the data best (since all variables are whole real numbers, the probability functions that best adjusted were lognormal and gamma). To further explore differences among sites for significant variables in the GLMM, *post hoc* tests (Tukey) were also performed for multiple comparisons.

Two Principal Component Analysis (PCA) were performed for each sampling year in order to explore the presence of trends in all sites and dates regarding environmental variables. For this analysis we used the variables included in the F_WQI (DO, TP, $N\text{-}NH_4$, Peri Chl-*a*) with the exception of fecal coliforms because it introduced bias on the results (estimate based on ranks). In order to compare F_WQI values between years, non-parametric tests were applied. We used the site average per year of sampling, provided we did not observe significant variation among months for each sampling year ($p > 0.05$; Kruskall Wallis test). For all statistics we used the R 3.5.0 and RStudio software (packages: factoextra, FactoMineR, lme4 and multicomp).

3. Results

3.1. Morphometric and environmental parameters

Buena Esperanza stream (BES) locations experienced lower water discharge at all sampling sites ($<1\ m^3\ s^{-1}$, $n = 4$) compared to the others two water courses ($>1.50\ 1\ m^3\ s^{-1}$ at AGR and $>2\ m^3\ s^{-1}$ at PR) during the two years sampled (Fig. 2). In addition, we detected on average higher water discharge during the second year of sampling in almost all the sampling sites (Fig. 2).

The average water temperature ranged between 5.4 °C and 8.7 °C in the first year (Appendix B.1), while in the second year mean values ranged between 5.7 °C and 9.1 °C (Appendix B.2). Concentrations of dissolved oxygen (DO) always exceeded 10 mg L^{-1} and did not differ significantly among watersheds or sampling sites (Appendices B.1 and B.2). The pH was circumneutral in most sites and all watersheds under

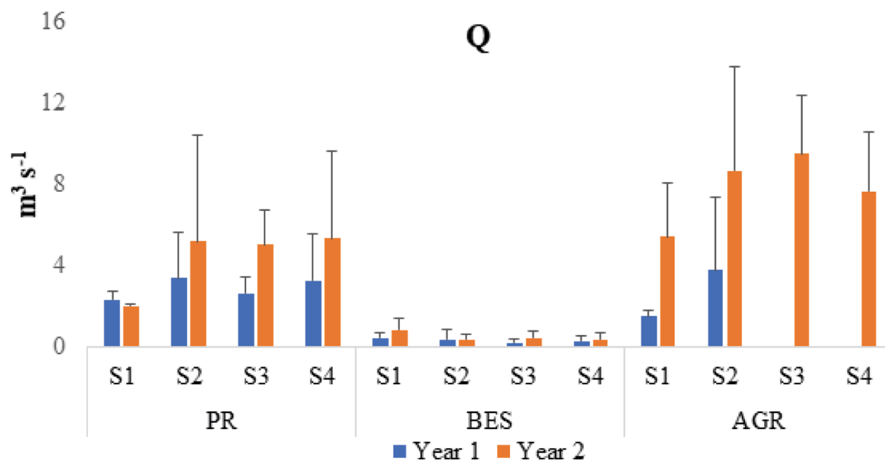

Fig. 2. Water discharge (Q, in $m^3 s^{-1}$), time-averaged values (+1 SD; n = 3) at each sampling site (S1, S2, S3, S4) corresponding to Pipo river (PR), Buena Esperanza stream (BES) and Arroyo Grande river (AGR) during the first year (blue bars) and second year (orange bars) of study. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

study in both years (Appendices B.1 and B.2). Conductivity values were higher in the lower sections (S4) in both sampling years (Appendices B.1 and B.2). Turbidity on average, oscillated between 0.3 and 48.9 NTU in the first year, and between 2.4 and 45.4 NTU in the second year. In both years, the low section (S4) of BES was on average more turbid than the other two water courses (Appendices B.1 and B.2).

