

## Article

# Diversity of Freshwater Mollusks from Lake Pampulha, Municipality of Belo Horizonte, Minas Gerais, Brazil

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**Abstract:** The artificially created Lake Pampulha, within the metropolitan area of Belo Horizonte, Minas Gerais State, lacks detailed information about its freshwater mollusks, representing a challenge for the assessment and conservation of this ecosystem. In this study, conducted during June and August 2021, we collected specimens on four different occasions and at five sampling points around the lake, using different sampling equipment, such as a shovel and a scoop. During these collections, we identified, enumerated and measured 1538 species of mollusks and additionally examined the presence of trematode larvae. We identified twelve species of fresh water bivalves and gastropods: *Biomphalaria straminea*, *Biomphalaria kuhmiana*, *Biomphalaria occidentalis*, *Drepanotrema cimex*, *Pomacea maculata*, *Stenophysa marmorata*, *Physa acuta*, *Gundlachia ticaga*, *Melanoides tuberculata*, *Pseudosuccinea columella*, *Omalonyx matheroni* and *Corbicula largillierti*. Echinostome and strigeocercaria types of larval trematodes were detected in *B. straminea*. Notably, some species of mollusks have not previously been recorded at Lake Pampulha. The analyses revealed differences in the composition and abundance of species, highlighting the higher number of mollusk species in areas more impacted by human actions. This study expands our understanding of mollusk diversity at Lake Pampulha, and provides valuable data for longitudinal comparisons of water quality and considerations of the conservation of native species. Furthermore, it highlights the importance of choosing appropriate sampling equipment, depending on the research objectives. The presence of invasive species of medical and veterinary relevance as intermediate hosts of parasites reinforces the need for efficient environmental protection strategies to preserve this artificial, aquatic environment widely used by the local population and by tourists.

**Keywords:** freshwater; biodiversity assessment; conservation; *Biomphalaria*; collectors; ecology



**Citation:** Coelho, P.R.S.; Thiengo, S.C.; de Mendonça, C.L.F.; de Oliveira, N.M.T.; dos Santos, S.B.; Caldeira, R.L.; Geiger, S.M. Diversity of Freshwater Mollusks from Lake Pampulha, Municipality of Belo Horizonte, Minas Gerais, Brazil. *Diversity* **2024**, *16*, 193. <https://doi.org/10.3390/d16040193>

Academic Editors: Michael Wink, Ioan Sirbu and Simone Varandas

Received: 30 December 2023

Revised: 23 February 2024

Accepted: 19 March 2024

Published: 24 March 2024



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## 1. Introduction

On a global scale, it is estimated that there are more than 5000 species of limnic mollusks. In Brazil, so far, 293 species have been officially described, but estimates speculate numbers between 340 and 359 species [1]. However, the lack of basic information about these species throughout the country makes it difficult to accurately assess their conservation status and distribution, which are considered crucial aspects for the definition of threat levels [1].

The diversity of limnic mollusks, a faunal group of significant relevance in aquatic ecosystems, has been the subject of various studies in which mollusks were used as ecological indicators [2–4]. However, Lake Pampulha, an artificial freshwater ecosystem, located in the municipality of Belo Horizonte, Minas Gerais, Brazil, lacks substantial information, with only a few studies available on this issue [5]. Lake Pampulha is included in the “Modern Pampulha Set”, designated as cultural heritage of humanity since 2016; it is widely recognized for its scenic beauty and ecological importance. However, the integrity of this ecosystem is intrinsically related to the diversity and interactions between the species that inhabit it, including its mollusks species.

Limnic mollusks, including freshwater snails and mussels, face global threats due to the degradation of aquatic environments, which result from natural factors and human interventions such as urbanization, pollution, deforestation and land use changes [6]. Their sensitivity to habitat changes, combined with low growth rates and limited dispersal, makes them particularly vulnerable to the impact of introduced exotic species and climate change [6,7]. Furthermore, their ability to serve as intermediate hosts for trematodes of medical and veterinary importance further increases the threats they face, intensifying dangers related to population reduction and habitat degradation [8].

The Lake Pampulha reservoir, initially designed in the late 1930s for water supply and flood control purposes, underwent a significant transformation, and became a recreation area and, unfortunately, with urbanization, a focus of schistosomiasis transmission. Given this context, several malacological studies were conducted here, but focused exclusively on *Biomphalaria* spp., the intermediate hosts of *Schistosoma mansoni* Sambon, 1907, the trematode that causes intestinal schistosomiasis [4,9–16].

In view of this, the study of mollusk diversity in Lake Pampulha plays a crucial role in assessing the conservation status of this unique aquatic ecosystem. A current (Figure 1) and retrospective analysis of its diversity can provide important information about the integrity of the ecosystem, allowing the identification of possible imbalances and pollution at an early stage, thus contributing to its preservation and sustainability.



**Figure 1.** Illustration of the malacological survey at Lake Pampulha, Belo Horizonte, Minas Gerais, Brazil, during the winter season 2021. Representative picture of the manual collection and monitoring of medically important limnic mollusks, as recommended by the Brazilian Ministry of Health, e.g., for *Biomphalaria* spp., natural, intermediate host of *Schistosoma mansoni*, the parasite that causes intestinal schistosomiasis.

