

Microfibers in the gut of invasive armored catfish (*Pterygoplichthys* spp.) (Actinopterygii: Siluriformes: Loricariidae) in an urban lake in the floodplain of the Grijalva River basin, Mexico

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Abstract

The intake of microplastics by freshwater fishes has been scarcely studied, and it is not yet clear whether the amount of particles these vertebrates ingest is associated with their feeding strategies. Hence, this study is focused on the suckermouth armored catfish (*Pterygoplichthys* spp.) under the hypothesis that, due to its detritivorous habits and demersal distribution, it may show evidence of the presence of microplastics in the sediments of a lake located in the Metropolitan Area of Villahermosa. A total of 21 organisms contained plastic microfibers throughout their digestive systems. These were treated with three solutions (hydrogen peroxide, potassium hydroxide, and sodium hypochlorite) to separate the microplastics. A total of 147 plastic particles of two categories were collected: microfibers (92%) and fragments (8%). Considering these results, as well as evidence in the literature, these species may constitute a global indicator of the incorporation into the trophic chain of microfibers deposited in the sediments of freshwater ecosystems.

Keywords

cosmopolitan, demersal, detritivorous, freshwater, invasive, Loricariidae, microfibers, non-native

Introduction

Plastics constitute the largest group of pollutants that enter freshwater ecosystems, mainly as discarded solid wastes and via wastewater treatment plant discharges, runoff, rain, and even air (Dris et al. 2016; Li et al. 2018; Zhang et al. 2019). Microplastics' (MP) particles

(<5 mm), have been scarcely documented for limnetic environments, and according to Lambert and Wagner (2018), less than 4% of publications on MP focus on this type of aquatic systems, and even fewer on lakes (Free et al. 2014; Yuan et al. 2019; Adeogun et al. 2020; Hurt et al. 2020). MP in freshwater environments are present in at least four forms: fibers, fragments, films, and pellets

(Free et al. 2014; Dris et al. 2018; Li et al. 2018; Vidal et al. 2021), and their origin may be determined depending on their form. For example, pellets come from personal care products (Fendall and Sewell 2009), while microfibers result from the decomposition of larger plastic objects like bags, clothing, and at present, personal protection articles that have been increasingly used in response to COVID-19 (Fadare and Okoffo 2020; Wu et al. 2020; Shruti et al. 2021; Yang et al. 2021). Microfibers have thus become the dominant form due to their numerical abundance in the digestive systems of fish and in freshwater ecosystem sediments (Wu et al. 2020; Vidal et al. 2021; Yang et al. 2021).

It has been suggested that the distribution of MP in these aquatic ecosystems is markedly heterogeneous and is mixed with other natural particles (Klein et al. 2018). Their distribution in the water column or in sediments is related to their density with respect to that of the water (Cole et al. 2011; Li et al. 2020). Due to their distribution in the water column and in sediments, MP may be easily ingested as prey, or incidentally by pelagic and demersal fish that employ different feeding strategies (Campbell et al. 2017; Lusher et al. 2017a; Abbasi et al. 2018; Parker et al. 2020). In addition, the association between MP and microbial organisms, and their adsorption of organic matter, favor their deposition and accumulation processes on lentic ecosystem substrates (Galgani et al. 2019; Parker et al. 2020; Yang et al. 2021) which, in turn, could increase their presence in the digestive system of detritivorous demersal fish.

