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TITLE

Massive plastic pollution in a mega-river of a developing country: sediment deposition and ingestion by fish (*Prochilodus lineatus*).

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KEY WORDS

Plastic waste; South America; large river; macroplastic; secondary microplastic; fish digestive tract.

ABSTRACT

The aim of this study was to determine the amount, composition and origin of plastic debris in one of the world largest river, the Paraná River in Argentina (South America), focusing on the impact of urban rivers, relationships among macro, meso and microplastic, socio-political issues and microplastic ingestion by fish.

We recorded a huge concentration of macroplastic debris of domestic origin (up to 5.05 macroplastic items per m²) dominated largely by bags (mainly high- and low-density

polyethylene), foodwrapper (polypropylene and polystyrene), foam plastics (expanded polystyrene) and beverage bottles (polyethylene terephthalate), particularly downstream from the confluence with an urban stream. This suggests inadequate waste collection, processing and final disposal in the region, which is regrettably recurrent in many cities of the Global South and Argentina in particular.

We found an average of 4654 microplastic fragments m^{-2} in shoreline sediments of the river, ranging from 131 to 12687 microplastics m^{-2} . In contrast to other studies from industrialized countries from Europe and North America, secondary microplastics (resulting from comminution of larger particles) were more abundant than primary ones (microbeads to cosmetics or pellets to the industry). This could be explained by differences in consumer habits and industrialization level between societies and economies.

Microplastic particles (mostly fibres) were recorded in the digestive tract of 100% of the studied *Prochilodus lineatus* (commercial species).

Contrary to recently published statements by other researchers, our results suggest neither macroplastic nor mesoplastics would serve as surrogate for microplastic items in pollution surveys, suggesting the need to consider all three size categories.

The massive plastic pollution found in the Paraná River is caused by an inadequate waste management. New actions are required to properly manage waste from its inception to its final disposal.

CAPSULE

Massive plastic pollution in a mega-river from Argentina, mainly caused by inadequate waste management.

1. INTRODUCTION

Plastic pollution is one of the great challenges for environmental management in our times.

Plastic debris is a combination of high persistence, low density, and extremely wide size distribution. This causes the behavior of plastic debris to show a far wider variety than most other materials, such as suspended fine sediments (Kooi et al. 2018). Plastic particles cause severe damage to freshwater and marine ecosystems (Galloway et al. 2017). In the oceans alone, the economic damage due to plastic pollution is estimated as high as 21 billion Euro (Beaumont et al. 2019). In spite of a great scientific effort to tackle this problem worldwide the state of our knowledge is yet deficient for different reasons. Firstly, despite wide

research efforts investigating plastic pollution in oceans, considerable less attention has been paid on freshwater systems (Blettler et al. 2018). Nevertheless, this imbalance seems to be reversing in the last years (e.g. Gündoğdu et al. 2018; Battulga et al. 2019; Mani et al. 2019; van Wijnen et al. 2019; Zhang et al. 2019).

Secondly, research on freshwater plastic pollution have been mainly carried out in industrialized countries (the Global North; Rochman et al. 2015; Blettler et al. 2018). This is not surprising due to the bias in the scientific output between the Global North and the Global South (Guterl 2012). However, this disparity causes concern, as increasing population levels, rapid urbanization, informal settlements, and the rise in consumption levels have greatly accelerated the solid waste generation rate in the Global South, where waste collection, processing and final disposal is still poor (Minghua et al. 2009; United Nations Human Settlements Programme 2016).

Thirdly, there is a clear dominance of microplastic over macroplastic studies in freshwater environments worldwide (less than 20% of the total surveys in freshwater systems have been focused on macroplastics; Blettler et al. 2018). Consequently, more macroplastics studies in freshwaters are urgently required since: i) studies estimating the amount of plastic exported from rivers into the ocean are limited due to the scarcity of field-data in rivers (Lebreton et al. 2017, Schmidt et al. 2017); ii) global studies estimating the amount of plastic exported from rivers into the ocean have evidenced a significantly (>100 times) greater input in terms of weight of macroplastics (compared with microplastics, Schmidt et al. 2017); iii) removing macroplastics in rivers (e.g. using artisanal boom barriers) is an effective/low-cost action to avoid plastics reach the ocean but, on the contrary, the same action on microplastic is virtually impossible. Microplastics can be categorized by their source. Primary microplastics are purposefully made to be that size

