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Seasonal and Spatial Distribution Patterns of Non-Native Fishes in Inland Waters of Guangxi

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Abstract: Fish invasions can damage the ecological environment of invaded areas, causing negative effects such as monotony of ecological types in invaded waters and endangerment of native species. It is important to monitor their presence and spread in invaded areas. This study aims to update the available data on non-native fish species in the inland waters of Guangxi and to determine their seasonal and spatial distribution in this region. Taking the inland waters of Guangxi as the study area, 34 sampling sites were set up in the major river systems, and systematic sampling was conducted in four seasons in 2023. The data showed that a total of 7690 non-native fish were collected from 23 species, belonging to 7 orders, 13 families, and 20 genera. Of the non-native fish species, 19 species were introduced for aquaculture purposes, and three species were introduced for ornamental purposes. The most non-native fish species were found in summer with 21 species, which were followed by 20 species in winter, 18 species in spring, and only 15 species in autumn. However, the distribution composition was similar in each season, with *Coptodon zillii* being dominant in each season, which was followed by *Oreochromis niloticus*. The Hongshuihe River had the highest number of non-native fish species with 16 species, followed by the Xunjiang River with 14 species, the Qianjiang River with 13 species, and the Npanjiang River had the lowest number of non-native fish species with 7 species. In addition, the Xunjiang River and the Qianjiang River showed significant separation in the PCoA results, and the overall test showed significant differences in non-native fish composition among the river system. The main reasons for the differences in their spatial distribution are geographical location and temperature. *O. niloticus* and *C. zillii* were found in all river systems and have become the main invasive non-native fish species in the inland waters of Guangxi.

Keywords: fish invasions; non-native fish species; seasonal; spatial distribution

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1. Introduction

Biological invasions, as a major factor affecting the structure and functioning of global ecosystems, are one of the three major problems facing the global environment along with climate change and the loss of biological habitats [1]. However, freshwater ecosystems are considered to be more vulnerable to biological invasions [2]. In freshwater ecosystems worldwide, fish is the most commonly introduced group [3]. Thousands of freshwater fish species have been introduced, either intentionally or unintentionally, into new environments around the world [4]. After successful invasion, they pose a major threat to the biodiversity and ecosystem function of aquatic ecosystems through predation [5], competition [6], habitat destruction [7], and disease transmission [8]. At the same time, it reduces the amount of fishing and the income of fishermen [9,10], resulting in a large number of economic losses [11,12].

The Guangxi Zhuang Autonomous Region (hereinafter referred to as Guangxi) located in southern China (104°26′–112°03′ E, 20°54′–26°23′ N) has typical Karst landforms and Danxia landforms. The rivers in the area belong to four basins (the Pearl River Basin, the Yangtze River Basin, the Red River Basin, and the rivers flowing into the Beibu Gulf) and span three climatic zones: the northern tropics, the southern tropics, and the middle tropics [13]. Guangxi is one of the world's biodiversity hotspots, one of the richest areas in terms of aquatic biodiversity, and an important production base for freshwater fisheries in China [14].

There are clear geographical and ecological orientations to the distribution of non-native fish species in China, and they are mainly distributed in southern China, especially in Guangxi, Guangdong, Hainan, and Fujian [15,16]. According to the research, at least 439 species of non-native freshwater fish species have been recorded in Chinese freshwaters, and southern China accounts for 81% of the total national aquaculture production of non-native fish [17]. Many non-native fish species have been introduced, farmed, sold, discarded, or escaped in the region [18]. The relatively warm climate and complex river network provide conditions for aquaculture production but also for the survival and spread of non-native fish species in natural waters [19]. Some non-native fish, such as *Tilapia* spp., *Cirrhinus cirrhosus*, *Clarias gariepinus*, *Cirrhinus mrigala*, and *Hypostomus* sp., have already established natural populations among the large number of fish introduced into Guangdong [20]. In addition, a study of the Pearl River basin showed that *Clarias gariepinus*, *Tilapias*, *Labeo rohita*, and *Cirrhinus mrigala* have become important exotic species for the local fishing industry [21]. *Oreochromis niloticus* has disrupted the functional patterns of fish communities, caused disruptions in the trophic structure of fish communities, and affected the trophic position and resource use of commercially harvested piscivorous fish in studies related to the rivers of Guangdong [22–24]. However, there are few studies on non-native fish in the inland waters of Guangxi.