All stream systems had relatively low nutrient concentrations (Appendices B.1 and B.2). Nevertheless, higher values for some nutrients were found in the lower sites (S4) of BES and AGR (Appendices B.1 and B.2). Nitrogen-ammonium ($N\text{-}NH_4$) concentrations differed among watersheds; PR showed the lowest concentrations (below 0.05 mg L^{-1}), while at BES we detected higher concentrations (>4 mg L^{-1}) and AGR reached intermediate concentrations (ca. 0.4 mg L^{-1}). The concentration of $N\text{-}NH_4$ in PR did not show significant variations among sampling sites (Fig. 3a), unlike BES, where the highest concentration was found at S4 ($p = 0.001$; Fig. 3b). Regarding AGR, although $N\text{-}NH_4$ concentration increased towards S4, no significant differences were detected among sampling sites (Fig. 3c). PR had higher $N\text{-}NH_4$ concentrations in January at all sampling sites ($p = 0.034$, data not shown).

Total phosphorus (TP) concentrations showed slight fluctuation among watersheds. At PR and BES, the maximum concentrations of TP reached were around 1 mg L^{-1}, while at AGR the highest value was 0.72 mg L^{-1}. TP concentrations at PR differed significantly between S3 and S4 ($p = 0.047$; Fig. 4a). At BES and AGR, the TP concentration in S4 was significantly higher than S1 and S2 ($p = 0.022$ and 0.007, respectively for BES, Fig. 4b, and $p = 0.001$ and 0.030 respectively for AGR, Fig. 4c). BES and AGR had higher TP values in April at all sampling sites ($p < 0.05$, data not shown).

3.2. Biological parameters

Peri Chl-*a* concentrations increased towards lower sampling sites and differed among watersheds with higher values found at BES and AGR (ca. 150 mg m^{-2}, Fig. 5a and b). In general, no differences were detected among sampling months with the exception of PR in November 2018, where lower values were found for all sites ($p = 0.044$).

In the first sampling year, fecal coliforms (FC) were found at higher amounts at the lower sites (S4) of BES and AGR (Appendix B.3). Whilst, in PR, FC were only detected in S4 in January 2018 but in smaller quantities than BES and AGR. FC were not detected at any site in March 2019 (Appendix B.3).

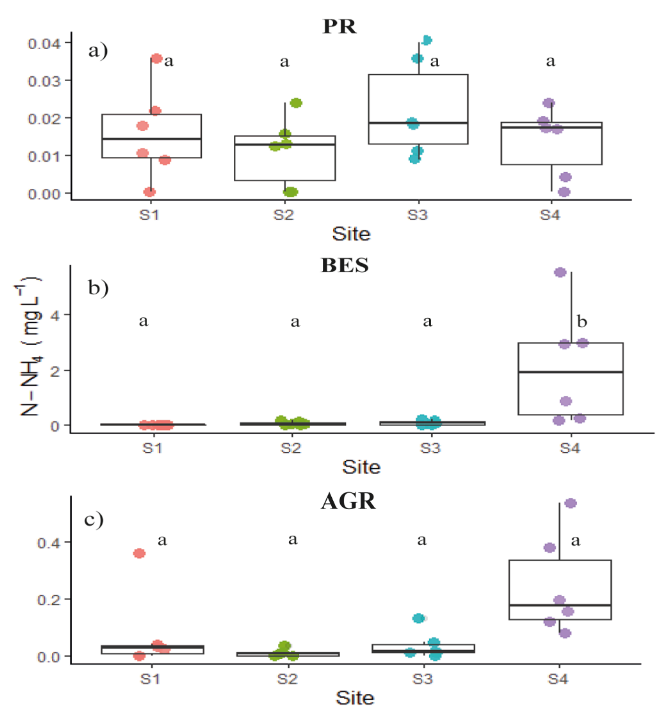

Fig. 3. Boxplot (median, quartile and whiskers CI = 95%) of nitrogen-ammonium concentrations ($N\text{-}NH_4$, in mg L^{-1}; n = 6) during the study period at each sampling site (S1, S2, S3, S4) corresponding to (a) Pipo river (PR), (b) Buena Esperanza stream (BES) and (c) Arroyo Grande river (AGR). Different letters in each figure represent significant differences among sites (based on Tukey HSD, $p < 0.05$). Note the different scale of the y-axis.