(e.g. microbeads used in cosmetics and personal care products, virgin resin pellets used in plastic manufacturing processes), while secondary microplastics are the result of larger items of plastic breaking down into smaller particles (Weinstein et al. 2016). Studies indicated that wastewater treatment plants (WWTPs) play an important role in releasing primary microplastics to the environment (Ou and Zeng 2018; Gündoğdu et al. 2018). Fourthly, the largest rivers in the world (also called mega-rivers) are located in developing countries (see Latrubesse et al. 2008). The great discharges, basin sizes and poor sanitary conditions of people living in these catchments, potentially increase the amount of plastic debris flowing through mega-rivers to the ocean. However, information about plastic pollution in mega-rivers of developing countries is still very scarce (Pazos et al. 2017, Blettler et al. 2018), even though all the plastic input conveyed by rivers is eventually released into oceans (Morritt et al. 2014) or accumulated in estuaries (Vermeiren et al. 2016). Fifthly, the ingestion of microplastics by fish, and the associated risks to human health, remain major knowledge gaps (Santos Silva-Cavalcanti et al. 2017), even though the major inland fisheries are located precisely in the most plastic polluted rivers (Lebreton et al. 2017) of the Global South (FAO 2016). The above suggests an urgent need to focus monitoring efforts in the most polluted rivers, specially where inland fisheries are crucial for local consumption and economies, as it is the case with the Paraná River. Taking into account the rationale outlined above, the objectives of this study were to determine: i) the amount, origin and composition of plastic debris deposited in sediments of a mega-river (Paraná River), ii) the plastic input conveyed by an urban stream joining the Paraná River; iii) quantitative relationship between macro, meso and microplastics in

sediments; iv) microplastic ingestion by *Prochilodus lineatus*, an iliophagous fish (that feeds mud containing detritus and associated organisms).

2. MATERIALS AND METHODS

2.1. Study area

La Plata basin is one the ten largest fluvial basins of the world, draining five countries (southern part of Brazil, the northern of Argentina, Bolivia, Uruguay and Paraguay), accounting for 17% of the surface area of the South America and supporting 19 large cities (with a population greater than 100,000 inhabitants). The Paraná River is the largest river of this basin, ranking ninth among the largest rivers of the world, according to its mean annual discharge to the Atlantic Ocean ($18,000 \text{ m}^3 \text{ s}^{-1}$; Latrubesse 2008). However, this river is also one of the world's top-ten rivers at risk due to anthropogenic pressure (Wong et al. 2007).

The study took place near Paraná city (Argentina), located on its eastern shore of the river, with a population of about 300,000 inhabitants. The collection, processing and final disposal of waste of this city is still deficient resulting in strongly polluted urban streams. We selected three sampling areas in the Paraná River bank sediments: upstream of the city (Escondida beach), in the city (Thompson beach, a municipal public beach), and in an island located in front of the city (Curupí island; Figure 1). Thompson is a recreational beach influenced by the mouth of a strongly polluted urban river ("Las Viejas" stream) that flows through the Paraná city. Fish were caught in the vicinity of the sampling sites. Due to flow conditions, we expected that the upstream site would be the least polluted, followed by Curupí island, whereas Thompson beach, is influenced by the strongly polluted "Las Viejas" stream crossing the city.

>>>>>> Figure 1.

2.2. Sampling.

We selected 2 transects of 50 m in length and 3 m wide for the macroplastic survey (Noik and Tuah 2015) in each sampling area. Transects were selected parallel to the riverbank, randomly chosen, and covering more than a 20% of the shoreline section (Lippiatt et al. 2013). All visible macroplastic items on the surface of each transect were collected by hand.

Plastic debris was sorted according to size and classified as macroplastic (> 2.5 cm), mesoplastic (5 mm to 2.5 cm), or microplastic (≤ 5 mm). This classification is currently used by the UNEP (Cheshire et al. 2009), NOAA (Lippiatt et al. 2013) and MSFD (2013).