Mostly, MP in the digestive system of freshwater fishes have been reported in bottom-feeding carnivores, since they account for 49% of the species studied (Sanchez et al. 2014; Peters and Bratton 2016; Jabeen et al. 2017; McNeish et al. 2018; dos Santos et al. 2020; Kuśmierk and Popiołek 2020; Vidal et al. 2021). In contrast, only 13% of registered species were detritivores (Silva-Cavalcanti et al. 2016; McNeish et al. 2018; Adeogun et al. 2020; Vidal et al. 2021). Among these detritivore fishes, stood out the demersal Siluriformes. For example, the armored catfish, *Hoplosternum littorale* (Hancock, 1828) had an intake of 83% and 3.6 MP particles per individual (N_{MP}/ind) in the Pajeú River, Brazil (Silva-Cavalcanti et al. 2016), and another two species, *Otocinclus arnoldi* Regan, 1909 and *Hypostomus commersoni* Valenciennes, 1836 with MP ingestion from 80% to 100% and 1.4 to 2.5 N_{MP}/ind in Uruguayan streams of the Plata River basin (Vidal et al. 2021).

In the northern region of Middle America, non-native invasive species of the armored catfish *Pterygoplichthys* spp. have been recorded as demersal detritivores and are numerically dominant in the urban and suburban aquatic ecosystems of the floodplain of the Grijalva River (Sánchez et al. 2019). Due to its cosmopolitan distribution (Orfinger and Gooding 2018), feeding, and morphology, it is expected that this catfish frequently swallows microplastics deposited in limnetic sediment. If tested, the armored catfish can be proposed as a global bioindicator to quantify the uptake into the trophic chain of microfibers accumulated in the sediments of lentic ecosystems or low hydraulic energy areas in rivers (meanders), such

as the low-lying areas or lakes associated with rivers on coastal plains where environmental conditions have negatively affected aquatic fauna (Sánchez et al. 2012; Torres-Martínez et al. 2017). In order to prove the hypothesis, the aim of this study was to quantify the microfibers ingested by armored catfish after having been deposited and fragmented on the substrate of a low-lying area or a riparian lake with environmental disturbance conditions similar to those recorded for other urban ecosystems (Yuan et al. 2019; Adeogun et al. 2020).

Methods

The catfish were caught in La Pólvara Lake, which is located (17°58'56"N, 092°55'30"W and 17°58'45"N, 092°55'31"W) in the Metropolitan Area of Villahermosa (MAV) in the Grijalva River basin (Fig. 1). It has an area of 40 650 m². It is a riparian depression that has been hydrologically isolated from the Grijalva River by urban infrastructure and has no marginal vegetation due to its rehabilitation as an urban park since 1985. For this reason, its volume is maintained by runoff and extreme or extraordinary flood events (Sánchez et al. 2012). The lake is surrounded by governmental and commercial buildings, schools, and houses. It is subject to strong pressure from activities that take place in the area, which have resulted in hypereutrophic conditions due to high concentrations of phosphorus (0.116–0.126 mg · L⁻¹) and chlorophyll *a* (69–101 µg · L⁻¹) and pollution due to high values of fecal coliforms (7900–240 000 MPN/100 mL) and a low number of aquatic species (Sánchez et al. 2012; Torres-Martínez et al. 2017; SEIACC 2021).

On 23 March 2015, 21 specimens of 280 to 370 mm total length (TL) were collected with a set gillnet (50 m long, 2 m depth, 6.5 cm mesh size) and a cast net (1 cm mesh size). The specimens were preserved in a 10% formalin solution for 15 days, after which they were rinsed in running water to eliminate the formaldehyde and kept in 70% alcohol. The identification included 13 females with gonads in stages III and IV of maturation (in accordance with Núñez and Duponchelle 2008), and eight males for which it was not possible to determine the maturation stage since the gonads were affected by the preservation in formaldehyde and alcohol. Regarding the ventral pigmentation pattern, eight specimens presented spots, seven vermiculations, and six mixed pigmentation (spots and vermiculations) (Table 1). In view of this, it was decided to keep the identification of the specimens at the genus level (*Pterygoplichthys* spp.) following the keys of Armbruster and Page (2006) as, due to the history of management and cultivation of these organisms and the limited of genetic studies in the Grijalva River basin (Vargas-Rivas et al. 2023), it has been suggested that the pigmentation patterns and the morphological characteristics are insufficient to identify species (Wu et al. 2011; Nico et al. 2012; Álvarez-Pliego et al. 2015).