Thorough research and monitoring, as well as comprehensive statistics on non-native fish, are essential for the control and management of the non-native fish. In this study, the species composition of non-native fish in 12 major river systems was recorded, and the seasonal and spatial distribution characteristics were investigated. Specifically, we aimed to (1) update the current status data of non-native fish species in the inland waters of Guangxi and discuss the introduction pathways of non-native fish; (2) determine the seasonal distribution patterns of non-native fish; (3) determine the spatial distribution patterns of non-native fish in the inland waters of Guangxi, while trying to confirm the differences among the 12 river systems and further discuss the factors. The results can update the status of non-native fish records in the inland waters of Guangxi, provide basic data for the in-depth study of non-native fish invasion, and provide evidence for the prevention and control of non-native fish.

2. Materials and Methods

2.1. Study Area

Located on the southern border of China, Guangxi has nearly 1000 rivers and abundant wild fish resources. The Karst landform is widespread with surface water seepage and a well-developed underground river system. Fish communities were surveyed in 2023 at 34 sites on rivers in the inland waters of Guangxi. The major rivers in the inland waters of Guangxi were systematically surveyed: Nanpanjiang River (NPJ), Hongshuihe River (HSH), Qianjiang River (QJ), Xunjiang River (XJ), Youjiang River (YJ), Zuojiang River (ZJ), Yongjiang River and Yujiang River (YYJ), Hejiang River (HJ), Guijiang River (GJ), Liujiang River (LJ), Nanliujiang River (NLJ), and other southern rivers flowing into the sea (OR) (Figure 1).

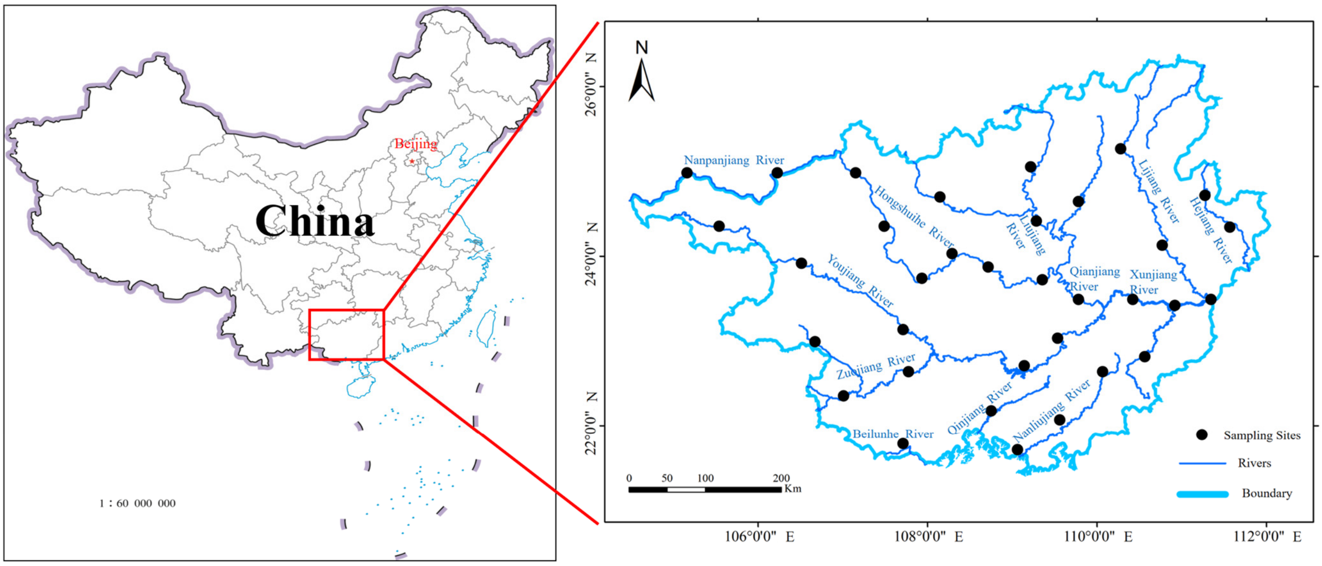

Figure 1. Map of sampling sites of non-native fish in inland waters of Guangxi.

