Evidence of microplastics in water and commercial fish from a high-altitude mountain lake (Lake Titicaca)

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ABSTRACT

Microplastic pollution is a widespread environmental concern. Like other anthropogenic pollutants, microplastics can reach aquatic ecosystems through rivers and interact with the aquatic biota. For instance, Lake Titicaca (between Bolivia and Peru), one of the great ancient lakes in South America (3,809 m a.s.l.), shows a pollution problem, particularly in the southern shallow basin (Lago Menor) in Bolivia. Nevertheless, our knowledge of the presence of microplastics and their interaction with the biota of Lake Titicaca is limited. Therefore, this study evaluated the presence of microplastics in the stomach content of the four fish species targeted by local fisheries in Lago Menor of Lake Titicaca (Orestias luteus, Orestias agassizii, Trichomycterus dispar, and Odonthestes bonariensis; N = 1,283), and looked for relationships with trophic guilds or fishing areas. Additionally, surface water was analyzed to evaluate the presence of microplastics in the water. The evaluation of microplastics was carried out by visual observations. We observed that the frequency of microplastic ingestion was low in all species (<5%). Conversely, microplastic was present in the water, with the highest quantity at the southern part of Lago Menor (103 ± 20 particles per L), without differences in the microplastic number between sites. Most microplastics counted in stomach contents were fibers, whereas water samples mainly contained fragments. Our results point to microplastic pollution in Lago Menor of Lake Titicaca. However, we could not determine the pollution rate due to considerable methodological limitations. Further research will be needed to robustly detect microplastics in Lake Titicaca and their impact on the fish species in the lake.

INTRODUCTION

Plastic is an important component in everyday life (Andrady & Neal, 2009), and its diverse applications, widespread use, particularly for disposable items, has increased drastically (Thompson et al., 2009; Li, Liu & Paul Chen, 2018). Plastic is nowadays considered one of the main pollutants in the environment and has attracted worldwide attention (Thompson et al., 2009; Cole et al., 2011; Eerkes-Medrano, Thompson & Aldridge, 2015). Plastic wastes are
observed even in isolated islands in the Pacific Ocean (Lavers & Bond, 2017). Microscopic plastic (i.e., microplastics) is currently a topic of interest to researchers. The plastic characteristics, fragmentation, environmental fate, and potential impacts are the main research topics (Blettler et al., 2018). The term microplastic was coined by Thompson et al. (2004) and further defined by other authors (see Arthur, Baker & Bamford, 2009; Cole et al., 2011; Frias & Nash, 2019; Hartmann et al., 2019). We define microplastic as “plastic particles ranging between 0.1 and 5,000 \( \mu m \)” (Desforges et al., 2014; EFSA Panel on Contaminants in the Food Chain, CONTAM). Microplastic pollution is well-studied in the marine environment, while research on plastic pollution in freshwater environments is still limited (Mani et al., 2016; Blettler et al., 2017; Blettler et al., 2018; Dusaucy et al., 2021). The majority of plastic pollution studies in freshwaters were carried out in Europe and North America, leaving research to do in Asia and Latin America, where progress has been made in recent years (Blettler et al., 2018; Alfonso, Arias & Piccolo, 2020; Galafassi et al., 2021).