We determined the total length (TL) [mm] and total weight (*W*) [g] of each organism. Individual MP particles were extracted from the digestive tract through a

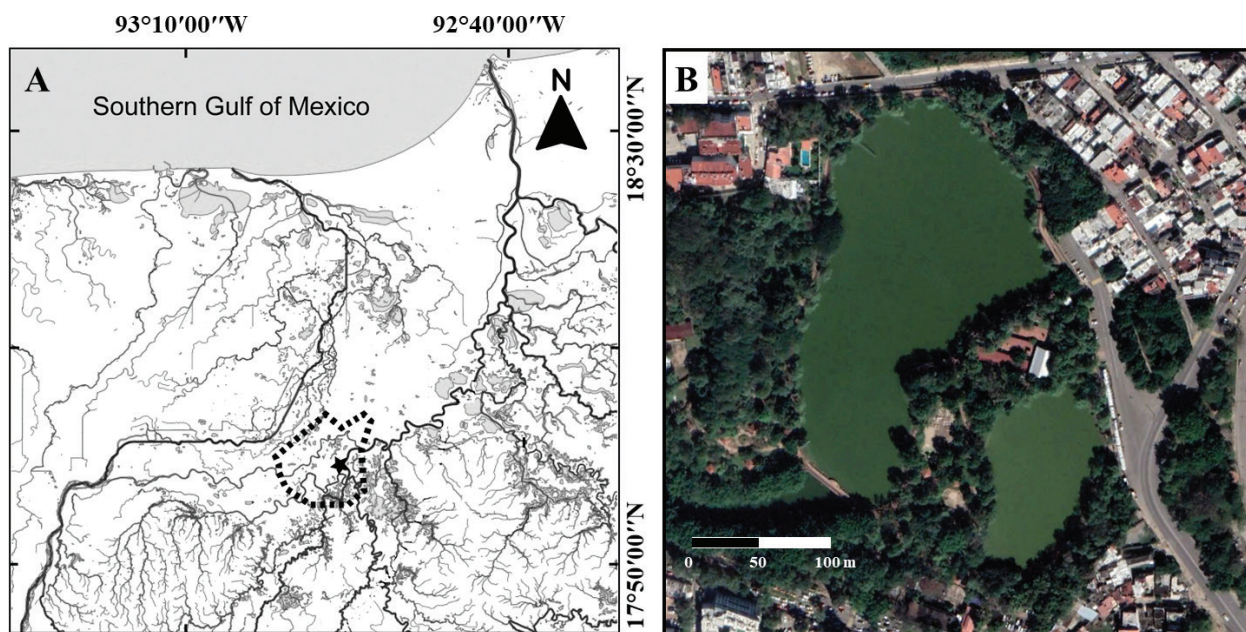


Figure 1. A) Study area; Metropolitan Area of Villahermosa. Map modified from INEGI (2021). B) La Pólvara lake (Satélite Airbus 2023).

dissection in the visceral cavity, by making two cuts in the ventral area of the fish from the anus to the origin of the right pectoral fin and repeating the process towards the left pectoral fin. The weight of the digestive tract (gut) (W_G) was determined [g] and it was placed in glass vessels of 110 mL and submerged in solutions of 30 mL of 35% hydrogen peroxide (H_2O_2) for 48 h and 30 mL of 4% potassium hydroxide (KOH) for 120 h at room temperature (Campbell et al. 2017; Abbasi et al. 2018) in order to dissolve tissues and organic matter without damaging or altering the forms, colors, and sizes of the MP. A solution of 4% sodium hypochlorite ($NaClO$) was later added for an additional week to the samples in which organic matter persisted (Collard et al. 2015). Once the organic matter had dissolved, the sample was washed with distilled water through a 10 μ m mesh sieve to separate and obtain the MP. The remainder was observed with a 10 \times lens on an OLYMPUS BX41 microscope to identify and classify the MP following the categories proposed by Free et al. (2014): 1) granule or particle (irregularly shaped cube with smooth to jagged edges and no smooth plane), 2) fiber (thin thread-shaped structure), 3) fragment (irregularly shaped cube with at least one smooth plane) and 4) film (transparent with two smooth planes).