We collected mesoplastic debris from triplicate samples (1 m^2) randomly located into each macroplastic-transects (after macroplastic being picked up; Lippiatt et al. 2013).

Mesoplastics particles were carefully removed from the top 3 cm of sediments of each 1 m^2 quadrat (using stainless steels of 5 mm mesh size to sieved the sediments). In a similar way, we took microplastics samples also per triplicate from the macroplastic-transects but using smaller quadrats ($0.25 \times 0.25 \text{ m} \times 3 \text{ cm}$ depth; Klein et al. 2015). Mesoplastic particles were hand-picked in the field using stainless steels (5 mm mesh size), while microplastic samples were directly transferred to the laboratory for processing.

All sampled (macro and mesoplastics and sediment with microplastics) were transferred to the laboratory for further analyses (see below).

Prochilodus lineatus (locally called “Sábalo”) is a dominant detritivorous fish species of great importance for commercial and artisanal fishing (Espínola et al. 2016). For the analysis of fish, we obtained 21 fresh specimens that were caught with gill nets of 14 and 16 cm between opposite knots at the respective sites of the study area, respecting local policies. Fish were caught in the early morning hours and transported to the laboratory on ice within 3 hours. For each individual, total length (cm) was measured and the body total weight (g) was also determined. Afterward, fish samples were cut open using a scalpel and gastrointestinal tracts were removed and immediately placed in clean glassware in order to minimize the risk of laboratory contamination (Bessa et al. 2018). In addition to the methods described below, we also noted the color of the eaten particles in order to identify potential preferences.

In order to avoid contamination from microplastics, potentially present in the laboratory environment, the use of cotton lab coats, gloves and mask was mandatory. Moreover, glassware and working place were cleaned with solution of ethanol (96%) before starting all experiments in order to conserve a sterile environment. From the beginning of the operations until the observation under the microscope, the samples were covered with aluminium foil.

The organic matter presents in the samples was digested with hydrogen peroxide (H_2O_2) (30%) at 60°C (Pazos et al. 2017; Jabeen et al. 2017). According to Sujathan et al. (2017), H_2O_2 is an oxidizing agent that no changes or bleach the structure of microplastic particles. According to our environmental principles, all sampling campaigns were performed using kayaks (zero emission and free noise pollution).

2.3. Samples analysis and processing.

Macroplastic particles were washed, counted and classified in the laboratory (item by item). The classification accounted for their functional origin (e.g. food wrappers, packaging, beverage bottles, shopping bags, personal care products, etc.) following the NOAA (Lippiatt et al. 2013) and resin composition. The ASTM International Resin Identification Coding System (RIC 2016) was used to recognise the plastic resin used in manufactured macroplastics (Gasperi et al. 2014). As the later procedure was not always possible to use (sometime this code is lost or not clearly visible), we used a FT-IR Spectrophotometer Shimadzu IR Prestige 21™ to identify the plastic resin (Song et al. 2015). According to Gündoğdu and Çevik (2017), mesoplastics were counted and classified in: Styrofoam, hard plastic, fishing line, and films.

Microplastic separation was performed following the method proposed by Masura et al. (2015). Thus, full samples were dried at 60°C per 24hs, weighed and sieved through a stainless steel sieve of 350 µm mesh size using a Retsch™ sieve shaker. The remaining material was transferred to a 1L beaker for wet 30% peroxide oxidation (H₂O₂), and located on a hot plate set at 60°C until all organic material digested (Yonkos et al. 2014). After completion, H₂O₂ was washed using distilled water through a 350 µm mesh size.

Afterwards, a concentrated saline NaCl solution (1.2 g cm⁻³) was added and strongly stirred for about one minute (Hidalgo-Ruz et al. 2012). Afterward, the supernatant with floating microplastics was extracted and washed with distilled water for further processing. This last step was repeated as many times as it was needed in order to catch every floating plastic particle.

Microplastics were separated from other materials (present in the supernatant) and classified under a Boeco™ zoom stereo microscope and a Nikon™ binocular microscope (10–40x). We used the criteria suggested by Norén (2007) to identify microplastics.