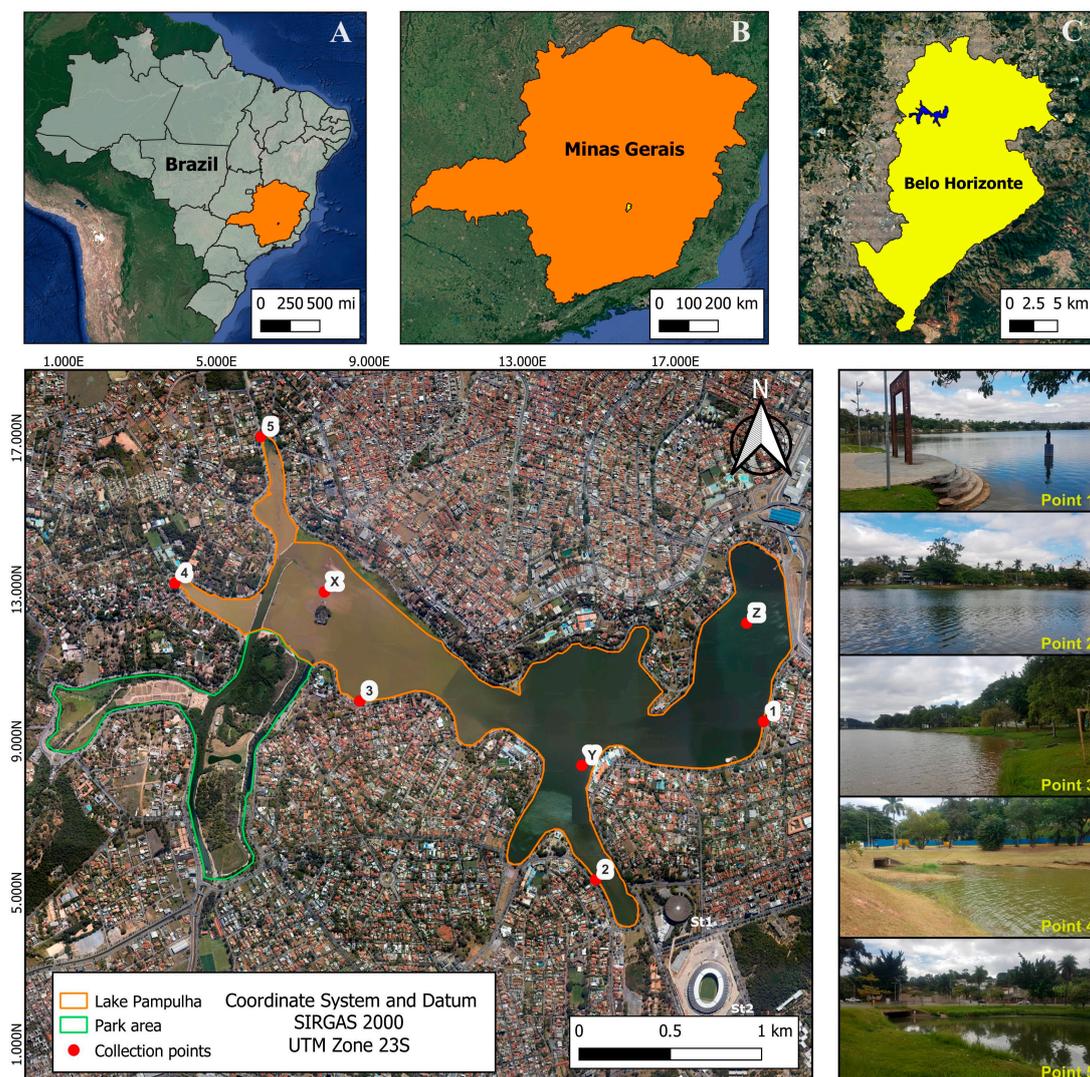
In this study, we sought to identify gaps not only in the approach to mollusk diversity, but also in the investigation of possible parasitic infections at Lake Pampulha. We used two pieces of equipment for the malacological survey and characterized the environment in a descriptive way, complementing the assessment of water quality with laboratory analyses.

The results cover the local fauna and point to the need for effective measures to conserve and improve the management of water resources.

## 2. Materials and Methods

### 2.1. Study Area

The present study was carried out at Lake Pampulha ( $19^{\circ}51'04''$  S  $43^{\circ}58'46''$  W), located in the city of Belo Horizonte, capital of the State of Minas Gerais, Brazil (Figure 2). The climate in the region is high-altitude tropical (Cwa), according to the Köppen classification, with a dry winter and a rainy summer. The average annual temperature is  $21.1^{\circ}\text{C}$ , with little variation throughout the year, while the average rainfall is 1463.7 mm, with an average of 292 mm in December, the wettest month, and an average precipitation of only 11.5 mm in June, according to the National Institute of Meteorology—INMET [17,18].



**Figure 2.** The illustrative maps (A–C) indicate the State of Minas Gerais (orange) within Brazil (A) and within the southeastern region of Brazil (B), where the capital is indicated in yellow and Lake Pampulha is indicated in blue, within the municipality (C). The bird’s-eye view indicates the extension of the water body (orange line) as well as the points of mollusk sampling (1–5) and water analysis (X–Z) at Lake Pampulha, Belo Horizonte, Minas Gerais. In the larger figure, it is possible to observe dense urbanization around the lake, as well as the tourist attractions close to point 2, with the sports stadiums Mineirinho and Mineirão (St1, St2). Each sampling point is illustrated by a photograph on the right (Points 1–5).

Lake Pampulha is part of the waters of the Onça Basin and consists of eight tributaries, the Mergulhão, Tejuco, Ressaca, Sarandi, Água Funda, Baraúna, Olhos D'água and AABB streams. The lake is a tributary of the Das Velhas River and a subtributary of the important São Francisco River. The "lake" exists as an artificial reservoir and was inaugurated in 1938, in order to serve as a water supply for the capital [16,17]. In the years following the formation of the reservoir, the site was used for water sports, especially the use of canoes, boats, speedboats and yachts, and even hosted municipal and national competitions [17,18]. Currently, despite the inferior water quality, the lake often features on picture postcards, promoting the city and its surroundings, which are often used for festive events, major sports events, and recreation [19] (Figure 2). Considering its historical and cultural importance, the architectural ensemble of Lake Pampulha was awarded cultural heritage of humanity by UNESCO in March 2016.

## 2.2. Methodological Framework for Mollusk Analysis: From Sampling Techniques to Statistical Insights

### 2.2.1. Sampling Methodology and Georeferencing Techniques

Mollusks were sampled at five locations, during the months of June and August 2021, during the dry winter season. The mollusks were captured by two trained collectors, with a sampling effort of 30 min per individual at each sampling point, according to the technique by Olivier and Schneiderman [20]. At each of the 5 collection points, 4 separate collections were carried out (2 collections per month with alternation of the collection devices), totaling 20 samplings for the entire survey (10 collections in June and 10 collections in August). For the design of the map in QGIS, the databases of the Brazilian Institute of Geography and Statistics (IBGE) and Cartography based on geocoding, projection/datum with SIRGAS 2000 UTM Zone 23S, were used. The georeferencing of the lake (latitude and longitude) was undertaken using portable GPS (Garmin, GPSMAP 62S).

### 2.2.2. Uniform Collection Practices and Environmental Considerations

In order to minimize variability in sampling intensity among different individuals, all collections were carried out by the same researchers. Specimens were collected from 8:00 am to 1:00 pm, following a pattern of alternation in the sequence of collection points on each sampling day. At least one week before collections took place, there was no rain in the neighborhoods and there were no anthropogenic interventions to the banks, such as cleaning or cutting the grass.

### 2.2.3. Collection Instruments and Statistical Evaluation of Mollusk Shells

Scoops and shovels were used to capture mollusks, as recommended by the Brazilian Schistosomiasis Control Program (Ministério da Saúde, 2008) [21] (Figure 3). All collected mollusks were stored in transparent plastic bags, together with decomposing leaves to maintain humidity. After sampling took place, the biological material was transported to the Laboratory of Intestinal Helminths at the Federal University of Minas Gerais, along with a capture form sheet, containing date, place of collection, and the identification of the collectors.

The dimensions of the shells of the genus *Biomphalaria* and *Pomacea* and the collection instruments (scoop and shovel), along with any association between them, were statistically evaluated using RStudio software (version 2023.09.1+494). The statistical analysis of these dimensions included the application of median tests, such as the Student's t test and the Mann–Whitney U-test.