2.2. Fish Sampling

Sampling was carried out in four seasons: February (spring), May–June (summer), August–September (autumn), and November–December (winter). Suitable nets were selected according to the characteristics of the different river habitats. Single-layer gillnets (20–50 m $\times$ 1–2 m, mesh size 1–6 cm) were used in shallow and fast-flowing river sections. In the slow-flowing river section, a bottom cage (0.3 m $\times$ 0.25 m, total length 8 m, mesh size 4 mm) was used as the main method with gillnets as an auxiliary method. At the same time, the markets in the vicinity of the sampling sites were surveyed, and the relevant fishing personnel were interviewed about the historical fishing status of non-native fish. To increase the comparability of the data, the same mesh size and fishing time were used for each sampling. Fish samples were identified according to the *Freshwater Fishes from Guangxi* [13], *Fishes in the Pearl River* [25], and *Manual of Identification of Common Exotic Aquatic Organisms in China* [26].

2.3. Data Analyses

The Bray–Curtis similarity measure was introduced to analyze the compositional similarity of non-native fishes in 12 river systems. Based on the sampling results, the proportions of each non-native fishes in the total catch of non-native fish were calculated, and a species-sample point matrix was generated [18]. After square root transformation, the similarity indices of the different streams were analyzed by clustering using Primer 5 version 5.2.8 [27].

In order to compare the structural differences of non-native fish in each river system, the non-native fish abundance was used as the raw data, which was transformed into a distance matrix by the Bray–Curtis similarity coefficient, and then the Principal Coordinates Analysis (PCoA) was performed. Next, the statistical significance of the overall differences and pairwise was assessed by Permutational Multivariate Analysis of Variance (PERMANOVA). The river system was a fixed factor, and the four seasons were levels, in which the number of randomizations is 999, and we chose the Bray–Curtis similarity to calculate pairwise distances. The whole test used the *vegan* package, and the *pairwiseAdonis* package was used for the pairwiseAdonis group test; all of the above analyses were performed using R 4.3.0 software.

3. Results

3.1. Species Composition

A total of 7690 individual alien fish were collected from 34 sampling sites in the inland waters of Guangxi during four seasons, and 23 non-native fish species were found: *Polyodon spathula*, *Neosalanx taihuensis*, *Prochilodus lineatus*, *Piaractus brachypomus*, *Labeo rohita*, *Cirrhinus mrigala*, *Megalobrama pellegrini*, *Cyprinus carpio* var. *haematopterus*, *Cyprinus carpio* var. *specularis*, *Leiocassis longirostris*, *Pterygoplichthys pardalis*, *Hypostomus* sp., *Ictalurus punctatus*, *Clarias lazera*, *Gambusia affinis*, *Oreochromis niloticus*, *Oreochromis mossambicus* × *O. niloticus*, *Sarotherodon galilaeus*, *Coptodon zillii*, *Parachromis managuensis*, *Lepomis macrochirus*, *Lepomis cyanellus*, and *Micropterus salmoides* (Table 1).

Table 1. List of non-native fish species found in inland waters of Guangxi.