The number of MP (N_{MP}) was counted per specimen and for all the organisms studied in order to obtain a mean number of MP per individual (N_{MP}/ind) following the protocol of Vidal et al. (2021). The relation between the number of MP and the gut weight (N_{MP}/W_G) was obtained using body measurements data. These results were compared with data recorded in previous publications where the MP in the stomach contents of freshwater detritivorous fish in urban aquatic environments, for which a search was carried out through Scopus and Web of Science, using as keywords: freshwater, fish, microplastic, detritivore, and urban.

Table 1. Individuals of suckermouth catfishes (*Pterygoplichthys* spp.) collected in La Pólvara Lake, Mexico.

TL [mm]	W [g]	W_G [g]	Sex	Pigmentation pattern	N_{MP}		N_{MP}/W_G
					Fiber	Fragment	
280	261	19	M	SV	9	–	0.47
280	264	23	M	SV	2	–	0.09
290	200	19	F	S	4	–	0.21
293	239	12	M	S	5	1	0.50
300	238	9	F	S	6	–	0.67
300	297	21	M	V	15	–	0.71
305	342	25	F	V	2	–	0.08
305	296	12	M	V	6	–	0.50
308	264	20	F	V	11	–	0.55
310	294	6	M	SV	3	–	0.50
310	254	10	F	S	2	–	0.20
310	244	22	M	SV	8	7	0.68
315	370	31	F	SV	2	–	0.06
320	458	31	F	V	4	–	0.13
320	556	43	F	V	6	–	0.14
320	260	18	F	V	10	2	0.67
330	351	27	F	S	6	–	0.22
335	425	40	F	S	12	1	0.33
352	290	20	F	SV	12	1	0.65
355	502	32	M	S	7	–	0.22
370	482	70	F	S	3	–	0.04

TL = total length, W = total fish weight, W_G = gut weight, F = female, M = male, S = spots, V = vermiculations, SV = spots with vermiculations, N_{MP} = number of microplastics per gut weight.

Results

MP were found in the 21 analyzed specimens, varying from two to up to 15 pieces in two animals (Table 1). A total of 147 particles of two categories were counted: microfibers (92%) and fragments (8%) (Fig. 2, Table 1). The mean number of MP per individual (N_{MP}/ind) was 7 ± 4.5 . Also observed in some of the processed samples were remains of plant matter, microcrustaceans (harpacticoid copepods), and detritus (Fig. 2). It was also possible to see that some fibers were tied up among filaments of aquatic vegetation.