### Scoop

- Perforated aluminum Scoop
- Size of holes: 3mm
- Scoop area: 307.9cm<sup>2</sup>
- Diameter of Scoop: 15cm
- Cable: 131cm total
- Weight: 481.78g

### Shovel

- Netted Shovel
- Screen: 1.5mm
- Shovel area: 170.5cm<sup>2</sup>
- Cable: 120cm
- Weight: 566.8g

**Figure 3.** Equipment used for the collection of mollusks, as recommended by the Brazilian Ministry of Health (Ministério da Saúde, 2008) [21]. The scoop was handcrafted and the shovel was provided by LOGNature®.

### 2.3. Molluska Taxonomy

The identification of mollusks was based on the available literature [21–28]. From each batch of planorbid snails collected, about 10% of the specimens were removed and sacrificed in hot water at 70 °C. The soft parts were fixed in Railliet–Henry solution and dissected under a stereomicroscope. A sample of foot tissue from *Biomphalaria* spp. was used to identify species via molecular methods, using the PCR-RFLP technique [29,30].

For reference and identification, each specimen was photographed and a voucher collection was maintained for all morphospecies. The entire field procedure was authorized by the Chico Mendes Institute for Biodiversity and Conservation, through the Biodiversity Authorization and Information System (SISBIO), permission number 68627. The collected material was deposited in the Medical Malacology Collection (Fiocruz/CMM), at the Mollusk Collection of Oswaldo Cruz Institute (Fiocruz/CMIOC) and at the laboratory for Limnic and Terrestrial Malacology of the State University of Rio de Janeiro (UERJ). To complement this, we consulted the literature and scientific collections to identify research on limnic mollusks from Lake Pampulha, in order to compare them with the data obtained in this study.

### 2.4. Community Ecology Analysis

Our research used two collection devices that employed a similar manual methodology. We compared these two devices in relation to several ecological aspects, including richness, dominance, equitability, Simpson's 1-D and Shannon\_H, which were calculated using PAST software 4.13 [31,32].

### 2.5. Parasitological: Evaluation of Trematodes

In the laboratory, after manual sorting, the mollusks were counted, measured with the aid of a digital caliper (precision 0.01 mm), observed macroscopically and then placed individually in cell culture plates containing 5–15 mL of non-chlorinated water, and then left overnight for examination before and after direct artificial photostimulation (60 W lamp) [33], except for the *Omalonyx* specimens. The following day, after 4 h of exposure to light, the material was examined for the emergence of cercariae and identification was based on morphological criteria, as described elsewhere [34]. Examinations were

performed on a weekly basis and after 30 days those snails that remained negative were crushed and compressed between glass plates, in order to search for possible sporocysts and/or metacercariae.

### 2.6. Water Quality Assessment

To characterize the water quality of Lake Pampulha, secondary data from ongoing surveillance by the Instituto Mineiro de Gestão das Águas (IGAM) were used, taken from the same time period as for the mollusk collections. Water monitoring included three specific sampling points: close to Ilha dos Amores (X), close to the São Francisco church (Y) and close to the spillway (Z) [35,36]. The parameters used in the water assessment included a Water Quality Index, which considers a combination of three groups of indicators: organic enrichment, fecal contamination and the presence of toxic substances.

## 3. Results

### 3.1. Malacology

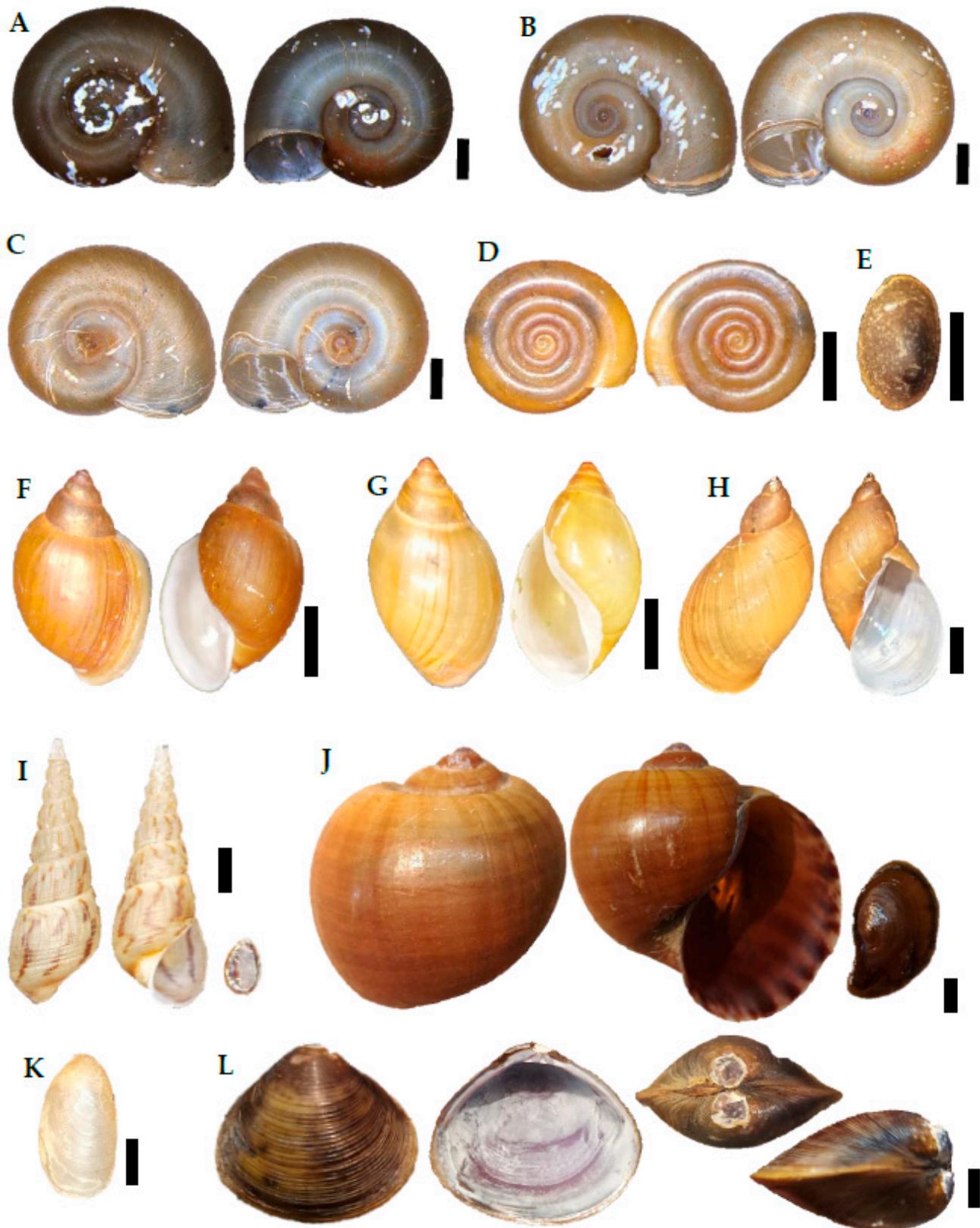
A total of 1538 specimens, belonging to twelve mollusk species, were collected and identified in the study area, which included *Biomphalaria straminea* (Dunker, 1848), *Biomphalaria kuhniiana* (Clessin, 1883), *Biomphalaria occidentalis* Paraense, 1981, *Drepanotrema cimex* (Moricand, 1837), *Pomacea maculata* Perry, 1810, *Stenophysa marmorata* (Guilding, 1828), *Physa acuta* (Draparnaud, 1805), *Gundlachia ticaga* (Marcus and Marcus, 1962), *Melanooides tuberculata* (Müller, 1774), *Pseudosuccinea columella* (Say, 1817), *Omalonyx matheroni* (Pontiez and Michaud, 1835) and *Corbicula largillierti* (Philippi, 1844) (Table 1 and Figure 4).