Order	Family	Genus	Species	Common Name
Acipenseriformes	Polyodontidae	*Polyodon*	*Polyodon spathula*	Mississippi paddlefish
Osmeriformes	Salangidae	*Neosalanx*	*Neosalanx taihuensis*	Yangtze icefish
Characiformes	Prochilodontidae	*Prochilodus*	*Prochilodus lineatus*	Streaked prochilod
	Serrasalmidae	*Piaractu*	*Piaractus brachypomus*	Pirapitinga
Cypriniformes	Cyprinidae	*Labeo*	*Labeo rohita*	Roho labeo
		Cirrhinus	*Cirrhinus mrigala*	Mrigal carp
		Megalobrama	*Megalobrama pellegrini*	
		Cyprinus	*Cyprinus carpio* var. *haematopterus*	
			Cyprinus carpio var. *specularis*	Mirror carp
Siluriformes	Bagridae	*Pseudobagrus*	*Leiocassis longirostris*	Chinese longsnout catfish
	Loricariidae	*Pterygoplichthys*	*Hypostomus* sp.	Suckermouth catfish
	Ictaluridae	*Ictalurus*	*Ictalurus punctatus*	Channel catfish
	Clariidae	*Clarias*	*Clarias lazera*	North African catfish
Cyprinodontiformes	Poeciliidae	*Gambusia*	*Gambusia affinis*	Mosquitofish
Perciformes	Cichlidae	*Oreochromis*	*Oreochromis niloticus*	Nile tilapia
			Oreochromis mossambicus × *O. niloticus*	Red tilapia
		Sarotherodon	*Sarotherodon galilaeus*	Mango tilapia
		Coptodon	*Coptodon zillii*	Redbelly tilapia
		Coptodon	*Parachromis managuensis*	Jaguar guapote
	Eleotridae	*Oxyeleotris*	*Oxyeleotris marmorata*	Marble goby
	Centrarchidae	*Lepomis*	*Lepomis macrochirus*	Bluegill
			Lepomis cyanellus	Green sunfish
		Micropterus	*Micropterus salmoides*	Largemouth black bass

The 23 non-native fish species collected represent 7 orders, 13 families, and 20 genera (Table 1); the orders most represented were the Perciformes with nine species, Cypriniformes with five species, and Siluriformes with four species (Figure 2a); the families most represented were Cyprinidae with five species, Cichlidae with five species, and Centrarchidae with three species (Figure 2b). In terms of the places of native region, seven originated from Asia, seven from North America, five from Africa, and one from Europe (Figure 2c). In terms of the purpose of introduction, 19 species are used as cultured fish, three species are used as ornamental fish, and one is used for biological control (Figure 2d).