However, items of doubtful origin were analysed with a FT-IR Spectrophotometer in order to confirm (or reject) their plastic composition (Frias et al. 2014; Li et al. 2016). Spectra ranges were set at 4000–400 cm^{-1} , using the IRsolution Agent software. The resulting spectra were directly compared with the reference library databases.

Microplastics were classified in Styrofoam (trademarked brand of closed-cell extruded polystyrene foam), hard plastic, film, fiber and fiber-roll (very large fibers twisted), according to Castañeda et al. (2014) and Gündoğdu and Çevik (2017).

2.4. Data analyses

Tables and figures were created to identify presence, abundance and type of plastic debris in order to compare the sampling sites between each other. Correlations were performed among the different plastic size ranges. In order to test spatial patterns of similarity in the abundance and type of microplastics, a Canonical Analysis of Principal (CAP) coordinates was performed. The CAP is a constrained ordination analysis that calculates unconstrained principal coordinate axes followed by canonical discriminant analysis on the principal coordinates to maximize the separation between predefined groups (Anderson, 2004). The Bray-Curtis dissimilarity index and 999 permutations were the parameters selected in this procedure. Subsequent one-way Permutational Multivariate Analyses of Variance (PERMANOVA) (Anderson, 2001) was conducted to determine differences between scores of the CAP Axis 1.

Statistical analyses were carried out using the CAP software Version 1.0 (Anderson, 2004) and the MULTIV software, version 2.4.2 [Pillar, 2004], with a statistical significance level was $p < 0.05$.

3. RESULTS

3.1. Macroplastics.

We recorded a total of 18 categories of macroplastic debris (based on the NOAA's classification; Lippiatt et al. 2013); being bag, foodwrapper, Styrofoam and beverage bottle the most abundant particles, representing almost the 80% of the total (Table 1).

>>>>> Table 1.

The three sampling sites have strong differences in amount (number of items) and type of macroplastic debris (Figure 2a). Thus, Escondida beach (4 km upstream Paraná city) showed the lower values (52 macro-items per transect; 150m²), with a heterogeneous composition of plastic types (13 different categories) but dominated by fishing lines (23 items). The Curupí island (in front of the Paraná city), was dominated by only 2 types of macroplastics: beverage bottles (81) and Styrofoam fragments (99). Finally, the Thompson beach (slightly downstream to the Las Viejas outlet) showed a clear dominance of shopping bags (490; many different colors and textures) and food wrappers (202.5), having the highest amount of plastics: 757.5 items per transect (i.e. 5.05 macroplastic particles per m²), 14 times more than the Escondida beach. By far, the most abundant plastic resins were HDPE, LDPE, PP and PS in the Thompson beach, EPS and PET in the Curupí island and Nylon in the Escondida beach. Cellulose acetate, Polyester and PVC resins were found at low densities.

>>>>> Figure 2.

3.2. Mesoplastics.

In contrast to macroplastics, mesoplastics had the highest abundance in the Escondida beach (55.6 items m^{-2}), followed by Curupí island (35.5 items m^{-2}) and Thompson beach (only 18.5 particles per m^2 ; Figure 2b). The average abundance of mesoplastic was close to 46 items m^{-2} , being foam plastic (Styrofoam) the dominant category (41.1 items m^{-2}) (Table 2).

>>>>>Table 2.

3.3. Microplastics.

Films and fibers were the dominant items in the microplastic samples (Table 3). An average of 4654 microplastic fragments (per m^2) was found in shoreline sediments of the three sampling (beaches and island). An average of 12687 micro-particles m^{-2} (81% of the total) were recorded in the Thompson beach, but only 131 in the Curupí island (Figure 2c). Microplastic film and fibber were extremely abundant in the Thompson beach.

>>>>> Table 3.

The CAP (and subsequent PERMANOVA) showed significant differences in abundance and type of microplastics between the three beaches (sampling sites) ($p\text{-values} = <0.003$; Sum of squares (Q) within groups= 2.829) (Figure 3).

>>>>> Figure 3.