**Table 1.** Diversity, distribution and identification of native and invasive species of limnic mollusks from Lake Pampulha, Belo Horizonte, Minas Gerais, Brazil, during the surveyed winter season of 2021.

| Specimens                        | Lake Pampulha |         |         |         |         |
|----------------------------------|---------------|---------|---------|---------|---------|
|                                  | Point 1       | Point 2 | Point 3 | Point 4 | Point 5 |
| <i>Biomphalaria straminea</i>    | X             | X       | -       | -       | X       |
| <i>Biomphalaria kuhniiana</i>    | -             | -       | -       | X       | -       |
| <i>Biomphalaria occidentalis</i> | -             | -       | X       | -       | -       |
| <i>Drepanotrema cimex</i>        | -             | -       | X       | -       | -       |
| <i>Gundlachia ticaga</i>         | X             | X       | -       | -       | -       |
| <i>Stenophysa marmorata</i>      | X             | X       | X       | -       | X       |
| <i>Physa acuta</i> *             | X             | X       | -       | -       | X       |
| <i>Pseudosuccinea columella</i>  | -             | X       | X       | -       | X       |
| <i>Pomacea maculata</i> *        | X             | X       | X       | X       | X       |
| <i>Melanooides tuberculata</i> * | X             | X       | X       | -       | -       |
| <i>Omalonyx matheroni</i>        | -             | X       | -       | -       | X       |
| <i>Corbicula largillierti</i> *  | -             | -       | -       | X       | -       |

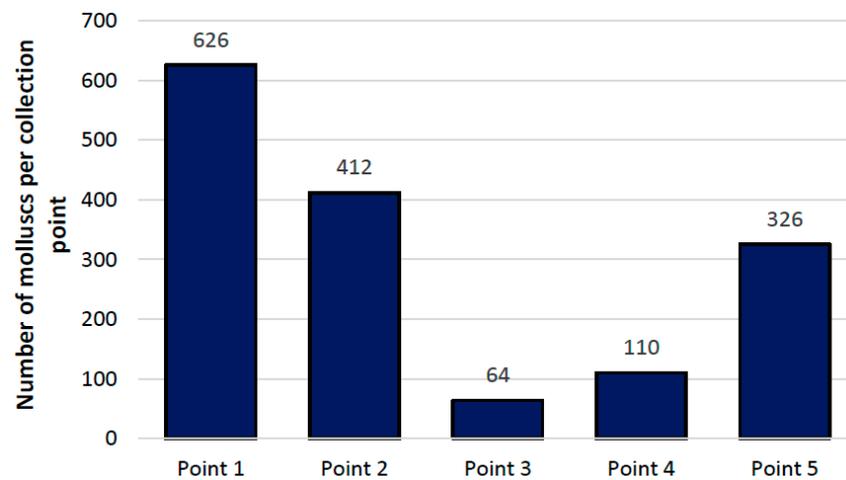
\*: non-native species.

Table 1 further outlines the distribution and diversity of these native and invasive species across different collection points (Point 1, Point 2, Point 3, Point 4, and Point 5) at Lake Pampulha, providing a comprehensive overview of the mollusk community in the ecosystem during the specified period.



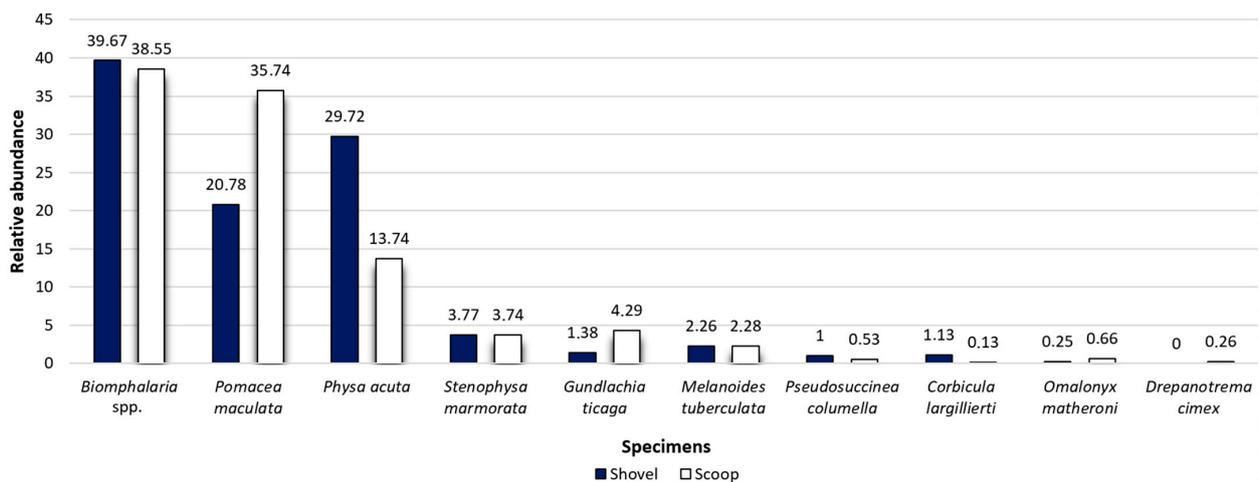
**Figure 4.** Species of limnic mollusks found at Lake Pampulha, Belo Horizonte, Minas Gerais, Brazil: (A) *Biomphalaria straminea*; (B) *Biomphalaria kuhniiana*; (C) *Biomphalaria occidentalis*; (D) *Drepanotrema cimex*; (E) *Gundlachia ticaga*; (F) *Physa acuta*; (G) *Stenophysa marmorata*; (H) *Pseudosuccinea columella*; (I) *Melanoides tuberculata*; (J) *Pomacea maculata*; (K) *Omalonyx matheroni*; (L) *Corbicula largillierti*. Scale bar: (A–D) 1 mm; (F–G) 2 mm; (H–I) and (K) 3 mm; (J) and (L) 5 mm.