The interaction of microplastics and aquatic biota is expected (Silva-Cavalcanti et al., 2017; Alfonso, Arias & Piccolo, 2020). Although microplastic ingestion does not directly impose fatal effects on fish, a negative correlation between fitness and plastic ingestion was observed (Lusher et al., 2014). The pathways of microplastics entering the fish include direct and indirect ingestion. Fishes are the group with most records of plastic ingestion and it is described that fish mainly ingests microplastics via predation activities (Li, Liu & Paul Chen, 2018; Azevedo-Santos et al., 2021). The ingestion of plastics by fish thus may be related to the trophic guild. For instance, omnivorous fish tend to consume more plastic than fish with a specialized diet (Mizraji et al., 2017). Understanding the feeding aspects across fish species facilitates the interpretation of plastic content in the stomach (Dantas, Barletta & Costa, 2015). Freshwater biota is vulnerable to microplastic pollution, particularly fish living in freshwater ecosystems near urban areas (Silva-Cavalcanti et al., 2017; Souza, Corrêa & Smith, 2020). Their vulnerability is caused by the disposal of plastics in freshwater ecosystems coming from direct dumping into rivers or through the disposal of synthetic fiber products from an increasing population and rapid urbanization (Blettler et al., 2018). For example, Lake Titicaca (shared between Bolivia and Peru), one of the highest and most emblematic ancient great lakes in South America (3,809 m a.s.l.), shows a pollution problem that has increased since the 1990s due to poor land-use planning (Molina et al., 2017). The lake is part of the TDPS endorheic system (Titicaca-Desaguadero-Poopo-Coipasa Salt lake), characterized by a north-south gradient, with a single outlet, which drains out to the Lake Uru-Uru and Poopo (Dejoux & Iltis, 1992; Cross et al., 2000; Revollo, 2001). Its physicochemical characteristics are atypical due to its high altitude and tropical location (\( \sim 16^\circ \)S) (Lazzaro & Gamarra, 2014). Lake Titicaca is also the most important water resource for the Andean Altiplano. However, Bolivia’s main source of anthropogenic pollution reaches the lake in Cohana Bay through the Katari River, which drains the densely populated area of El Alto city, with more than 1.2 million inhabitants, industries, and a poor waste water treatment (Molina et al., 2017). Moreover, the pollution on the Bolivian side of Lake Titicaca is exacerbated in the southern shallow basin (Lago Menor), with large areas encompassing depths between 5 to 10 m (Dejoux & Iltis, 1992; Lazzaro & Gamarra, 2014).
Lake Titicaca also hosts a highly endemic biota, dominated by members of the killifish genus *Orestias* (more than 20 species) and the catfish genus *Trichomycterus* (2 species) (*Ibañez et al., 2014*). In addition to the exotic piscivores, rainbow trout (*Oncorhynchus mykiss*), and silverside (*Odontesthes bonariensis*) were introduced in the 1940s (*Loubens, 1992; Loubens & Osorio, 1992*). Although Lake Titicaca provides fish for ~3 million people in the region the fish stock has been drastically declining, mainly by anthropogenic causes (*Ibañez et al., 2014; Monroy et al., 2014; ALT, 2020*). A lack of knowledge remains on fish response to pollution. The native and exotic fishes are the base of artisanal fisheries in Lake Titicaca. Most of the native species (*i.e.*, *O. luteus*, *O. agassizii*, and *T. dispar*) predominantly occupy benthic habitats (*Lauzanne, 1992; Ibañez et al., 2014; De La Barra et al., 2020*) with an omnivorous diet, although with few detailed studies. Conversely, *O. bonariensis* is a piscivore fish, and is the main predator of native *Orestias* (*Loubens & Osorio, 1992; Monroy et al., 2014*). Frequently, the species that are economically important for commercial fisheries are the target of studies on plastic ingestion. However, scarce information exists on microplastic contamination in the fish community of Lake Titicaca. This study, therefore, aims to evaluate the presence of microplastics in the stomach contents of four fishery resources from the Lago Menor of Lake Titicaca caught by local fishermen and to assess whether there is an association with trophic guilds or fishing areas. Additionally, surface water was analyzed to evaluate the presence of microplastics in the water. Finally, our study contributes to the growing body of research on microplastics in freshwater environments.

**MATERIALS & METHODS**

**Study area**

Lake Titicaca is divided into two basins: the oligotrophic deepest northern basin Lago Mayor (7,131 km$^2$; mean depth = 100 m; max depth = 285 m; water volume = 900 km$^3$), and the smaller southern shallower basin Lago Menor (1,428 km$^2$; mean depth = 9 m; max depth = 40 m; water volume = 12 km$^3$) (*Dejoux & Iltis, 1992*). The difference between basins in Lake Titicaca make Lago Menor more susceptible to pollution (*Molina et al., 2017; Guédron et al., 2017*). Similarly, due to its shallow water column, the entire water column of Lago Menor is mixed because of the strong winds (*Achá et al., 2018*). Therefore, small shallow portions of the lake are now eutrophic (*Molina et al., 2017; Achá et al., 2018*). Lake Titicaca hosts a strong artisanal fishing activity of considerable importance for the income of rural families. The fisheries in Lago Menor have four main target species (*O. agassizii, O. luteus, T. dispar* and *O. bonariensis*), which are caught all year round (*Lino & Padilla, 2014; ALT, 2020*). Nonetheless, as stated previously, heavily polluted water drains to the Lago Menor through the Katari River from untreated sewage (*Molina et al., 2017*). This study was carried out in Lago Menor.

**Fish sampling**

We identified the main landing zones arriving from the same fishing areas, and the rural communities of Huarina, Cachilaya, and Desaguadero were selected (*Fig. 1*). We followed the fishing activity during the last weeks of May and June 2021 and bought samples...
composed of 60 to 100 specimens (when possible) of each of the four target species from the fishermen. Often the fish arrived alive at the landing zone, so the entire sample was euthanized in ice, which also reduced digestive action and preserved stomach content. Then, fish were transported in coolers to the laboratory (between one and two hours travel time). Each fish specimen was measured: total length (TL, mm; digital Vernier CD-20CP Mitutoyo) and weight (W, g; PT 120 Sartorius Laboratory precision balance, 0.01 g). After the measurements were taken, the specimens were dissected, and the stomachs were separated. Next, stomach content was washed with distilled water and diluted in case of abundant stomach content. All stomach content was transferred to Petri dishes, where the microplastics were evaluated under a stereomicroscope with a magnification power between 6 X to 50 X (WILD M3, Heerbrugg). Microplastics were identified, counted, and categorized by color and type (fragment and fiber) (McCormick et al., 2016). The visual identification of microplastics may have several limitation, but is still the simplest and most widely used (Li, Liu & Paul Chen, 2018). Our visual evaluation followed personal observations comparing plastic fragments and fibers found in the stomachs with plastic materials found in water samples. In order to prevent any contamination, we used lab coats, gloves, and caps during the evaluation. In addition, all work surfaces and tools were sterilized with ethanol, and all utensils and Petri dishes were carefully assessed before the stomach contents were transferred to ensure the Petri dishes were not contaminated with any plastic. The quantification of microplastics ingested was based on the frequency of occurrence (FO) (Hyslop, 1980), calculated from the equation: \( \text{FO} (%) = \frac{F_i}{F_t} \times 100 \); where \( F_i \) is the number of stomachs containing microplastics and \( F_t \) is the total number of stomachs examined.