3.3. PCA

The PCAs of the two sampled years showed that about 70% of the total variance was explained by the first two dimensions (Figs. 6 and 7). During the first sampling year (Fig. 6), component 1 explained 48.2% of the total variance; the variables that more contributed to this dimension were $N\text{-}NH_4$ (36.9%) and Peri Chl-*a* (27.6%). The variables that contributed more to the second component were DO (65.4%) and to a lesser extent TP (23.1%).

During the second year (Fig. 7), component 1 explained 45.4% of the

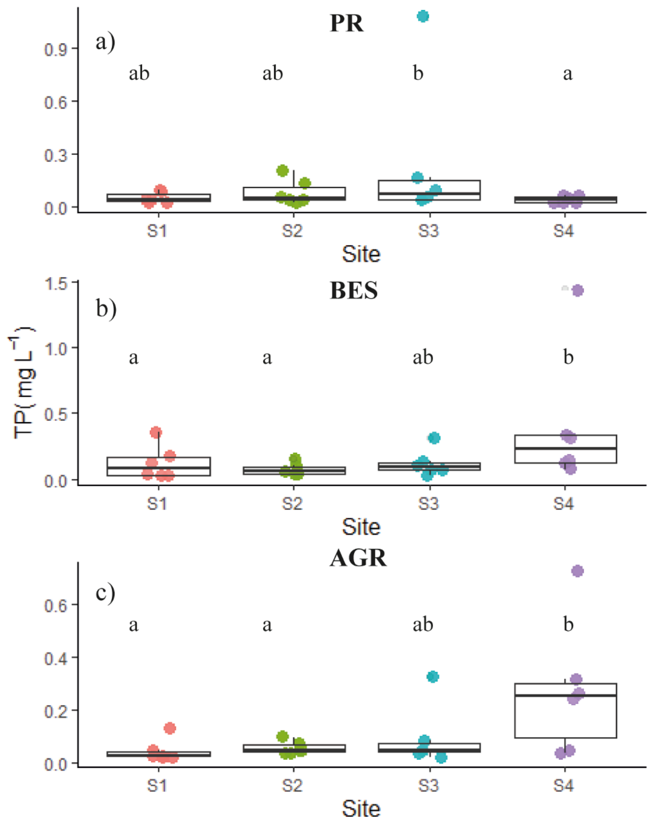

Fig. 4. Boxplot (median, quartile and whiskers CI = 95%) of total phosphorus concentrations (TP in mg L^{-1}, n = 6) during the study period at each sampling site (S1, S2, S3, S4) corresponding to (a) Pipo river (PR), (b) Buena Esperanza stream (BES) and (c) Arroyo Grande river (AGR). Different letters in each figure represent significant differences among sites (based on Tukey HSD, $p < 0.05$). Note the different scale of the y-axis.