The results presented in Figure 5 provide an overview of the relative abundance of mollusks at five collection points at Lake Pampulha. Point 1 stands out as an area with a substantial mollusk count ( $n = 626$ ), demonstrating its richness in aquatic species. Point 2 follows, with 412 specimens, while Point 3 shows a lower abundance, with 64 collected mollusks. Point 4 and Point 5 had intermediate counts of 110 and 326, respectively.



**Figure 5.** Total numbers of mollusks collected per collection point at Lake Pampulha, Belo Horizonte, Minas Gerais, Brazil, in June and August 2021.

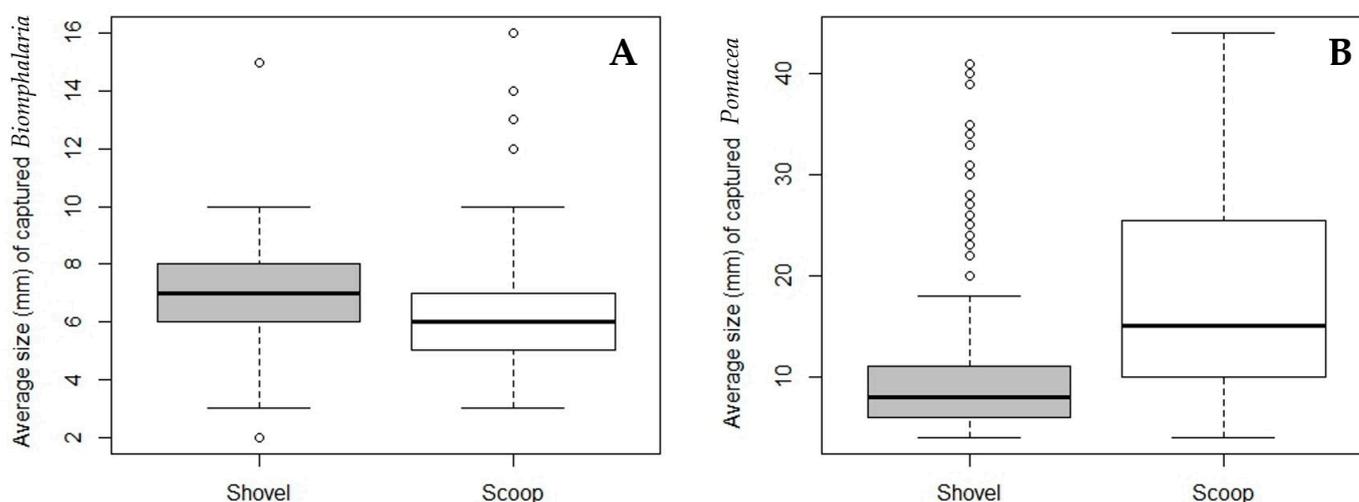
Figure 6 shows differences in the percent distribution of aquatic mollusks when subjected to two different types of sampling equipment: shovel and scoop. A subtle predominance of *Biomphalaria* was observed in shovel sampling in relation to scoop sampling, while *Pomacea maculata* presented a more expressive variation between the two sampling devices. *Physa acuta*, in turn, was more easily captured by the shovel. It is important to highlight that *Drepanotrema cimex* was hardly present in either of the equipment-type groups, suggesting possible scarcity or preference for environments not covered by the methods used.



**Figure 6.** Relative frequency of mollusk species collected from Lake Pampulha, Belo Horizonte, Minas Gerais, Brazil, during the winter season 2021 and depending on the sampling device used (shovel or scoop).

After collection, the mollusks were measured and subjected to comparisons between collection equipment, aiming to identify performance standards (Figure 7). The results of the Wilcoxon test reveal significant differences for *Biomphalaria* (A) and *Pomacea* (B), when

collected with the shovel and scoop equipment ( $p$ -value:  $<0.0001$  and  $p$ -value:  $\leq 0.0001$ , respectively).



**Figure 7.** Mean average shell size of *Biomphalaria* (A) and *Pomacea* (B), collected at Lake Pampulha with two different sampling devices, e.g., shovel (grey boxplots) and scoop (white boxplots). Bold horizontal lines indicate the mean values, with upper and lower boxes indicating 75 and 25% confidence intervals, respectively, and whiskers indicating the 95 and 5% confidence intervals.

### 3.2. Community Ecology Analysis

The results of the ecological diversity analysis, considering the different collection points and sampling equipment used (shovel or scoop), reveal small variations in the compositions of the communities at each sampling point (Table 2). In general, the shovel device appeared to yield slightly lower diversity indices, with richness, evenness and Shannon index (Shannon\_H) values, when compared with the scoop device. Furthermore, the shovel tended to exhibit higher dominance values, indicating that some species can increase the dominance index when this equipment is used during collection. On the other hand, the scoop method generally showed higher diversity indices, suggesting that it captures greater species richness and a more equitable distribution of abundances.

**Table 2.** Richness, dominance, equitability and diversity indices calculated with the different sampling devices at the five different sampling points at Lake Pampulha, city of Belo Horizonte, Minas Gerais, Brazil.

| Indexes      | Point 1 |        | Point 2 |        | Point 3 |        | Point 4 |         | Point 5 |        |
|--------------|---------|--------|---------|--------|---------|--------|---------|---------|---------|--------|
|              | Shovel  | Scoop  | Shovel  | Scoop  | Shovel  | Scoop  | Shovel  | Scoop   | Shovel  | Scoop  |
| Richness     | 6       | 6      | 6       | 7      | 5       | 5      | 2       | 3       | 6       | 4      |
| Dominance    | 0.4177  | 0.4484 | 0.4799  | 0.3213 | 0.3015  | 0.4634 | 0.7545  | 0.9158  | 0.338   | 0.6387 |
| Equitability | 0.5787  | 0.5993 | 0.6065  | 0.7505 | 0.8889  | 0.6356 | 0.5971  | 0.2064  | 0.6979  | 0.5109 |
| Simpson 1-D  | 0.5823  | 0.5516 | 0.5201  | 0.6787 | 0.6985  | 0.5366 | 0.2455  | 0.08418 | 0.662   | 0.3613 |
| Shannon H    | 1.037   | 1.074  | 1.087   | 1.46   | 1.431   | 1.023  | 0.4139  | 0.2267  | 1.25    | 0.7083 |

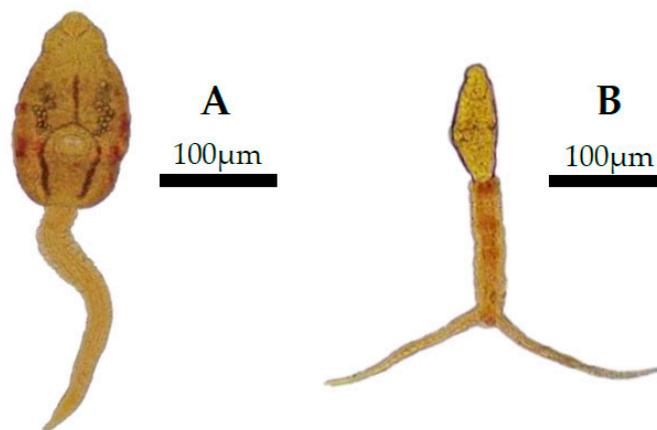
Although both devices demonstrated equal performance capacities in collecting *Biomphalaria*, it is crucial to select the most appropriate equipment to achieve specific objectives in ecological studies.