Table 4 shows that the density values of the size classes (macro, meso and microplastic) were not surrogate of each other (no correlations were detected). While some weak tendencies could be detected (ex.: high concentration values of macro and microplastics in the Thompson beach), they were not statistically significant. Particularly, the mesoplastic abundance showed a completely independent tendency. For ex.: lowest values of macroplastic were found in the Escondida beach, but mesoplastic showed the highest concentration in the same beach. While the highest concentrations of macro- and microplastics were found in the Thompson beach, the mesoplastic concentration there was the lowest one.

>>>>> Table 4.

3.4. Fish ingestion.

All fish were contaminated with at least one microplastic. The number of items recorded in the digestive tracts of adult *P. lineatus* averaged 9.9 microplastic particles, The maximum value of microplastic particles recorded in an individual was 27 (Figure 4). Particle sizes ranged between 0.5 to 3mm and recorded colours were blue (most of them), black, yellow, red and transparent.

>>>>> Figure 4.

4. DISCUSSION

4.1. Massive plastic concentration: geo-political issues and societies.

Macroplastic materials are the most visible form of plastic pollution. Blettler et al. (2017) reported an average of 172.5 macroplastic items per transect of 150 m² (~1.15 items m²) in a floodplain lake of the Paraná River, located only 18km from our sampling area. In the present study, we found almost twice that amount: 340.8 macroplastics per 150 m² (~2.27 m²).

While several studies on macroplastics have been performed in water surface of rivers (Gasperi et al. 2014; Faure et al. 2015; Baldwin et al. 2016; Lahens et al. 2018) and lakes (Faure et al. 2015), macroplastic studies in riverine sediments are still scarce, especially for beaches. Some examples include Imhof et al. (2013) in the Garda lake (Italy) and Faure et al. (2015) in 6 lakes of Switzerland. However, direct comparison with the present study are unfeasible since these authors considered macroplastics as the particles higher than 5mm (including mesoplastic size).

The great amount of macroplastic debris recorded in the Thompson beach and Curupí island, as well as the origin of them (household waste, Table 1), suggest a deficient waste collection, processing and final disposal in the Paraná city. Waste management is one of the key environmental issues concerning urban hydrosystems on a global scale, however, in the Global South it still remains strongly based on uncontrolled dumping and/or littering (Guerrero et al. 2013). As a result, serious environmental problems (Al-Khatib et al. 2010) and increasing plastic pollution (Battulga et al. 2019) occur, particularly in freshwater systems. Municipalities in low-income countries are spending lower proportion of their budgets on waste management, and yet over 90% of waste in low-income countries is still openly dumped (Kaza et al. 2018). In addition, increasing population levels and the rise in consumption levels have greatly accelerated the solid waste generation rate in Argentina

(waste generation rates: 1.14 kg/capita/day; Kaza et al. 2018). The present study shows, in part, this global trend.

Most of the macroplastics recorded in the present research were shopping bags, followed by food wrappers and foam packaging (almost 80%; Table 1). The first communities to embrace the anti-plastic bag norm were in the Global South, with those in the Global North only doing so much more recently (Clapp and Swanston, 2009). However, an anti-plastic-bag municipal ordinance was not adopted in the Paraná city before 2017.

Results from available microplastics studies in freshwater systems are extremely variable according to the used methodology used (e.g. grab sampler, sediment core, manta net, pump, etc), size range reported (including nanoplastic), reporting unit (e.g. m^2 , m^3 , l, kg), environment (river, lake, reservoir, estuary, sewage, etc), and sampling compartment (water surface or column, bottom or beach sediment, etc). As a result, comparisons between worldwide studies are very difficult. We found an average of 5239 microplastics m^{-2} (size range: 0.35-5mm) in bank sediments of the Paraná River, ranging from only 75 to a maximum of 34443 microplastics m^{-2} (Table 3). Castañeda et al. (2014) found about 13832 m^{-2} polyethylene microbeads, retained by a 0.5 mm sieve, from industrial effluents in the St. Lawrence River sediments (Canada). Klein et al. (2015) have record about 228-3763 microparticles kg^{-1} in shore sediments of the Rhine and Main rivers in Germany (microplastic size: 0.2-5 mm). Moreover, Su et al. (2016) have reported a range of 15-1600 microplastics l^{-1} (>0.3 mm) in the Middle-Lower Yangtze River (China), Wang et al. (2016) recorded 178-544 microplastics l^{-1} (<5 mm) in the Beijiang River sediments, and Peng et al. (2017) found 410-1600 microplastics kg^{-1} (0.05-5 mm) in some rivers of Shanghai, most of them fragments, spheres and fibers.