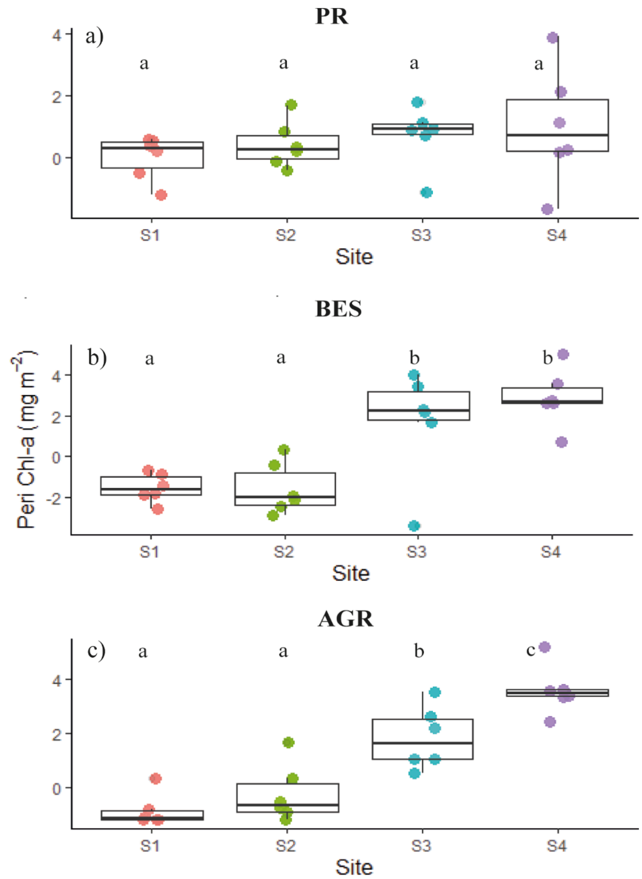

Fig. 5. Boxplot (median, quartile and whiskers CI = 95%) of periphyton chlorophyll-a concentrations (Peri Chl-*a*, in mg m^{-2}; n = 6) during the study period at each sampling site (S1, S2, S3, S4) corresponding to (a) Pipo river (PR), (b) Buena Esperanza stream (BES) and (c) Arroyo Grande river (AGR). Different letters in each figure represent significant differences among sites (based on Tukey HSD, $p < 0.05$). Note scale of the y-axis (log scale).

total variance and, alike the first year, N-NH_4 and Peri Chl-*a* were the variables more related to this component (44.4% and 38.3%, respectively), whilst DO and TP contributed more to dimension 2 (45.4% and 49.4%, respectively) (Fig. 7).

Overall, in both PCA, the sites in the upper sections of the watersheds (squares and crosses in Figs. 6 and 7) were more similar to each other and were negatively related to dimension 1. On the other hand, lower sections at BES and AGR (pink and orange circles) were mostly related to N-NH_4 and Peri Chl-*a* concentrations.

3.4. Fuegian water quality index (F_WQI)

During the first year, on average, water quality was very good at all sampling sites except at low and middle-low sections (S4 and S3) of BES and AGR where, according to the F_WQI, the water quality was worse, ranging from good to very bad (Fig. 8). The F_WQI at PR showed little variation in the longitudinal gradient (86 in S1 to 79 in S4) compared to BES and AGR. At BES the index showed broader variation: it was lower in S4 and S3 (21.5 and 50.3, respectively) in contrast to S2 and S1 (80 and 88, respectively). The F_WQI at AGR watershed ranged on average from 83.3 in S1 to 36.7 at S4, while S2 and S3 were more similar to S1 (79.7 and 70.7, respectively).

Regarding the second year of sampling, the F_WQI in PR showed a narrower range of variation among the studied watersheds, between 90.1 and 78.3. Alike the first year, PR showed very good water quality and was not affected in the different sections of the watershed by organic inputs (Fig. 8). At BES, the F_WQI decreased from S1 to S4 (89.4 to 46.4, respectively). Likewise, AGR presented worse water quality at S4 (43.5) compared to the rest of the sites (91.4 in S1, 92.5 in S2 and 72 in S3).

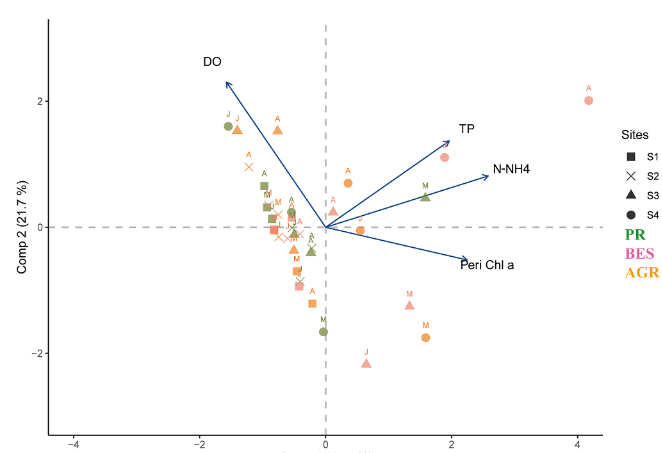

Fig. 6. Principal component analysis (PCA) for the first sampling year considering the quantitative variables used for the construction of the Water Quality Index: Nitrogen-ammonium (N-NH_4), dissolved oxygen (DO), total phosphorus (TP) and periphyton chlorophyll-*a* (Peri Chl-*a*). The sections of the watersheds (sites) are represented by different symbols and colors are used to represent water courses in the different sampling dates: January (J), March (M), April (A).