Simpson’s 1-D index measures the probability that two randomly chosen sampling units belong to the same species. Higher values indicate lower diversity, as two units are less likely to belong to different species. Finally, the Shannon\_H index, considering both

richness and evenness, combines information on the number of species and the distribution of abundances to provide a global measure of diversity in the studied community [31,32].

### 3.3. Evaluation of Parasitic Trematodes

The analysis of trematode larvae, associated with the collected mollusks, revealed the presence of larval stages identified as cercariae of the types echinostome (9/603) and Strigeocercariae (24/603). It is noteworthy that infection was observed in 33/603 specimens of *Biomphalaria* spp., resulting in an infection rate of 5.5%. It is also of importance to highlight that the cercariae were eliminated exclusively by mollusks of the species *Biomphalaria straminea* (Figure 8).



**Figure 8.** Cercariae eliminated by *Biomphalaria straminea*, identified as types echinostome (A) and Strigeocercaria (B) (photographed SDPTOP mode: OD400UHW-P).

### 3.4. Descriptive Characterization of Collection Environments and Additional Water Parameters

Point 1 stands out for the greater relative abundance of mollusks collected compared to the other locations (Figure 5). There are several hypotheses that can explain this phenomenon. One of them is related to environmental characteristics, notably the accumulation of organic matter. Furthermore, it is important to mention that this point is known for being the scene of religious rituals, especially linked to the veneration of Iemanjá. During these rituals, numerous offerings are deposited in the water, including grains of rice and beans, creating a favorable environment for the increase in mollusk biomass.

Point 2 was the only environment where trees were close to the banks of the lake, with decomposing leaves accumulating at the bottom of the lake. This environment revealed the highest richness of mollusk species during the collections (Table 2). On the other hand, point 3 stood out for its lower abundance compared to the other collection sites (Figure 5). The distinct characteristics of this environment, such as the scarcity of vegetation, the significant accumulation of plastic waste, the oiliness and the stagnant water, most probably influenced the peculiar ecological composition observed in this area.

Points 4 and 5 were located close to the mouths of their tributaries, AABB and Olhos d'água, respectively. Both locations shared similar environmental problems, such as a large volume of urban sewage, the presence of a bad odor and the accumulation of plastic waste. These environmental challenges may have affected water quality and mollusk biodiversity at these locations. The presence of urban sewage is especially worrying, as it can affect the health of aquatic ecosystems and the survival of many species. This discussion highlights the complexity of the interaction between the environment and the biodiversity of the fauna, and the importance of considering these factors in ecological studies. Furthermore, these areas are at epidemiological risk due to contamination by water-borne parasitic diseases, such as schistosomiasis and fascioliasis.

To complement the characterization of the water quality of Lake Pampulha, we used biological, physical and chemical data that demonstrate the deterioration of water quality.

We used parameters provided by IGAM, which indicate that points X, Y and Z did not meet the thresholds established for the parameters of Biochemical Oxygen Demand (BOD), *Escherichia coli*, cyanobacteria, cyanotoxin–microcystin, total phenols and phosphorus compounds. Additionally, Point “X” also exceeded the thresholds for free cyanide.

#### 4. Discussion

The results of this study reveal crucial insights into the malacofauna of Lake Pampulha during the winter season of 2021. Notably, Point 1 stood out with an expressive count of mollusks, most probably associated with religious rituals and the accumulation of organic matter, thus highlighting the complex interactions between cultural practices and aquatic ecosystems. In addition, the presence of parasites in one mollusk species pointed to possible risks for public health. Furthermore, environmental factors, such as plastic waste and urban sewage, emerged as threats to water quality and biodiversity, demanding specific conservation strategies. The results from previous studies corroborate that impacted environments can induce significant changes in the biodiversity and dynamics of aquatic ecosystems [37]. These findings emphasize the urgency of collaborative efforts towards the sustainable preservation of these ecosystems.

##### 4.1. Malacology

###### 4.1.1. Invasive Mollusks

The introduction of invasive species was identified as the main cause of reducing biodiversity, causing the displacement of native species and changes in the structure of biological communities, and facilitating the spread of diseases [38–41]. In the present study, it was observed that around a third of the species collected at Lake Pampulha consisted of invasive mollusks.

Among the invasive species, we highlight the unprecedented record of *Corbicula largillierti* at Lake Pampulha, a freshwater bivalve belonging to the family Corbiculidae (Bivalvia, Heterodonta, Veneroidea), originating in Asia and endemic in the Yangtze River system, in China [42]. This bivalve has already been identified in several regions of South America [42]. In Brazil, the first observation occurred in the Pantanal region of Mato Grosso State, as recorded by [43], and in a first record from Minas Gerais State [44].

*Physa acuta*, also known as *Physella acuta* and originally from North America, has a wide distribution throughout Brazil, as documented in several studies [4,45–47]. These snails are common in eutrophic lakes and river environments in various regions of the world, covering Europe, Asia, Africa, Australia, North America and South America [48–53]. The presence of this organism represents a threat to the species *S. marmorata*, which shares the same niche and presents similar morphological characteristics. *Stenophysa marmorata*, corresponding to the species *Physa marmorata*, “Name uncertain”, a native species, is listed as “vulnerable” in the catalog of threatened species of the Ministry of the Environment (MMA) and the Chico Mendes Institute for Biodiversity Conservation (ICMBio), although further studies are required in order to make assertions on its current distribution.

Among the collected species, *M. tuberculata*, originally from Southeast Asia and the northern and eastern regions of Africa, is classified as an invasive species introduced in Brazil in the 1960s [54]. In 1986, the first appearance was reported at Lake Pampulha [55]. Over the years, the species has established significant population densities in this location [56,57], a phenomenon that was repeated in several parts of the country [54,58–60].