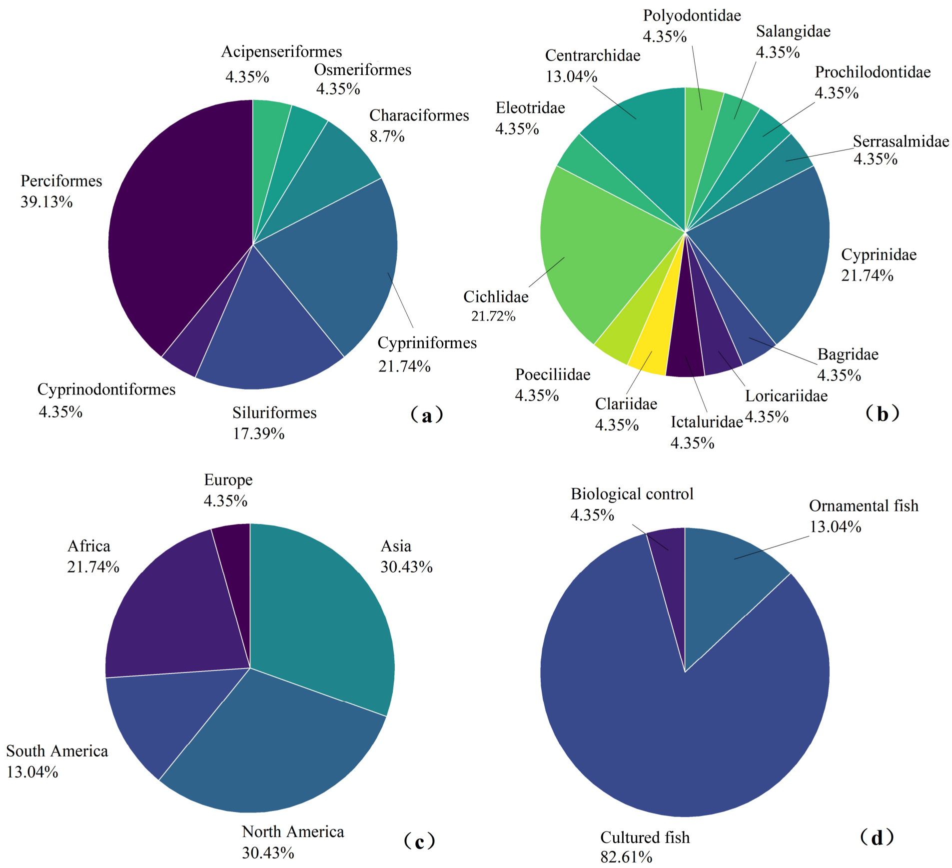

Figure 2. The non-native fish species distribution in the inland waters of Guangxi (**a**) by order, (**b**) by family, (**c**) by native region, and (**d**) by introduction vector.

3.2. Seasonal Distribution

The percentage of non-native fish abundance in each season showed that *C. zillii* was dominant in each season, which was followed by *O. niloticus* (Figure 3). Summer has the highest number of non-native fish species with 21 species, which was followed by 20 species in winter with 18 species in spring but only 15 species in autumn. In addition, *P. spathula* and *M. pellegrini* were only collected in summer (Table 2).

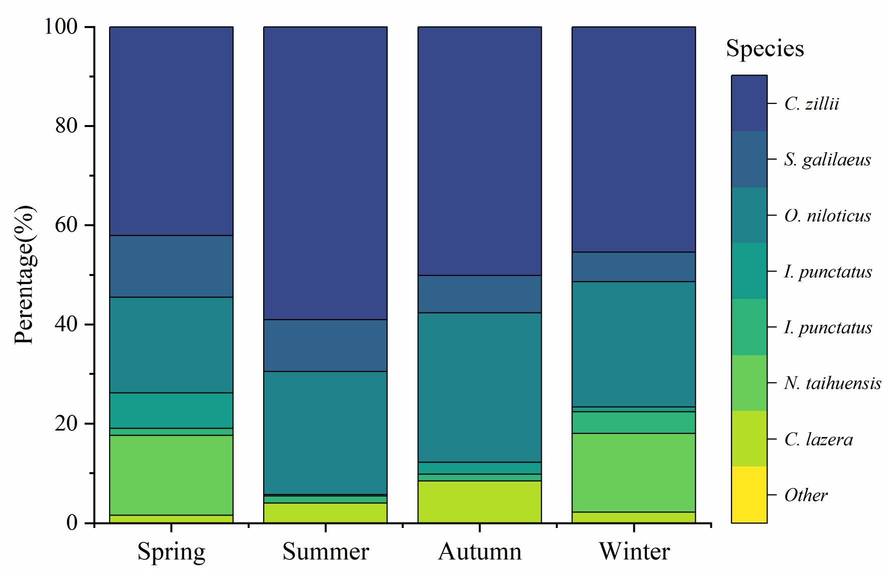

Figure 3. Percentage of non-native fish abundance in each season.

Table 2. Seasonal distribution of non-native fish in the inland waters of Guangxi.

Specie	Spring	Summer	Autumn	Winter
Polyodon spathula	0	1	0	0
Neosalanx taihuensis	1	0	0	1
Prochilodus lineatus	1	1	1	0
Piaractus brachypomus	1	1	0	1
Labeo rohita	1	1	1	1
Cirrhinus mrigala	1	1	1	1
Megalobrama pellegrini	0	1	0	0
Cyprinus carpio var. *haematopterus*	1	1	1	1
Cyprinus carpio var. *specularis*	0	1	0	1
Leiocassis longirostris	1	1	1	0
Hypostomus sp.	1	1	1	1
Ictalurus punctatus	1	1	1	1
Clarias lazera	1	1	1	1
Gambusia affinis	0	1	1	0
Oreochromis niloticus	1	1	1	1
Oreochromis mossambicus × *O. niloticus*	1	1	1	1
Sarotherodon galilaeus	1	1	1	1
Coptodon zillii	1	1	1	1
Parachromis managuensis	1	1	1	1
Oxyeleotris marmorata	1	0	0	1
Lepomis macrochirus	1	1	1	1
Lepomis cyanellus	0	1	0	1
Micropterus salmoides	0	1	0	1

Note: "0" denotes absence of the species, and "1" its presence.