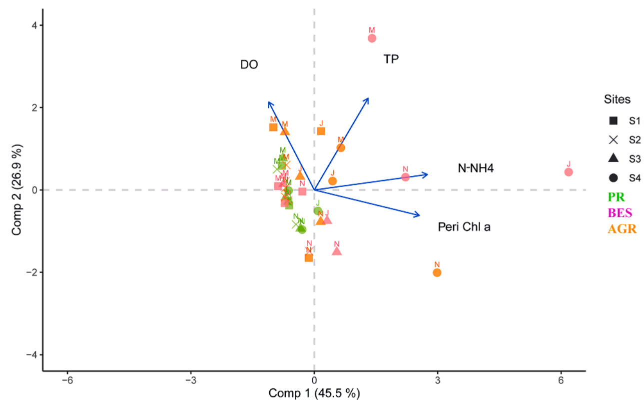

Fig. 7. Principal component analysis (PCA) for the second sampling year considering the quantitative variables used for the construction of the Water Quality Index: Nitrogen-ammonium (N-NH_4), dissolved oxygen (DO), total phosphorus (TP) and periphyton chlorophyll-*a* (Peri Chl-*a*). The sections of watersheds (sites) are represented by different symbols and colors are used to represent each water course in the different sampling dates: November (N), January (J), March (M).

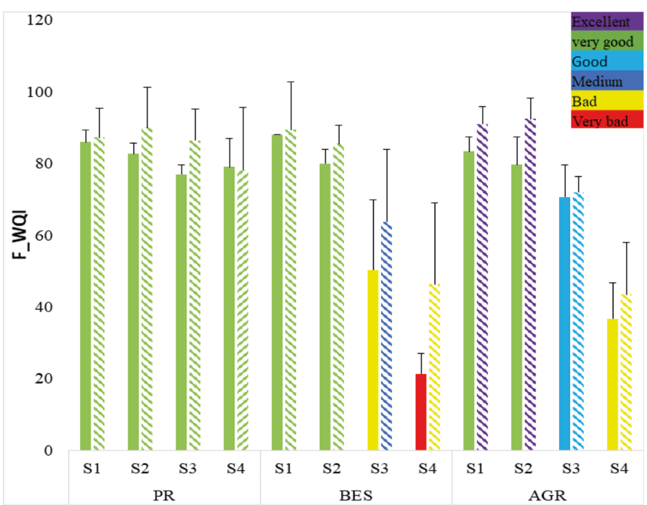

Fig. 8. Average values of the Fuegian Water Quality Index (F_WQI) (n = 3) during the study period at each sampling site (S1, S2, S3, S4) in Pipo river (PR), Buena Esperanza stream (BES) and Arroyo Grande river (AGR). Solid bars represent the average values of the index during the first year, while the hatched bars represent the average values for the second year. Inset: water quality rank with the associated color code.

4. Discussion

The aim of this study was to evaluate the water quality of three urban streams for the Argentinean city of Ushuaia, Tierra del Fuego and to develop a practical water quality index, the F_WQI, which includes physical, chemical, and biological variables. Geographically, Pipo River (PR) is away from the principal urban area of the city and the upper section of the PR belongs to a protected area (National Park Tierra del Fuego). However, due to the population increase that occurred in recent years, urbanization is progressively developing towards such area. In fact, our results showed that the PR watershed has a lower percentage of urbanized area in contrast to Buena Esperanza stream (BES) and Arroyo Grande river (AGR) (Table 1). This implies lower impervious surface at the watershed scale, which helps to prevent deterioration of the water resource. Many studies describe that urban watersheds with large percent of the catchment with impervious surface show decreased perviousness to precipitation, leading thus to decreased infiltration and increased surface runoff with the consequent stream impairment due to larger nutrient loads (Arnold and Gibbons, 1996; Walsh et al., 2005; Booth et al., 2016). Thus, we detected at the urbanized site close to the outlet (S4) of BES watershed higher values of conductivity, turbidity, nutrients (N-NH_4, TP) and fecal coliforms (Appendices B.1, B.2, B.3), being this watershed the smallest in size, with lower water discharge and the highest percentage of urbanization in contrast to PR and AGR (Fig. 1; Fig. 2; Table 1). Dilution is a physical process that is directly related with water discharge, since with lower water discharge, the dilution power of pollutants decreases. This fact could be related to the lower water quality values found at BES. Hence, we must not ignore the relative influence of the hydrology and morphology of the watersheds, since those could explain larger vulnerability to the impacts of urbanization.