**Water sampling**

The water samples were collected during the survey of the project “Working for a healthier aquaculture in Lake Titicaca” (TEAM Project, VLIR-UOS) in Huatajata, Central Islands, and Desaguadero (July, 2021), and Nacoca and San Jose (August 2021; Fig. 1). Samples were collected along transects from the shoreline to open waters of the Lago Menor. Surface water (between 0.5 to 1 m depth) was collected using a 1 L Van Dorn bottle. Three replicates were taken along of each transect, where 5 L of water was collected in each replicate. The collected water was accumulated in carefully washed gallon jugs prior to use. Then, the 5 L of water was filtered in a 20 µm mesh, concentrated in 50 mL falcon tubes, and stored on ice until it arrived at the laboratory for evaluation under controlled conditions. In the laboratory, the individual 50 mL of water sampled was subsequently suspended with distilled water and filtered once again through a 10 µm sieve to avoid losing any portion of the sample. Next, the sieved material was transferred to a Sedgwick-Rafter counting cell with a glass dropper pipette and visually examined under a microscope (Olympus CX43, 4X, and 10X objectives) coupled to a 3.1-megapixel digital color camera for microscopy (Olympus LC30). The microplastics were identified, counted, and categorized by color and type (fragment and fiber) (McCormick et al., 2016). Finally, 10–15 items of microplastics in fragments and fibers were randomly selected, photographed, and measured using the microscope’s camera software (LCmicro, Olympus Image Analysis Software 2.2 version).
The Bolivian Ministry of Environment fully approved the development of the project investigations from which this study is derived (MMAYA/VMABCCGDF/DGBAP/MEG 340/2019).

**Data analysis**

The data presented did not meet the assumptions required for conducting a one-way ANOVA. Therefore, a Kruskal–Wallis test was performed to look for differences in number
of microplastics ingested per fish species, trophic guilds and fishing areas. The Kruskal-Wallis test was also used to test differences in the number of microplastics per site. All statistical analysis were performed using Infostat software (Di Rienzo et al., 2017).

RESULTS

A total of 1,283 fish were evaluated, dominated by *O. agassizii* (32%) (Supplemental Material). We observed microplastics inside the stomach of 44 specimens (3% of the evaluated fishes). Microplastics ingestion was observed in all fishing areas, but none was observed in the stomach of *O. agassizii* and *O. bonariensis* at Desaguadero. The ingested microplastics observed in fish were dark-colored fibers (blue and black). The fibers were usually found as filaments (Fig. 2A) and rarely as a bundle (Fig. 2B). The frequency of occurrence of fibers was low in all species (FO <5%; Fig. 3). The carnivorous species (*O. bonariensis*) showed microplastics at much lower frequency (0.3%). Finally, the mean number of microplastics found per species, trophic guilds and fishing areas were not significantly different ($H = 2.64, p = 0.504$; $H = 1.26, p = 0.297$; and $H = 4.35, p = 0.111$, respectively).

A total of 3,395 particles of microplastics were counted in water samples ranging between 56 and 602 particles per sample. Microplastic quantity was highest at Desaguadero (103 ± 20 particles per L; Table 1), although there were no differences between sites ($H = 7.36, p = 0.118$). Of the total number of microplastics identified, 69.4% were blue, 29.4% red, and 1.2% black. The majority of microplastics counted were fragments (54.2%). Desaguadero was the site with the highest quantity of microplastic fragments (75 ± 19 particles per L). Conversely, a higher quantity of microplastic fibers was found in the Central Islands (41 ± 34 particles per L; Fig. 4). The size of particles ranged from 5.63 µm to 2,756.92 µm for fibers and from 2.15 µm to 1,249.34 µm for fragments. The mean size in length of
Figure 3  Frequency of occurrence (FO) percentage of microplastics found in gut content of the fish studied in Lago Menor of Lake Titicaca. Frequency of microplastics fibers found in the stomach content of four target species of Lago Menor artisanal fisheries. Bars indicate the SD.