Blettler et al. (2017), using the same methodology as the present study, have recorded a much lower average of 704 microplastics m^{-2} (size range: 0.35-5mm) in beach sediments of lentic environments of the Paraná River (a floodplain lake located 18 km from the sampling area of the present study). Xiong et al. (2018) reported 50-1292 microplastics m^{-2} (>0.1 mm) in the Qinghai Lake (China); most of them were films, fibers and foams.

In spite of the limitations and weaknesses of the above comparisons (i.e. different size ranges, units, environments), available information suggest a significant microplastic pollution present in sediments of the Paraná River.

The variation of microplastics abundance and type between sampling sites was statistically significant (Figure 4), showing a clear differentiation per sampling beach. Thompsons beach showed the highest concentration of microplastics, while Escondida revealed the most heterogeneous distribution (sampling stations ranged from low to high microplastic concentration).

Microplastic can occur either in a primary (beads) or secondary form (originating from the breakdown of larger plastic items; Cole et al. 2011). The relative importance of primary versus secondary sources of microplastics is still unknown. We found both of them, but the secondary ones were considerably more abundant (Table 3).

Particular attention should be paid to synthetic clothes, which are an important source of fibers via washing (Conkle et al. 2018). In our study, fiber was the only primary microplastic (Cole et al. 2013) recorded. However, it should be noted that some authors consider fiber as secondary (e.g.: Dris et al. 2015). Other primary microplastics such as microbeads, capsules or pellets (used in cosmetics and personal care products, industrial scrubbers used for abrasive blast cleaning and virgin pellets used in plastic manufacturing processes, respectively) were absent. Similar lack of microbeads was observed in the

Yangtze River (Zhang et al. 2015) and the Three Gorges Reservoir (Zhang et al., 2017) in China, the Saigon River in Vietnam (Lahens et al. 2018), and the Paraná River estuary in Argentina (Pazos et al. 2018). Nevertheless, a great presence of microbeads was observed in the Rhine and St. Lawrence Rivers (Mani et al. 2015 and Castañeda et al. 2014, respectively) and in Laurentian Great Lakes (Eriksen et al. 2013). In some countries benefiting from advanced waste treatment facilities (mainly in Europe and North of America), secondary microplastics releases are even lower than primary microplastics (Gouin et al. 2015). Losses of primary microplastics can occur during the production, transport or recycling stages of plastics, or during the use phase of products containing microplastic (e.g. microbeads originated from facial cleansers widely used in developed nations; Napper et al. 2015; Gouin et al. 2015). This contrasts with secondary microplastics that mostly originate from mismanaged waste during the disposal of products containing plastics (Boucher and Friot 2017). The absence of microbeads in the Paraná River system could be explained by these differences in consumer habits and waste management between societies and countries. Herein, almost 50% of the recorded microplastics were film particles (as a secondary product of advanced bag breakdown process), 33.1% fibers (used in textiles) and 18.7% resulting from larger particles of plastic of uncertain origin breaking down into smaller items (probably beverage bottle, foodwrapper and foams) (Table 3). In contrast, other studies in rivers from developing countries have reported a dominance of microplastic fibers (Zhang et al. 2015; Lahens et al. 2018), even in the Paraná River estuary (Pazos et al. 2018).

The variable ratios between macro- or mesoplastics in our study have shown that these data cannot serve as surrogates for microplastics monitoring (Table 4). This is important since

surveys of macroplastics debris can be easily conducted by volunteers, who have played important roles in many debris monitoring programs (Ribic et al. 2012).