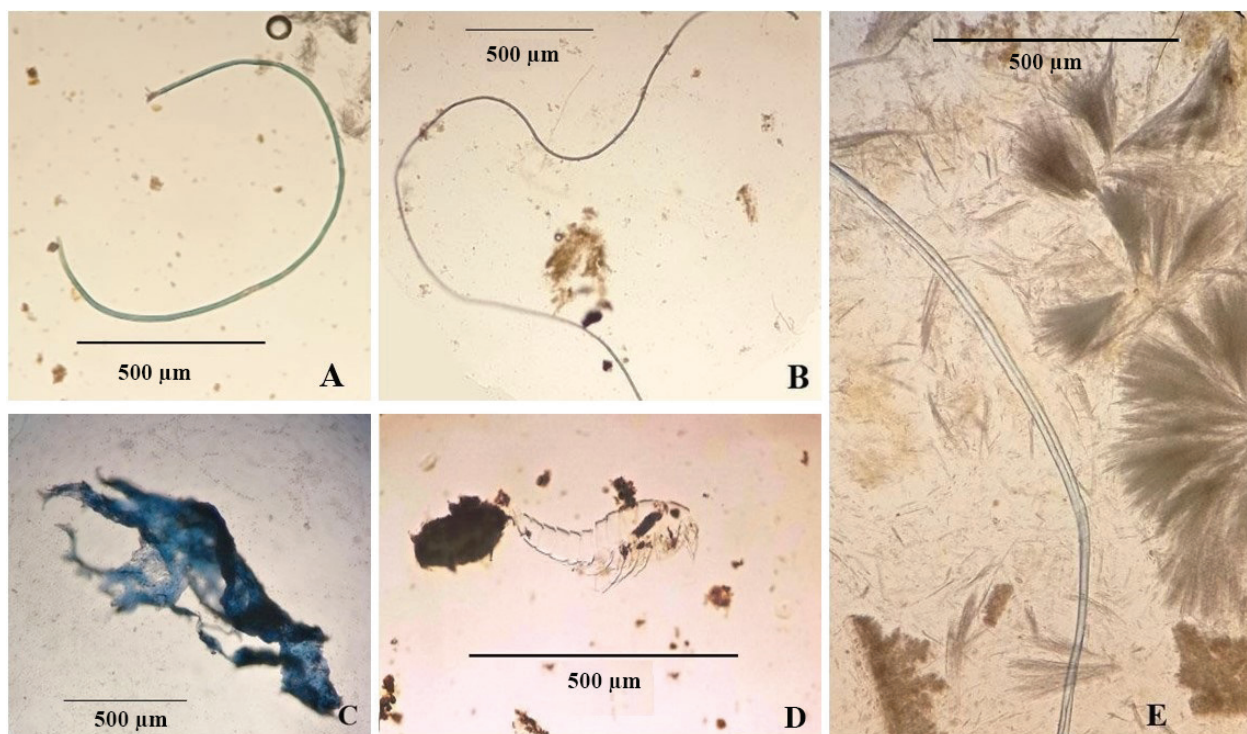


Figure 2. Microplastics found in the digestive tract of armored catfish (*Pterygoplichthys* spp.) and other elements detected. **A** and **B**) Fiber, **C**) Fragment, **D**) Copepod, **E**) Remains of vegetation and organic matter.

Discussion

The high percentage of microfibers (92%) in all plastic fragments identified in the stomach contents of the armored catfish is consistent with the predominance of this category of MP in freshwater demersal detritivore fish (Silva-Cavalcanti et al. 2016; McNeish et al. 2018; Kuśmierk and Popiołek 2020; Li et al. 2020; Vidal et al. 2021; Yang et al. 2021; Blankson et al. 2022), and in turn explains that one of the main sources of microfibers in La Pólvara Lake is the decomposition of larger materials that are transported by different means (Allen et al. 2017; Dris et al. 2018; Li et al. 2018; Zhang et al. 2019). Related to the high environmental pressure caused by human activities, the hydrological isolation of lakes around the world has generated hypereutrophic conditions, low biological diversity, damage to organisms, and the presence of dominant invasive species (Li et al. 2018; Escalera-Vázquez et al. 2020; Jiang et al. 2022), where runoff and rainfall remained as the main contributions of water inputs as a result of the hydraulic isolation and the microtopography (D'Arcy and Carignan 2011). These conditions of hydraulic isolation are more marked during the dry season when runoff and rainfall are scarce and the volume and level of the water decrease, allowing the retention of materials and their settlement on the sediments (Jeppesen et al. 2015; Cruz-Ramírez et al. 2019; Yang et al. 2021). This condition has been frequently reported in urban and suburban lakes located on the coastal

plain of the Grijalva River basin (Sánchez et al. 2012, 2019; Salcedo et al. 2018; Cruz-Ramírez et al. 2019).