Table 1  Microplastic debris in the water of Lago Menor of Lake Titicaca. Mean microplastic number per liter debris in the water samples of Lago Menor of Lake Titicaca.

| Site             | Mean of microplastics counted ± SD | Range of microplastics counted (min-max) |
|------------------|-----------------------------------|----------------------------------------|
| Huatajata        | 36.00 ± 0.57                      | 35.60–36.40                            |
| Central Islands  | 58.87 ± 41.41                     | 11.20–86.00                            |
| Nacoca           | 23.93 ± 5.82                      | 18.40–30.00                            |
| San Jose         | 24.60 ± 16.69                     | 12.80–36.40                            |
| Desaguadero      | 103.13 ± 19.88                    | 81.40–120.40                           |

Notes.
Number of microplastics found = 3,395 particles (Fibers: 45.8%; Fragments: 54.2%).

fragments and fibers was larger in Central Islands (fragments: 117.5 ± 183.7 µm; fibers: 645.3 ± 584.7 µm; Fig. 5).

DISCUSSION
We observed that 3% of the fish evaluated in our study had ingested microplastics. This low occurrence of plastic particles was much lower than other freshwater fish species reported (Galafassi et al., 2021). Furthermore, the relationship between microplastic ingestion, species, trophic guild, and fishing areas was not evident. However, it is notable that the carnivorous species (O. bonariensis) had the lowest occurrence of microplastics. Conversely, omnivorous species, particularly O. agassizii, had a relatively higher ingestion of microplastics. An important ecological characteristic of this species is its feeding plasticity,
Figure 4  Number of microplastics in water samples in Lago Menor of Lake Titicaca. Box plot for the microplastic number found in the water samples in different sites in Lago Menor of Lake Titicaca. Visual examination was performed using an Olympus CX43 microscope, 4X and 10X objectives.

Figure 5  Size of microplastics found in the water samples in Lago Menor of the Lake Titicaca. Bar plot of the size in length of microplastics found in the water samples in different sites in Lago Menor of Lake Titicaca. Bars indicate the SD. Visual examination was performed using an Olympus CX43 microscope, 4X, and 10X objectives, and photographed using an attached Olympus LC30 digital camera. No contrast, gain, and image saturation modification to the photographs were made.

which allows it to take advantage of almost all food items in its habitat (Vila, Pardo & Scott, 2007; Ibañez et al., 2014; De La Barra et al., 2020). Specifically, our result may be consistent with the hypothesis that the amount of plastics ingested by omnivorous fish is
associated with their wider food web (Mizraji et al., 2017). It has frequently been suggested that fish living in freshwater environments near urbanized regions have a higher risk of microplastic ingestion, and a direct relationship with the level of urbanization has been observed (Silva-Cavalcanti et al., 2017; Souza, Corrêa & Smith, 2020; Galafassi et al., 2021). However, several studies present evidence that higher environmental concentrations of microplastics do not necessarily translate into higher ingestion by aquatic biota (McNeish et al., 2018; Talley, Venuti & Whelan, 2020; Sembiring et al., 2020). Such is the case for our results because the microplastic numbers in the water were higher than in the stomachs. Life history may thus have an even more significant influence on the level of microplastic ingestion (Talley, Venuti & Whelan, 2020). It is important to highlight here that more research is needed to obtain further information on microplastic ingestion by fish in Lake Titicaca. Surely, microplastics can cause health complications to wildlife in aquatic ecosystems. These complications can include intestinal obstruction, gut microbiota, physical injury, and liver stress, among others (Browne et al., 2008; Wright, Thompson & Galloway, 2013; Guerrero et al., 2021; Huang et al., 2022). Furthermore, aspects such as transit time and the ability of the digestive system to get rid of microplastics should be considered in future studies to understand the impact of these pollutants in fish of Lake Titicaca.

The mean numbers of microplastic abundance in freshwater systems vary significantly (Li, Liu & Paul Chen, 2018). Human activities, sampling location and sampling approaches could contribute to differences in microplastic number (Eerkes-Medrano, Thompson & Aldridge, 2015). In our study, the number of microplastics was variable (ranging from 11 to 120 particles per liter), showing higher quantities in the southern part of Lago Menor. Specifically, the highest quantity of microplastics was observed in Desaguadero. It was not possible to differentiate whether this higher number of microplastics is a local phenomenon or is a result of the natural watershed gradient. There is no accurate information on the amount of waste discharged into Lake Titicaca. Moreover, there is no data on the Lago Menor’s water flows (Lazzaro & Gamarra, 2014). The number of microplastics was also higher near the Central Islands, possibly because of the spread of pollution from wastewater inlets into the lake (Duwig et al., 2014; Archundia et al., 2017). The Katari River, the main tributary of Lago Menor, is undoubtedly the main wastewater source in this shallow lake (Molina et al., 2017; Achá et al., 2018). However, physicochemical changes observed in Lago Menor suggest other sources of pollutants, in addition to those transported by the Katari River (Achá et al., 2018). In recent years, demographic growth in the Central Islands in Lago Menor has led to rapid urbanization, with no sewage water or solid waste management, adding to the pollutants discharged into Lake Titicaca. There is a strong link between the rapid growth of urban and industrial areas and the detriment of the water quality of Lake Titicaca, as there is deficient wastewater management (Molina et al., 2017; Archundia et al., 2017; Baltodano et al., 2022). This situation thus highlights the need for a monitoring system for Lake Titicaca, one of the 20 largest lakes in the world.