4.2. Role of urban streams in plastic dissemination.

Urban rivers and streams suffer from multiple interactive stressors, especially in the Global South (Wang et al. 2012; Wantzen et al. 2019). In this study, Las Viejas urban stream seems to play a crucial role transporting huge amounts of waste plastics and depositing them into the Thompson beach, immediately downstream to the confluence with the Paraná River (Figure 1d). This sampling area showed the highest concentration of macro and microplastic debris (Figure 2 and 4). Las Viejas stream flows all through the Paraná city, concentrating and transporting the municipal solid waste improperly managed. According to Xu et al. (2019) the development of sewer systems has not caught up with the urbanization speed in developing countries, with serious consequences for urban river water quality. Thus, many urban rivers become the end points of plastic pollution (McCormick et al. 2014, 2016). In the same way as rains and severe floods can dramatically increase the plastic levels in the sea (Gündoğdu et al. 2018), it is highly probable that the same phenomenon operates in urban streams discharging to large river systems.

On the other side, the Curupí island showed an average of 190 macroplastics per transect (against 780 in the Thompson and only 52 in the Escondida beach; Table 1). This sampling site was dominated by two domestic items: beverage bottles and foam packaging fragments (Styrofoam; Figure 2). We hypothesize that these plastics arrived from Las Viejas stream. Floating waste is transported by the Paraná River current and dominant southern winds unto the Curupí island shores. This process could be facilitated by the high buoyancy of these items (EPS density: $11\text{--}32\text{ kg m}^{-3}$; while density of PET is 950 kg m^{-3} bottles initially

float due to the air trapped inside). Otherwise, shopping bags and food wrappers (most abundant items in the Thompson beach) were not recorded in the island which is, probably, related to their low buoyance (density of HDPE: 950 kg m^{-3} ; LDPE: $917\text{-}930 \text{ kg m}^{-3}$, PP: 946 kg m^{-3} ; PS: 1066 kg m^{-3}).

Finally, there are no urban river confluences in the Escondida beach, which was the least polluted sampling area. This beach showed a completely different plastic debris composition. While shopping bags, Styrofoam and beverage bottles were present, the dominant item was fishing line. It suggests that the main impact is given by the beach users, most of them artisanal and sports fishermen, and not by municipal waste poorly treated coming from large cities upstream.

The most common plastic polymers recorded in this study were HDPE, LPDE, PP, PS and EPS, which can be very harmful to wild fauna (Kyaw et al. 2012). Moreover, PP and PS have been extensively recorded in food wrappers particles (Table 1). Finally, EPS (often referred as StyrofoamTM) products (takeout containers, dispensable cups, foam trays, etc) were widespread found in our study (Table 1). EPS is commonly reported as one of the top items of debris recovered from shorelines and beaches worldwide (Lee et al., 2013; Ocean Conservancy 2017). As a result, EPS products are now discussed for a ban in several countries (UNEP 2018). In the present study, EPS was the most abundant mesoplastic debris (almost 90%; Table 2). Zbyszewski et al. (2014) and Driedger (2015) reported a similar proportion in mesoplastics from the Great Lakes.

4.3. Ingestion of plastic by fish and potential impacts

Today, the ingestion of plastic has been reported in approximately 150 fish species worldwide (Jabeen et al. 2017), causing internal blockages and injury to the digestive tract

of fish (Cannon et al. 2016; Nadal et al. 2016). We recorded microplastics in the digestive tract of 100% of the sampled *P. lineatus* specimens, corroborating a similar study in the Paraná River estuary (Pazos et al. 2017). The latter could be explained from the detritivorous feeding strategy of this species and the high amount of microplastics recorded in the study area. Thus, the occurrence frequency of microplastics in fish from Paraná River seems to be higher than in other South American rivers. For example, in the Amazon estuary and northern coast of Brazil microplastics were found in 13.8% of digestive tracts examined (Pegado et al., 2018), 23 % and 13.4 % in the Goiana estuary (Possatto et al. 2011 and Ramos et al. 2012, respectively). However, we recognize that the low number of specimens studied here does not allow generalizations.

In our study, most of the recorded microplastics in fish were fibers (90%). In agreement, several studies worldwide have also reported greater number of ingested fibers compared to other microplastic types (Neves et al. 2015; Bellas et al. 2016; Nadal et al. 2016; Pazos et al. 2017). The reasoning behind the dominance of fibers is the diverse nature of this microplastic type, which may originate from the degradation of clothing items, furniture and fishing gear. Indeed, washing (through a washing machine) a single item of synthetic clothing resulted in the release of about 2000 microfibers (Browne et al. 2011; Carney Almroth et al. 2018). Mesoplastics ingested by fish were not recorded in this study. In fact, this range size has been scarcely recorded in fish digestive tracts (Jabeen et al. 2016).