A recently published study by Tremblay et al. (2020) assessed the relative influence of the watershed and geomorphic features on nutrients and carbon fluxes in a pristine and moderately urbanized stream. The authors calculated areal fluxes (μg m^{-2} s^{-1}) between two river/stream points considering the measured concentrations of nutrients, water discharges and the streambed. They detected net export (positive areal fluxes) of N-NO_3 and N-NH_4 in the peri-urban reaches whilst in the pristine reaches the flux was negative (retained by the water course). We applied this mass balance equation (Appendix C.1) for one sampling date (March 2019) in order to explore how the areal fluxes for the N-NH_4 concentrations were in the middle zone (S2 to S3) of the studied urban watersheds and we obtained a similar result. Despite the fact that the three watersheds studied in this work are urbanized, the PR watershed, with less urbanized area presented a positive areal flux but lower in magnitude than BES and AGR (0.59, 3.01 and 21.7 μg m^{-2} s^{-1} for PR, BES and AGR, respectively). Even though the mass balance for N-NH_4 was larger for AGR than BES, the average water quality provided by the F_WQI, which includes more variables, resulted in better quality at S3 in AGR than S3 at BES (Fig. 8). According to these results, hydrological (Fig. 2) and morphological differences (Table 1) among watersheds could influence the real impact associated with urbanization in water quality of urban streams at Ushuaia city.

Among biological variables, some water quality indices incorporate coliform bacteria (Cude, 2001; Said et al., 2004; Van Hop et al., 2008) because the inputs of sewage effluents without treatment usually contain high amounts of these microorganisms which are of sanitary relevance. On the other hand, phytoplankton chlorophyll-*a* can also be found in the literature included in the calculation of water quality indices (Tian et al., 2019; Gikas et al., 2020; Ustaoğlu, et al., 2020). In this work, the concentrations of phytoplankton chlorophyll-*a* were in general very low (data not shown) as previously recorded by Diodato (2018) and Rodríguez et al. (2020) in the area. Nevertheless, we detected that Peri Chl-*a* was sensitive to the chemical changes in water quality along the gradient of organic pollution in our studied rivers (Fig. 5). At more urbanized watersheds (BES and AGR) Peri Chl-*a* increased in the middle-low and close to the outlet (Fig. 5b, 5c). This suggest that Peri Chl-*a* would be more sensitive to the extra input of N and P nutrients, showing significant differences between the higher sections (S1 and S2) and the lower sections (S3 and S4) as a result of the influence of point and non-point sources of organic pollution. Higher sensitivity to organic pollution is also evidenced in the PCA results for both years, where Peri Chl-*a* was one of the variables which more contributed to explain the variability in component 1. Similar results were obtained by Busse et al. (2006) in Southern California. These authors reported an increase in algal biomass with urbanization, reaching higher concentrations in the most urbanized streams, obtaining even higher values than in this study. Additionally, there are studies that have used the periphyton to assess the beaver dam effect in streams in the area nearby Ushuaia city (García and Rodríguez, 2018; Rodríguez et al., 2020). The average values of Peri Chl-*a* reported in the mentioned studies, which correspond to streams without urbanization and without beaver activity, are similar to the values obtained in this work for the reference sites (S1) in all the watersheds (0.35 mg m^{-2} in Rodríguez et al., 2020 and 0.39–0.30 mg m^{-2} in García and Rodríguez, 2018).