It is worth mentioning that the methods used to evaluate water and the stomach content of the collected fish were based on visual observations and could therefore be biased. Furthermore, none of the samples studied were subjected to a purification
process nor density separation. Therefore, our results should be taken cautiously as no analytical methods was performed. Although the current techniques for quantitative and qualitative research of microplastics include spectroscopy and liquid chromatography (Li, Liu & Paul Chen, 2018), the availability of this specialized equipment is not always easily accessible to researchers in developing countries. Unfortunately, our study has methodological limitations and does not meet all the requirements and quality assurance quality control proposed by Cowger et al. (2020). Specifically, visual observation of plastics can be misleading, and, more importantly, the calculated number of microplastic presence could be an underestimate or overestimate of the real number. In addition, the results of visual classification are strongly affected by several factors (i.e., carelessness of the assessor or the quality of the microscopy) (Hidalgo-Ruz et al., 2012). To date, there is no information on microplastic pollution in Bolivia or Lake Titicaca (Castañeta et al., 2020; Galafassi et al., 2021). Therefore, despite its methodological limitations, our study provides evidence of the pollution of microplastics in Lake Titicaca’s habitat and fishery resources.

**CONCLUSIONS**

In this study, we report the presence of microplastics in the stomachs of the four target species of artisanal fisheries in the Lago Menor ok Lake Titicaca. Our results suggest that the frequency of microplastic ingestion by fish is low, and was not related to the trophic guild or fishing areas. However, the presence of microplastics in the water was particularly high in Desaguadero, near the outlet of Lago Menor. The difference between microplastics found in stomach contents and the water column suggests that microplastic pollution and urbanization were not related in Lago Menor. However, we cannot ensure the pollution rate in Lago Menor due to considerable methodological limitations. Nevertheless, despite these limitations, this study represents relevant information assessing the microplastic content in the emblematic Lake Titicaca. Further research would be needed for more robust detection of microplastic quantity for Lake Titicaca fishes across large spatial scales.

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**ADDITIONAL INFORMATION AND DECLARATIONS**

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**Competing Interests**
The authors declare there are no competing interests.

**Author Contributions**
- Erick Loayza conceived and designed the experiments, performed the experiments, analyzed the data, prepared figures and/or tables, authored or reviewed drafts of the article, and approved the final draft.
- Amaya C. Trigoso Barrientos analyzed the data, prepared figures and/or tables, authored or reviewed drafts of the article, and approved the final draft.
- Geert P.J. Janssens conceived and designed the experiments, prepared figures and/or tables, authored or reviewed drafts of the article, and approved the final draft.

**Animal Ethics**
The following information was supplied relating to ethical approvals (i.e., approving body and any reference numbers):

The Bolivian Ministry of Environment fully approved the development of the project investigations from which this study is derived (MMAYA/VMABCCGDF/DGBAP/MEG/340/2019).

**Field Study Permissions**
The following information was supplied relating to field study approvals (i.e., approving body and any reference numbers):

The Bolivian Ministry of Environment fully approved the development of the project investigations from which this study is derived (MMAYA/VMABCCGDF/DGBAP/MEG/340/2019).

**Data Availability**
The following information was supplied regarding data availability:

The raw data are available in the Supplementary Files.

**Supplemental Information**
Supplemental information for this article can be found online at http://dx.doi.org/10.7717/peerj.14112#supplemental-information.
REFERENCES

Achá D, Guédron S, Amouroux D, Point D, Lazzaro X, Fernandez PE, Sarret G. 2018. Algal bloom exacerbates hydrogen sulfide and methylmercury contamination in the emblematic high-altitude Lake Titicaca. *Geosciences* 8:438 DOI 10.3390/geosciences8120438.

Alfonso MB, Arias AH, Piccolo MC. 2020. Microplastics integrating the zooplanktonic fraction in a saline lake of Argentina: influence of water management. *Environmental Monitoring and Assessment* 192:117 DOI 10.1007/s10661-020-8080-1.

ALT. 2020. Diagnóstico Binacional Pesquero y Acuícola en el ámbito del Sitema Hídrico Lago Titicaca. Rio Desaguadero: Lago Poopó y Salar Coipasa-TDPS.

Andrady AL, Neal MA. 2009. Applications and societal benefits of plastics. *Philosophical Transactions of the Royal Society B: Biological Sciences* 364:1977–1984 DOI 10.1098/rstb.2008.0304.

Archundia D, Duwig C, Spadini L, Uzu G, Guédron S, Morel MC, Cortez R, Ramos ORamos, Chincheros J, Martins JMF. 2017. How uncontrolled urban expansion increases the contamination of the Titicaca Lake Basin (El Alto, La Paz, Bolivia). *Water, Air, & Soil Pollution* 228:44 DOI 10.1007/s11270-016-3217-0.

Arthur C, Baker J, Bamford H. 2009. Proceedings of the international research workshop on the occurrence, effects, and fate of microplastic marine debris. NOAA marine debris program.