In the case of La Pólvara Lake, hypereutrophic conditions (Sánchez et al. 2012), fecal coliforms above the allowed for public recreational use limits regarding services to the public (SEIACC 2021), a decrease in biodiversity that includes the absence of macrocrustaceans and the presence of non-native species like the armored catfish (Sánchez et al. 2012, 2019), have been recorded. Also, histopathological alterations such as testicular degeneration in the threadfin shad *Dorosoma petenense* (Günther, 1867) and melanomacrophage centers in the ovaries of *D. petenense* and *Thorichthys meeki* Brind, 1918 (see Torres-Martínez et al. 2017) have been detected. Although the reason for these types of lesions has not been investigated in La Pólvara Lake, background studies have reported that changes in the liver tissue of Japanese rice fish, *Oryzias latipes* (Temminck et Schlegel, 1846) (see Rochman et al. 2013) are tied to the plastic chemical compounds being bioaccumulated and affecting fish metabolism (Kuśmierk and Popiołek 2020; Parker et al. 2020).

The previously mentioned disturbance conditions are supplemented with the results of the presently reported study which show that the amount of microfibers recorded in the digestive systems of the armored catfish specimens is very similar to that recorded for other species of aquatic environments adjacent to urban centers (McNeish et al. 2018; Vidal et al. 2021). This proves the frequency of contributions of this type of particles that result from the de-

composition of larger plastics (bags, clothing, consumer products) discarded by people that then enter La Pólvara Lake. This, together with the topographical characteristics and the above-stated human impacts, increases the concentration of MP in these aquatic ecosystems (Free et al. 2014; Peters and Bratton 2016). Likewise, some demersal freshwater fish species with similar feeding strategies to those of suckermouth armored catfish have registered high concentrations of MP, dominated by microfibers. For instance, in urban lakes of $7.63 N_{MP}/ind$ (Yuan et al. 2019), in urban rivers of $3.6–6.4 N_{MP}/ind$ (Silva-Cavalcanti et al. 2016; McNeish et al. 2018), and in suburban streams of $0.63–21.4 N_{MP}/ind$ (Vidal et al. 2021) (Table 2).

At present, the geographical comparison of the quantification and description of MP in the digestive systems of fish is still inaccurate since the results have been reported through only three procedures: 1) the mean number of particles and the categories determined in all the individuals reviewed (Silva-Cavalcanti et al. 2016; McNeish et al. 2018; Yuan et al. 2019; Vidal et al. 2021); 2) the range of MP observed (Silva-Cavalcanti et al. 2016; Yuan et al. 2019; Adeogun et al. 2020); and 3) the use of fluorescence techniques to count MP (Adeogun et al. 2020). However, the available information makes it possible to see that the intake of these pollutants by bottom-dwelling detritivorous fish in lentic environments is greater than that taken by fish in river environments where detritivorous fish recorded values $<6.4 N_{MP}/ind$, while mean values were $>7 N_{MP}/ind$ in the first case, including those reported in this study (Table 2).

The presence of MP in freshwater fishes has been documented since 2014, and since then the number of studies related to these freshwater vertebrates has increased. However, more research is needed to better determine whether the type of feeding of each species is related to the amount and type of MP found in their guts. The presently reported study documents the presence of microfibers in an invasive, bottom-feeding detritivorous loricariid fish that may be used as a global fish indicator of MP accumulated in sediments and their possible transfer to the trophic chain, as was observed in this study and in others that studied this type of species (Silva-Cavalcanti et al. 2016; McNeish et al. 2018; Vidal et al. 2021).

The microfibers detected in the stomach contents of the *Pterygoplichthys* spp. specimens appear to have not been affected by the use of reagents (10% formaldehyde and 70% alcohol) in the preservation process, considering the high number of detected particles (147). However, Lusher et al. (2017b) indicated that synthetic polyamides such as nylon may be damaged by concentrations of 10% formaldehyde and suggested that other preservation techniques like drying or freezing be used in studies of MP in fish. In the case of not being able to preserve fish as is suggested, an alternative could be the dissection of the digestive tract and its preservation in 4% diluted formalin and a saturated solution of sodium borate to reduce acidity and maintain a neutral pH.