Ampullarids of the genus *Pomacea* Perry, 1810 have been recorded at Lake Pampulha since the 1940s. Their populations increased considerably after the reconstruction of the dam in the late 1950s, when the species was initially designated as *P. haustum* (Reeve, 1856) according to [11]. This nomenclature was adopted by several researchers in their subsequent studies [4,11,15,61–63]. Later on, it was reclassified as *P. insularum* (d’Orbigny, 1839) according to Paulinyi and Paulini (1972) [64], and, most recently, as *Pomacea canaliculata* (Lamarck, 1822) [65] and *Pomacea dolidoides* (Reeve, 1856) [66]. However, the identification of *P. haustum* has been undertaken using biological evidence such as the color of the egg

mass, which is green for this species, and is clearly different from the pink egg mass of the species present in the region, as observed by [64]. This study highlights the exclusive presence of the species *P. maculata* in all analyzed points, as confirmed by Hayes et al., 2012, clarifying that *P. insularum* is now considered synonymous with *P. maculata* [67]. It is important to highlight that *P. maculata* is also classified as an invasive species.

#### 4.1.2. Taxonomic Complexities and Unexplored Diversity

With regard to the species *B. occidentalis* and *B. kuhniiana*, this was their first recording at Lake Pampulha, although these species have already been present in the scientific collections of Fiocruz/CMM since 2005 and 2018, respectively. These species have received little attention regarding their distribution, as they are not considered to be of medical relevance. However, it is worth mentioning that *B. kuhniiana* was previously identified in Minas Gerais, as mentioned by Coelho et al., 2022 [68]. In this respect, the close morphological similarity between different species within the genus (11 species and one subspecies [69]) can facilitate errors in correct identification. Therefore, an integrative taxonomic approach (morphological and molecular) is recommended.

In malacological surveys, it is possible for different species of *Biomphalaria* to be present at the same collection point. Of the 603 specimens of *Biomphalaria* collected, only 10% were thoroughly identified, leaving a considerable number of as-yet unexplored diversity. In this study, the analyses of the relative frequency and average size of the shells were restricted to the genus *Biomphalaria*, due to the complexity and the need for more specific resources for a more accurate identification of the specimens. The discovery of a potentially greater diversity within this group of mollusks highlights the importance of future investigations and improvements of identification techniques, with the aim of understanding the ecology and distribution of these species in a more comprehensive and detailed way.

*Pseudosuccinea columella* was already mentioned in studies carried out during the last century at Lake Pampulha [70,71], a species that was formerly classified as *Lymnaea columella*.

Based on information from the literature and from scientific collections, we listed a total of 23 distinct species of mollusks that have previously been found at Lake Pampulha (Table S1). It is important to emphasize that some species, which were not collected in our study, such as *B. glabrata*, *B. tenagophila*, *B. schrammi*, *Drepanotrema lucidum*, *Drepanotrema depressissimum*, *P. haustum*; *P. canaliculata*, *P. dolidoides*, *Ancylus* sp., *Hebetancylus moricandi* and *Pisidium* sp., are detailed in the Supplementary Material Table S1. However, contrary to recent malacological surveys and/or findings in scientific collections (Fiocruz/CMM 2023; UERJ Collection 2023), no specimens of *B. schrammi*, *Drepanotrema depressissimum*, *P. canaliculata*, *P. dolidoides* and *Ancylus* sp. were found. The absence of these species in the sampling areas does not necessarily exclude them from Lake Pampulha as a whole, as they may be present in other regions of the reservoir not found by this study. Furthermore, it is plausible that changes in environmental conditions, adding to the continuous pollution of the reservoir, may have played a limiting role in the distribution of these mollusk species in the region. It is worth mentioning that *Ancylus* sp., although it was mistakenly identified at Lake Pampulha by [11], does not occur naturally in America, it being a species exclusive to Europe. Finally, the species *P. dolidoides* was shown to be distributed in the northern region of Brazil, with no records in the southeastern region [67].

#### 4.2. Sampling Challenges

The evaluation of two different collecting devices has revealed different and contrasting performances in relation to the sizes of the mollusks and collected species. We observed that the scoop produced inverse results for *Pomacea* and *Biomphalaria* compared to the shovel. In this regard, a field observation relevant to this disparity stands out: the absence of edges on the shovel made it difficult to collect *Pomacea*, especially for species of mollusks with a more rounded shape, which could roll and escape during sampling. The shovel also caused the loss of smaller specimens of *Biomphalaria*, which, due to their reduced weight, were easily swept away from the shovel.

However, it is clear that the use of any of these pieces of equipment is very useful in malacological surveys of the Schistosomiasis Control Programs, allowing the effective epidemiological monitoring of *Biomphalaria* specimens.

Regarding the collection of mollusks that inhabit deep sediment, such as *Corbicula*, we observed a reduced efficiency with the sampling devices, such that no living specimens were collected. A more efficient capture of this type of mollusk would require the use of a specific type of digging equipment. Thus, this field observation pointed out the importance of the kind of sampling device and its influence on the collected mollusk species, depending on differences in the morphology, size, and habitat of the species.

#### 4.3. Parasitology: Trematode Infections

In the context of species that were identified as potential intermediate hosts of parasitic diseases, *B. straminea* stands out for its role in the transmission of *S. mansoni*, and *P. columella*, with a similar function in the life cycle of *Fasciola hepatica* (Linnaeus, 1758). In addition, the non-native species *M. tuberculata* has been identified as a possible intermediate host for 37 species of trematodes in veterinary and medical parasitology [47,72]. It is important to highlight that at Lake Pampulha, the last record of the presence of *S. mansoni* in *B. glabrata* was documented by Pinto et al., 2013 [73], as well as the presence of the parasite *Centrocestus formosanus* (Nishigori, 1924) in *M. tuberculata*, by the same group of researchers [74].

*Biomphalaria straminea* was first reported at Lake Pampulha in the 1980s, subsequently serially replacing other predominant snail species that are intermediate hosts of *S. mansoni*, such as *B. glabrata* and *B. tenagophila* [73]. To date, *B. straminea* has not yet been found naturally infected with *S. mansoni* at Lake Pampulha, but the species *B. glabrata* and *B. tenagophila* were [10–12,14,61,62,75].

In the present study, only specimens of the species *B. straminea* ( $n = 33$ ) showed infections with cercariae of the echinostome and strigeocercariae types. These trematodes generally have complex life cycles, involving amphibians, reptiles, birds and mammals as definitive hosts [34]. In order to further identify and characterize the aforementioned cercariae, the recovery of adult parasites is indispensable, as are additional studies involving molecular analyses of molecular and/or experimental infections [34,76]. According to recent data [77], 27 species of trematodes have already been found in different vertebrate and non-vertebrate hosts (mollusks, fish, insects and birds) collected at Lake Pampulha, indicating a wide diversity of trematode parasites at this site.

Regarding the possible biological competition between *S. mansoni* intermediate hosts and other snail species, such as *Pomacea* sp. and *M. tuberculata*, studies carried out over more than four decades in the Pampulha region have indicated that, despite coexistence, populations of *Biomphalaria* spp. have remained stable [4,11,15,16,77], as also evidenced by our findings. This observation is in line with typical scenarios of eutrophic and unstable environments, in which snails encounter abundant access to food [78,79].