Azevedo-Santos VM, Brito MFG, Manoel PS, Perroca JF, Rodrigues-Filho JL, Paschoal LRP, Goncalves GRL, Wolf MR, Blettler MCM, Andrade MC, Nobile AB, Lima FP, Ruocco AMC, Silva CV, Perbiche-Neves G, Portinho JL, Giarrizzo T, Arcifa MS, Pelicice FM. 2021. Plastic pollution: a focus on freshwater biodiversity. *Ambio* 50:1313–1324 DOI 10.1007/s13280-020-01496-5.

Baltodano A, Agramont A, Reusen I, van Griensven A. 2022. Land cover change and water quality: how remote sensing can help understand driver–impact relations in the Lake Titicaca Basin. *Water* 14:1021 DOI 10.3390/w14071021.

Blettler MCM, Abrial E, Khan FR, Sivri N, Espinola LA. 2018. Freshwater plastic pollution: recognizing research biases and identifying knowledge gaps. *Water Research* 143:416–424 DOI 10.1016/j.watres.2018.06.015.

Blettler MCM, Ulla MA, Rabuffetti AP, Garello N. 2017. Plastic pollution in freshwater ecosystems: macro-, meso-, and microplastic debris in a floodplain lake. *Environmental Monitoring and Assessment* 189:581 DOI 10.1007/s10661-017-6305-8.

Browne MA, Dissanayake A, Galloway TS, Lowe DM, Thompson RC. 2008. Ingested microscopic plastic translocates to the circulatory system of the Mussel, Mytilus edulis (L). *Environmental Science & Technology* 42:5026–5031 DOI 10.1021/es800249a.

Castañeta G, Gutiérrez AF, Nacaratte F, Manzano Carlos A. 2020. Microplastics: a contaminant that grows in all environmental areas, its characteristics and possible risks to public health from exposure. *Revista Boliviana de Quimica* 37:160–175 DOI 10.34098/2078-3949.37.3.4.
Cole M, Lindeque P, Halsband C, Galloway TS. 2011. Microplastics as contaminants in the marine environment: a review. Marine Pollution Bulletin 62:2588–2597 DOI 10.1016/j.marpolbul.2011.09.025.

Cowger W, Booth AM, Hamilton BM, Thaysen C, Primpke S, Munno K, Lusher AL, Dehaut A, Vaz VP, Liboiron M, Devriese LI, Hermabessiere L, Rochman C, Athey SN, Lynch JM, De Frond H, Gray A, Jones OAH, Brander S, Steele C, Moore S, Sanchez A, Nel H. 2020. Reporting guidelines to increase the reproducibility and comparability of research on microplastics. Applied Spectroscopy 74:1066–1077 DOI 10.1177/0003702820930292.

Cross SL, Baker PA, Seltzer GO, Fritz SC, Dunbar RB. 2000. A new estimate of the Holocene lowstand level of Lake Titicaca, central Andes, and implications for tropical palaeohydrology. The Holocene 10:21–32 DOI 10.1191/095968300671452546.

Dantas DV, Barletta M, Costa MF. 2015. Feeding ecology and seasonal diet overlap between Stellifer brasiliensis and Stellifer stellifer in a tropical estuarine ecocline: feeding ecology and diet overlap of stellifer spp. Journal of Fish Biology 86:707–733 DOI 10.1111/jfb.12592.

De La Barra E, Maldonado M, Vila I, Ibañez C, Jegú M, Carvajal-Vallejos FM. 2020. Review of the biology and taxonomy of the genus Orestias Valenciennes 1839 (Actinopterygii, Cyprinodontiformes). Hidrobiología Neotropical y Conservación Acuática 1:185–224.

Dejoux C, Iltis A. 1992. Introduction. In: Dejoux C, Iltis A, eds. Lake Titicaca: a synthesis of limnological knowledge. Monographiae Biologicae. Dordrecht: Springer Netherlands, XVI–XX.

Desforges J-PW, Galbraith M, Dangerfield N, Ross PS. 2014. Widespread distribution of microplastics in subsurface seawater in the NE Pacific Ocean. Marine Pollution Bulletin 79:94–99 DOI 10.1016/j.marpolbul.2013.12.035.

Di Rienzo J, Casanoves F, Balzarini M, Gonzalez L, Tablada M, Robledo C. 2017. InfoStat. Available at http://www.infostat.com.ar/.

Dusaucy J, Gateuelle D, Perrette Y, Naffrechoux E. 2021. Microplastic pollution of worldwide lakes. Environmental Pollution 284:117075 DOI 10.1016/j.envpol.2021.117075.

Duwig C, Archundia D, Lehembre F, Spadini L, Morel MC, Uzu G, Chincheros J, Cortez R, Martins JMF. 2014. Impacts of anthropogenic activities on the contamination of a Sub Watershed of Lake Titicaca, are antibiotics a concern in the Bolivian Altiplano? Procedia Earth and Planetary Science 10:370–375 DOI 10.1016/j.proeps.2014.08.062.

Eerkes-Medrano D, Thompson RC, Aldridge DC. 2015. Microplastics in freshwater systems: a review of the emerging threats, identification of knowledge gaps and prioritisation of research needs. Water Research 75:63–82 DOI 10.1016/j.watres.2015.02.012.