In relation to the water quality index of forested urban streams in Ushuaia, we detected changes along the watersheds. During the first year of sampling, BES and AGR showed worse water quality in their outlets (S4), reaching average values below optimum (category "good") according to the F_WQI (Fig. 8). These results are consistent with Pesce and Wunderlin (2000), Debels et al. (2005), and Ewaid and Abed (2017) who, through the application of different indices, reported "bad" or "poor" water quality values at those sampling sites downstream to sewage discharges. We associated lower index values with effluent discharges and non-point pollution in the watersheds and due to the absence of sewage treatment infrastructure. Currently, only 65% of the city is connected to the main sewage system; from this fraction, approximately 3/4 of liquid wastes have been redirected to a pre-treatment plant that finally ends in the submarine emissary in Golondrina Bay (Beagle Channel). The other quarter is now being discharged into AGR until the secondary treatment plant will begin operating in 2021 (Diodato et al., 2020). The remaining 35% of the city has no connection to the sewage network and hence, water courses and the coast of Beagle Channel receive raw effluents (Torres et al., 2009). In general, the discharge of effluents from wastewater usually provides high concentrations of ammonium and fecal coliform bacteria into the watercourses, which coincides with our results (Table A.5; Fig. 3b and 3c) and with values reported by Diodato (2013) for the same area.

On average, values of the F_WQI of all sites at Pipo river (PR) watershed were higher than 76, indicating a very good water quality (Fig. 8). Although we expected to find worse water quality in sampling sites downstream the urbanization, water quality at PR was not impaired. Even if, F_WQI values decreased downstream the urbanized area of the PR (S4), the variables included to calculate the F_WQI, showed a narrower fluctuation throughout the watershed than at BES and AGR; and only TP showed significant differences between S3 and S4. According to the information provided by the Dirección Provincial de Obras y Servicios Sanitarios (DPOSS) (pers. com.), PR do not have a mixed sewage system, therefore pluvial and sewage effluents are separated. This improved/highly efficient system could explain the low amount of organic pollutants that enter into the course through natural runoff. Thus, PR would only be receiving occasional contributions of wastewater from the overflow of a near pumping station. The other two watersheds under study (BES and AGR) have a mixed sewage system, where pluvial and domestic effluents are combined. In this case, the total volume of loaded water into the streams is greater and is almost composed by raw effluents.

In relation to the second year, we obtained on average, higher values of the water quality index (except in S4 at PR) than those found in the first year. Besides, changes in the categorization of the F_WQI were detected in the middle-low section (S3) and in the low section (S4) at BES. These results may be linked, on the one hand, to the higher water discharge during the second year of sampling (Fig. 2), which could be associated with more dilution of the contaminants, as we mentioned above. There are contamination studies that show better water quality in periods with higher water flow (Pesce and Wunderlin, 2000; Almeida et al., 2007). In the watersheds under study, higher flows and dilution effects are generally associated with summer time, when defrosting events occurs (Iturraspe and Urciolo, 2000). Although in some sampling sites maximum water discharge values are observed in November 2018, we did not detect temporal trends in water discharge within each sampling year.

On the other hand, infrastructure works and improvements in the sewage system have recently been carried out as a part of an integrated sanitation plan in the Province. The negative result of fecal coliform bacteria during the last sampling date (March 2019) could be related to a reduction in effluent discharges which was also observed by other authors (Albizzi et al. in press; Diodato unpublished results). Particularly, infrastructure improvements were carried out during the year 2018, specifically in the mid-lower section of BES. Then, the improvement in the water quality in these sections of BES (S3 and S4) could be also directly related to the decrease in the input of domestic wastewater into the streams. However, as we only have two years of sampling, more research is needed to infer whether the water quality improvement we observed might be owed to discharge increase during the second year or infrastructure associated effects which would last longer.

5. Conclusion

The urbanization of Ushuaia in Tierra del Fuego negatively affected the water quality of urban forested streams that cross the city, particularly, in sites downstream. We detected very good water quality in all sections of the Pipo river watershed. Buena Esperanza stream and Arroyo Grande river showed water quality impairment: BES in the middle-low site (S3) and lower site (S4) and AGR in the lower site (S4). We encourage to use the F_WQI contemplating the inclusion of periphyton chlorophyll-*a* (Peri Chl-*a*) which proved to add sensitivity to the water quality index in temperate rivers and streams where this community is in general ubiquitous.

CRediT authorship contribution statement

María Granitto: Conceptualization, Methodology, Software, Writing - original draft, Formal analysis, Investigation. **Soledad Diodato:** Visualization, Writing - review & editing, Investigation. **Patricia Rodríguez:** Conceptualization, Visualization, Writing - review & editing, Supervision, Resources, Project administration, Funding acquisition.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ecolind.2021.107614.

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