The changed environmental conditions at Lake Pampulha favor the epidemiological chain for the transmission of parasitic trematode species. However, most malacological studies focusing on trematode larvae other than *S. mansoni* are based on spot samples, and longitudinal analyses of infection rates were generally not performed [4,15,58,80–82].

#### 4.4. Eutrophication and Water Quality

The malacological diversity herein reported may be related to the reservoir's eutrophication state, caused by the high content of organic matter dumped at the site, along with biotic factors, such as the proliferation of phytoplankton algae and aquatic macrophytes, and abiotic factors, such as nutrients, conductivity and oxygen. These conditions may favor the growth and reproduction of mollusk species and may result in denser populations. The presence of considerable mollusk populations, together with diverse water-associated vertebrate communities (birds, reptiles, small mammals, and fish), might create a favorable environment for trematode infections and high infection rates in these hosts [83–89].

Water quality parameters monitored by IGAM play a crucial role in assessing the environmental condition of water bodies. These indicators not only make it possible to analyze the impacts of pollution, but also allow for monitoring the effects of revitalization interventions at Lake Pampulha. The use of mollusks, especially freshwater snails, as bioindicators in aquatic ecological studies is recognized due to the remarkable sensitivity of some species to environmental changes [90–93]. However, the need to develop specific protocols to evaluate the impacts of these contaminants on mollusks in the Brazilian context is highlighted [94,95]. Such studies must provide a comprehensive overview, including ecotoxicological assessments, and analyses of growth, reproduction and physiological responses [96].

Finally, it is important to emphasize that the collection areas were characterized by highly impacted urban environments, with a scarce diversity of aquatic macrophytes and a lake bed predominantly composed of fine sediments. For a more accurate comparison between sampling devices, it would be desirable to conduct collections in less impacted environments as well.

## 5. Conclusions

The marked presence of invasive species, exemplified by the unprecedented record of the species *C. largillierti*, represents an urgent challenge for biodiversity and aquatic ecosystems. Ongoing monitoring is vital, in order to understand and mitigate the impacts of these introductions on native communities.

Data analyses have indicated some variation in the collected species and sizes of mollusks, depending on the collection equipment, thus stressing the importance of specific sampling devices. The tools used have shown effectiveness in our survey, especially for *Biomphalaria*, thus proving its usefulness for monitoring in schistosomiasis endemic areas. This contributes not only to the understanding of population dynamics, but also to the development of targeted control strategies.

It should be pointed out that additional research is essential to further elucidate the role of parasites associated with the malacofauna and therefore contribute to the promotion of public health, as well as to connect basic research with environmental preservation.

The detailed characterization of sampling points and the adjacent environment, as well as data on water quality, emphasize environmental challenges, such as plastics, urban sewage and organic waste, which threaten the health of ecosystems and mollusk biodiversity in this region. Considering such factors in ecological studies is crucial, and, in our opinion, specific strategies to face such complex challenges should always include a focus on preservation.

The recognition of Lake Pampulha as part of the World Heritage Site emphasizes its cultural relevance, imposing conservation obligations that require the active participation of local authorities in the protection of this important legacy. Coordinated efforts are essential to ensure the integrity of the ecosystem and associated cultural heritage, placing emphasis on the intersection between environmental conservation and cultural value, in order to promote the sustainability of the region.

In summary, our findings highlight the interconnection between malacological dynamics, environmental conditions and cultural practices, and the necessity of integrative efforts in preserving ecosystems and ensuring long-term health.

**Supplementary Materials:** The following supporting information can be downloaded at: <https://www.mdpi.com/article/10.3390/d16040193/s1>, Table S1. Malacofauna of Lake Pampulha, Belo Horizonte, MG, Brazil: An integrative analysis based on literature data, scientific collections and an updated survey.

**Author Contributions:** All authors contributed to the study conception. Conceptualization, P.R.S.C. and S.M.G.; methodology, P.R.S.C., S.C.T., C.L.F.d.M., S.B.d.S., R.L.C. and S.M.G.; software, P.R.S.C.; validation, P.R.S.C., S.C.T., C.L.F.d.M., N.M.T.d.O., S.B.d.S., R.L.C. and S.M.G.; formal analysis, P.R.S.C., S.C.T., C.L.F.d.M., N.M.T.d.O., S.B.d.S., R.L.C. and S.M.G.; investigation, P.R.S.C. and

S.M.G.; field work, P.R.S.C. and S.M.G.; resources, P.R.S.C., S.C.T., C.L.F.d.M., N.M.T.d.O., S.B.d.S., R.L.C. and S.M.G.; data curation, P.R.S.C. and S.M.G.; writing—original draft preparation, P.R.S.C., S.C.T., C.L.F.d.M., N.M.T.d.O., S.B.d.S., R.L.C. and S.M.G.; writing—review and editing, P.R.S.C., S.C.T., C.L.F.d.M., N.M.T.d.O., S.B.d.S., R.L.C. and S.M.G.; visualization, P.R.S.C., S.C.T., C.L.F.d.M., N.M.T.d.O., S.B.d.S., R.L.C. and S.M.G.; supervision, S.M.G.; project administration, P.R.S.C. and S.M.G.; funding acquisition, P.R.S.C., S.C.T., C.L.F.d.M., S.B.d.S., R.L.C. and S.M.G. All authors have read and agreed to the published version of the manuscript.

**Funding:** P.R.S.C. received a grant from the “Coordination for the Improvement of Higher Education Personnel- Brazil” (CAPES)—Financial Code 001, and acknowledges the support from the Postgraduate Program in Parasitology at the Federal University of Minas Gerais for their financial assistance towards the publication of this research. The snail molecular biology studies were partially financed by the “National Council for Scientific and Technological Development” (CNPq # 308869/2017-6), from the René Rachou Institute—Fiocruz, and from UFMG.

**Data Availability Statement:** Data are contained within the article.

**Acknowledgments:** We would like to thank the company LOGNature® for providing the Shovels, especially Bárbara de Oliveira Sanches and Juliana Maria Dumont Kleinsorge. It is important to highlight that LOGNature® had no influence on the design or results of the present study. We also thank Lucas José de Almeida Lana for producing the Scoops, Frederico Dutra Kirst for the photographs of specimens of the *Biomphalaria* genus, Jeferson Kelvin Alves de Oliveira Silva for delivering the material for identification at Fiocruz/CMIOC and William Joseph Raworth for reviewing the manuscript. We also appreciate the support from the Postgraduate Program in Parasitology at the Federal University of Minas Gerais for funding the publication of this research. Furthermore, we would like to thank the Medical Malacology Collection (Fiocruz/CMM), the Mollusc Collection of the Oswaldo Cruz Institute (Fiocruz/CMIOC) and the Malacological Collection of the State University of Rio de Janeiro (UERJ) for providing the data used in this study and for depositing our material.

**Conflicts of Interest:** The authors declare no conflicts of interest.

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