EFSA Panel on Contaminants in the Food Chain (CONTAM). 2016. Presence of microplastics and nanoplastics in food, with particular focus on seafood. EFSA Journal 14:e04501 DOI 10.2903/j.efsa.2016.4501.
Frias JPGL, Nash R. 2019. Microplastics: finding a consensus on the definition. *Marine Pollution Bulletin* 138:145–147 DOI 10.1016/j.marpollbul.2018.11.022.

Galafassi S, Campanale C, Massarelli C, Uricchio VF, Volta P. 2021. Do freshwater fish eat microplastics? A review with a focus on effects on fish health and predictive traits of MPs ingestion. *Water* 13:2214 DOI 10.3390/w13162214.

Guédron S, Point D, Acha D, Bouchet S, Baya PA, Tessier E, Monperrus M, Molina CI, Groeleau A, Chauvaud L, Thebault J, Amice E, Alanoac L, Duwig C, Uzu G, Lazzaro X, Bertrand A, Bertrand S, Barbraud C, Delord K, Gibon FM, Ibáñez C, Flores M, Fernandez Saavedra P, Ezpinoza ME, Heredia C, Rocha F, Zepita C, Amouroux D. 2017. Mercury contamination level and speciation inventory in Lakes Titicaca & Uru-Uru (Bolivia): current status and future trends. *Environmental Pollution* 231:262–270 DOI 10.1016/j.envpol.2017.08.009.

Guerrera MC, Aragona M, Porcino C, Fazio F, Laurà R, Levanti M, Montalbano G, Germanà G, Abbate F, Germanà A. 2021. Micro and nano plastics distribution in fish as model organisms: histopathology, blood response and bioaccumulation in different organs. *Applied Sciences* 11:5768 DOI 10.3390/app11135768.

Hartmann NB, Hüffer T, Thompson RC, Hassellöv M, Verschoor A, Daugaard AE, Rist S, Karlsson T, Brennholt N, Cole M, Herrling MP, Hess MC, Ivleva NP, Lusher AL, Wagner M. 2019. Are we speaking the same language? Recommendations for a definition and categorization framework for plastic debris. *Environmental Science & Technology* 53:1039–1047 DOI 10.1021/acs.est.8b05297.

Hidalgo-Ruz V, Gutow L, Thompson RC, Thiel M. 2012. Microplastics in the marine environment: a review of the methods used for identification and quantification. *Environmental Science & Technology* 46:3060–3075 DOI 10.1021/es2031505.

Huang J-N, Wen B, Xu L, Ma H-C, Li X-X, Gao J-Z, Chen Z-Z. 2022. Micro/nano-plastics cause neurobehavioral toxicity in discus fish (Symphysodon aequifasciatus): insight from brain-gut-microbiota axis. *Journal of Hazardous Materials* 421:126830 DOI 10.1016/j.jhazmat.2021.126830.

Hyslop EJ. 1980. Stomach contents analysis—a review of methods and their application. *Journal of Fish Biology* 17:411–429 DOI 10.1111/j.1095-8649.1980.tb02775.x.

Ibañez C, Hugueny B, Esquer Garrigos Y, Zepita C, Gutierrez R. 2014. Biodiversidad ictica en el lago Titicaca. In: Pouilly M, Lazzaro X, Point D, Aguirre M, eds. *Línea base de conocimientos sobre los recursos hidrológicos en el sistema TDPS con enfoque en la cuenca del Lago Titicaca*. Quito, Ecuador: IRD-UICN, 134–153.

Lauzanne L. 1992. Native species the Orestias. In: Dejoux C, Iltis A, eds. *Lake Titicaca: a synthesis of limnological knowledge*. *Monographiae Biologicae*, Dordrecht: Springer Netherlands, 405–419.

Lavers JL, Bond AL. 2017. Exceptional and rapid accumulation of anthropogenic debris on one of the world’s most remote and pristine islands. *Proceedings of the National Academy of Sciences of the United States of America* 114:6052–6055 DOI 10.1073/pnas.1619818114.

Lazzaro X, Gamarra C. 2014. Funcionamiento limnológico y fotobiología del Lago Titicaca. In: Pouilly M, Lazzaro X, Point D, Aguirre M, eds. *Línea base de conocimientos sobre los recursos hidrológicos en el sistema TDPS con enfoque en la cuenca del Lago Titicaca*. Quito, Ecuador: IRD-UICN, 134–153.
sobre los recursos hidrológicos e hidrobiológicos en el sistema TDPS con enfoque en la cuenca del Lago Titicaca. Quito: IRD-UICN, 155–217.

Li J, Liu H, Paul Chen J. 2018. Microplastics in freshwater systems: a review on occurrence, environmental effects, and methods for microplastics detection. Water Research 137:362–374 DOI 10.1016/j.watres.2017.12.056.