Table 2. Freshwater detritivorous fishes with microplastics (MP) recorded in the digestive tract (gut).

Reference	Species	Family	Hab.	N	N_{MP}/Ind	N_{MP}	Fibers [%]	Env.
Silva-Cavalcanti et al. 2016	<i>Hoplosternum littorale</i> (Hancock, 1828)	Callichthyidae	Dem	48	3.6	1–24	46.6	UR
McNeish et al. 2018	<i>Catostomus commersonii</i> (Lacepède, 1803)	Catostomidae	Dem	16	6.4	–	97	SR
Jabeen et al. 2017	<i>Carassius auratus</i> (Linnaeus, 1758)	Cyprinidae	BenPel	11	7.63	1–18	46.8	UL
Blankson et al. 2022	<i>Sarotherodon melanotheron</i> Rüppell, 1852	Cichlidae	Dem	19	–	1–34	–	UL
Blankson et al. 2022	<i>Chrysichthys nigrodigitatus</i> (Lacepède, 1803)	Caroteidae	Dem	3	–	1–3	–	UL
Vidal et al. 2021	[Total value for 6 species listed below]						85	
Vidal et al. 2021	<i>Ancistrus taunayi</i> Ribeiro, 1918	Loricariidae	Dem	2	1	–	–	SS
Vidal et al. 2021	<i>Otocinclus arnoldi</i> Regan, 1909	Loricariidae	Dem	7	1.43	–	–	SS
Vidal et al. 2021	<i>Hypostomus commersoni</i> Valenciennes, 1836	Loricariidae	Dem	5	2.4	–	–	SS
Vidal et al. 2021	<i>Rineloricaria</i> sp.	Loricariidae	Dem	14	1.07	–	–	SS
Vidal et al. 2021	<i>Steindachnerina biornata</i> (Braga et Azpelicueta, 1987)	Curimatidae	BenPel	8	0.63	–	–	SS
Vidal et al. 2021	<i>Hisonotus nigricauda</i> (Boulenger, 1891)	Loricariidae	BenPel	3	0.67	–	–	SS
Presently reported study	<i>Pterygoplichthys</i> spp.	Loricariidae	Dem	21	7	1–15	92	UL

Hab. = habitat, Dem = demersal, BenPel = benthopelagic, N = number of fish specimens examined, N_{MP} = number of microplastics, N_{MP}/Ind = number of microplastics per individual studied, Fibers = the percentage of fibers among microplastic fragments found; Env. = environment, UR = urban river, SR = suburban river, UL = urban lagoon, SS = suburban stream.

Conclusion

The proposal to use the armored catfish as an indicator of the dominant microfibers present in urban aquatic environment sediments is very feasible considering the results obtained in this study. Its use may also serve a dual purpose: 1) to detect these pollutants in an urban lake due to its high tolerance and dominance in altered environments, and 2) as an invasive species in several regions of the five continents, its scientific use may constitute a method to

regulate the populations of these non-native fish without affecting autochthonous species.

In addition, the predominance of microfibers found in the stomach contents of armored catfish and other bottom-feeding species in urban aquatic environments suggests that the main source of pollutants is the weathering or fragmentation of plastic products of urban use. Related to this, the activation of sanitary measures (since 2019) in response to COVID-19 produced an exponential consumption of medical articles of personal use including

facemasks, gloves, masks, and sanitizing towels, several of them a main source of plastic microfibers (Fadare and Okoffo 2020; Wu et al. 2020; Shruti et al. 2021), which in turn increased their presence in the solid urban wastes. In this context, a further and acute increase of MP in aquatic organisms and ecosystems may be expected, mainly in urban lakes where the suckermouth armored catfish is a dominant species.

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