5. CONCLUSIONS

1. The recorded plastic debris concentration (macro, meso and microplastics) was several times higher than the values previously reported in the Paraná River floodplain. Comparisons with other studies worldwide are still difficult, since methodological

protocols are not yet standardized; however, they suggest massive pollution levels in this mega-river of South America.

2. Macroplastics recorded herein have a domestic origin (shopping bags, food wrappers, beverage bottles and packaging foam fragments), suggesting an inadequate waste collection, processing and final disposal in the region, which is regrettably recurrent in the Global South. The further research must not overlook macroplastics in this geopolitical region, particularly if reliable estimates of global plastic waste entering to the ocean from rivers are intended.

3. Secondary microplastics (originated from the breakdown of larger plastic items) were more abundant than primary ones (manufactured as microbeads, capsules, pellets used in industry). Microbeads (commonly found in industrialized regions) were absent in the Paraná River. This finding contrasts with studies performed in freshwater environments of developed countries, suggesting a difference in consumer habits and levels of industrialization between societies and economies from the developed and developing world.

4. Most of the recorded plastic debris proceed from a highly polluted urban stream, which runs through the Paraná city. Urban rivers, particularly in the Global South, are vulnerable to different urban processes and activities that cause pollution and degradation of the water ecosystem.

5. We recorded microplastic particles in the digestive tract of 100% of *P. lineatus* specimens, most of them were fibers. While we recognize the low number of collected fish, this finding evidenced that microplastics have penetrated in the aquatic food webs and ecological niches in the Paraná River, reinforcing the necessity of more studies.

6. Contrary to our expectations, the macroplastic or mesoplastic items would not serve as surrogates for microplastic surveys (and vice versa), suggesting that all plastic debris sizes should be considered in further studies.

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CAPTIONS

Figure 1. Location of the Paraná River (study area, Entre Ríos Province, Argentina) in the Global South (a). Escondida beach (b), Curupí island (c), and Thompson beach (at the confluence of Las Viejas urban stream with the Paraná main channel) (d).

Figure 2. Bubble chart showing macro- (a), meso- (b) and microplastic (c) densities at each sampling area. Where: f-w: foodwrapper, sty: Styrofoam, b-b: beverage bottle, fishing-line, h-p: hard-plastic piece, fib: fibber.

Figure 3. Ordination plot of the Canonical Analysis of Principal coordinates (CAP) showing significant differences in abundance and type of microplastics between the three sampling sites (Escondida beach, Thompson beach, Curupí island).

Figure 4. Microplastic particles (fibers and others) found in the digestive tracts of *P. lineatus*. Number of items (a), fibers and a piece of plastic film (b).

769

770 **Table 1.** Type (origin/use), density per transect (150 m²), standard deviation, abundance
771 (%) and resin composition of macroplastic debris (total and per sampling site). Where,
772 HDPE: high-density polyethylene; LDPE: low-density polyethylene; PP: Polypropylene;
773 PS: Polystyrene; EPS: Expanded polystyrene; PET: Polyethylene terephthalate; Nylon: dry
774 polyamide; PE: Polyethylene; PVC: Polyvinyl chloride.

775

776 **Table 2.** Type, density (m²), standard deviation, and abundance (%) of mesoplastic debris
777 per sampling site.

778

779 **Table 3.** Type, density (m²), standard deviation, and abundance (%) of microplastic debris
780 per sampling site.

781

782 **Table 4.** Correlations among the different plastic seize ranges.

783

784 **Images and figures:**

785

786

"Islands" of macroplastic debris
flowing in urban rivers

Freshwater birds are using garbage
as nesting material

Birds become entangled in freshwater
debris (fishing line)

Abandoned fishing nets are killing
freshwater fish

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		Marine environment	Freshwater
Topic of research	Entanglement (%)	98.3	1.6
	Nest-material (%)	16.6	0
		Freshwater microplastic	Freshwater macroplastic
	Macroplastics (%)	80	20

791