3.3. Spatial Distribution

Cluster analysis based on the Bray–Curtis similarity measure showed that the twelve river systems could be divided into three assemblages: lower reaches of mainstems (QJ and XJ), northern tributaries (LJ, HJ, and GJ), and southern river systems (OR, YJ, HSH, YYJ, NPJ, ZJ, and NLJ) (Figure 4). The HSH has the highest number of non-native fish species with 16 species, which was followed by 14 species in the XJ and 13 species in the QJ. The NPJ has the lowest number of non-native fish species, with seven species, which was followed by eight species in the GJ and nine species each in the HJ and OR. *P. spathula* was found only in HSH, *M. pellegrini* only in XJ, *C. carpio* var. *specularis* only in LJ, and *L. cyanellus* only in GJ. In addition, *O. niloticus* and *C. zillii* were found in all river systems (Table 3). In terms of the number of non-native fish, *O. niloticus* and *C. zillii* already occupy most of the reaches; *L. rohita* was more common in XJ, and *C. lazera* is more common in QJ (Figure 5).

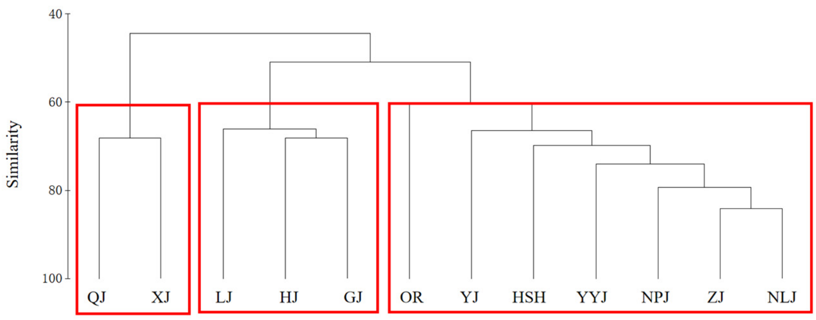

Figure 4. Cluster result based on the Bray–Curtis similarity measure.

Table 3. Spatial distribution of non-native fish in the inland waters of Guangxi: Nanpanjiang River (NPJ), Hongshuihe River (HSH), Qianjiang River (QJ), Xunjiang River (XJ), Youjiang River (YJ), Zuojiang River (ZJ), Yongjiang River and Yujiang River (YYJ), Hejiang River (HJ), Guijiang River (GJ), Liujiang River (LJ), Nanliujiang River (NLJ), other southern rivers flowing to the sea (OR).

Specie	NPJ	HSH	QJ	XJ	YJ	ZJ	YYJ	HJ	GJ	LJ	NLJ	OR
Polyodon spathula	0	1	0	0	0	0	0	0	0	0	0	0
Neosalanx taihuensis	0	1	1	1	0	0	0	1	1	0	0	0
Prochilodus lineatus	0	1	1	1	0	0	1	0	0	0	0	0
Piaractus brachypomus	0	1	0	0	0	1	0	0	0	1	1	1
Labeo rohita	0	1	1	1	1	1	0	1	0	1	1	0
Cirrhinus mrigala	0	1	1	1	0	1	1	1	0	1	1	1
Megalobrama pellegrini	0	0	0	1	0	0	0	0	0	0	0	0
Cyprinus carpio var. *haematopterus*	0	0	0	0	1	0	0	0	0	0	1	1
Cyprinus carpio var. *specularis*	0	0	0	0	0	0	0	0	0	1	0	0
Leiocassis longirostris	1	1	1	1	0	0	0	0	0	0	0	0
Hypostomus sp.	1	1	1	1	1	1	1	0	1	0	1	1
Ictalurus punctatus	1	1	1	1	1	1	1	0	0	1	1	0
Clarias lazera	1	1	1	1	1	1	1	1	1	1	1	1
Gambusia affinis	0	1	1	1	0	0	0	0	0	0	0	0
Oreochromis niloticus	0	1	1	1	0	0	1	1	0	1	1	0
Oreochromis mossambicus × *O.niloticus*	1	1	1	1	1	1	1	1	1	1	1	1
Sarotherodon galilaeus	1	1	1	1	1	1	1	0	0	0	1	1
Coptodon zillii	1	1	1	1	1	1	1	1	1	1	1	1
Parachromis managuensis	0	1	0	0	1	0	1	1	0	0	0	0
Oxyeleotris marmorata	0	0	0	0	1	1	0	0	0	1	0	1
Lepomis macrochirus	0	0	0	0	1	0	0	0	1	1	1	0
Lepomis cyanellus	0	0	0	0	0	0	0	1	1	1	0	0
Micropterus salmoides	0	0	0	0	0	0	0	0	1	0	0	0