Lino F, Padilla V. 2014. Uso actual de recursos acuáticos y servicios ecosistémicos del sistema TDPS. In: Pouilly M, Lazzaro X, Point D, Aguirre M, eds. Línea base de conocimientos sobre los recursos hidrológicos e hidrobiológicos en el sistema TDPS con enfoque en la cuenca del Lago Titicaca. Quito: IRD-UICN, 219–250.

Loubens G. 1992. Introduced species 1. Salmo gairdneri rainbow trout). In: Iltis A, Dejoux C, eds. Lake Titicaca: a synthesis of limnological knowledge. Monographiae Biologicae, Dordrecht: Springer Netherlands, 420–426.

Loubens G, Osorio F. 1992. Introduced species 2. Basilichthys bonariensis (the pejerrey). In: Iltis A, Dejoux C, eds. Lake Titicaca: a synthesis of limnological knowledge. Monographiae Biologicae, Dordrecht: Springer Netherlands, 427–444.

Lusher AL, Burke A, O’Connor I, Officer R. 2014. Microplastic pollution in the Northeast Atlantic Ocean: validated and opportunistic sampling. Marine Pollution Bulletin 88:325–333 DOI 10.1016/j.marpolbul.2014.08.023.

Mani T, Hauk A, Walter U, Burkhardt-Holm P. 2016. Microplastics profile along the Rhine River. Scientific Reports 5:17988 DOI 10.1038/srep17988.

McCormick AR, Hoellein TJ, London MG, Hittie J, Scott JW, Kelly JJ. 2016. Microplastic in surface waters of urban rivers: concentration, sources, and associated bacterial assemblages. Ecosphere 7:e01556 DOI 10.1002/ecs2.1556.

McNeish RE, Kim LH, Barrett HA, Mason SA, Kelly JJ, Hoellein TJ. 2018. Microplastic in riverine fish is connected to species traits. Scientific Reports 8:11639 DOI 10.1038/s41598-018-29980-9.

Mizraji R, Ahrendt C, Perez-Venegas D, Vargas J, Pulgar J, Aldana M, Patricio Ojeda F, Duarte C, Galbán-Malagón C. 2017. Is the feeding type related with the content of microplastics in intertidal fish gut? Marine Pollution Bulletin 116:498–500 DOI 10.1016/j.marpolbul.2017.01.008.

Molina C, Lazzaro X, Guédron S, Achá D. 2017. Contaminación de la Bahía de Cohana, Lago Titicaca (Bolivia): Desafíos y oportunidades para promover su recuperación. Ecología en Bolivia 52:65–76.

Monroy M, Maceda-Veiga A, Caiola N, De Sostoa A. 2014. Trophic interactions between native and introduced fish species in a littoral fish community: trophic interactions in Lake Titicaca fishes. Journal of Fish Biology 85:1693–1706 DOI 10.1111/jfb.12529.

Revollo MM. 2001. Management issues in the Lake Titicaca and Lake Poopo system: importance of developing a water budget. Lakes and Reservoirs: Research and Management 6:225–229 DOI 10.1046/j.1440-1770.2001.00151.x.

Sembiring E, Fareza AA, Suendo V, Reza M. 2020. The presence of microplastics in water, sediment, and milkfish (Chanos chanos) at the downstream area of Citarum River, Indonesia. Water, Air, & Soil Pollution 231:355 DOI 10.1007/s11270-020-04710-y.
Silva-Cavalcanti JS, Silva JDB, França EJ de, Araújo MCB de, Gusmão F. 2017. Microplastics ingestion by a common tropical freshwater fishing resource. *Environmental Pollution* **221**:218–226 DOI 10.1016/j.envpol.2016.11.068.

Souza C, Corrêa C, Smith W. 2020. Food ecology and presence of microplastic in the stomach content of neotropical fish in an urban river of the upper Paraná River Basin. *Ambiente & Água - An Interdisciplinary Journal of Applied Science* **15**:e2551 DOI 10.4136/ambi-agua.2551.

Talley TS, Venuti N, Whelan R. 2020. Natural history matters: plastics in estuarine fish and sediments at the mouth of an urban watershed. *PLOS ONE* **15**:e0229777 DOI 10.1371/journal.pone.0229777.

Thompson RC, Moore CJ, vom Saal FS, Swan SH. 2009. Plastics, the environment and human health: current consensus and future trends. *Philosophical Transactions of the Royal Society B: Biological Sciences* **364**:2153–2166 DOI 10.1098/rstb.2009.0053.

Thompson RC, Olsen Y, Mitchell RP, Davis A, Rowland SJ, John AWG, McGonigle D, Russell AE. 2004. Lost at sea: where is all the plastic? *Science* **304**:838–838 DOI 10.1126/science.1094559.

Vila I, Pardo R, Scott S. 2007. Freshwater fishes of the Altiplano. *Aquatic Ecosystem Health & Management* **10**:201–211 DOI 10.1080/14634980701351395.

Wright SL, Thompson RC, Galloway TS. 2013. The physical impacts of microplastics on marine organisms: a review. *Environmental Pollution* **178**:483–492 DOI 10.1016/j.envpol.2013.02.031.