Note: "0" denotes absence of the species, and "1" denotes its presence.

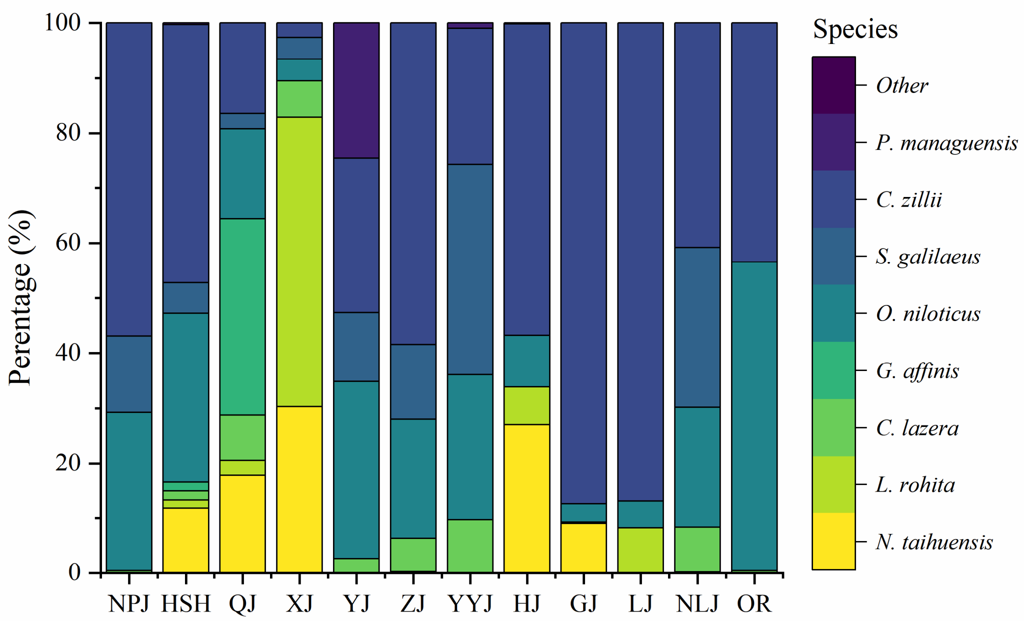

Figure 5. Percentage of non-native fish abundance in each river systems.

The principal coordinate analysis (PCoA) of non-native fish in each river system showed that Pco1 and Pco2 accounted for 32.45% and 20.16% of the overall variability, respectively (Figure 6). XJ and QJ are separated from other river systems across the first principal coordinate, and the remaining river systems overlap; the river systems tend to be discrete across the second principal coordinate, while some of them are clearly separated, such as ZJ, YYJ, NPL, and LJ. The PERMANOVA of the overall differences at all river system grouping levels showed that the non-native fish structure was significant ($p < 0.001$) (Table S1). The PERMANOVA of paired comparisons showed that the results were similar to the results of PCoA, and some river systems showed significant differences ($p < 0.05$) (Table S2).

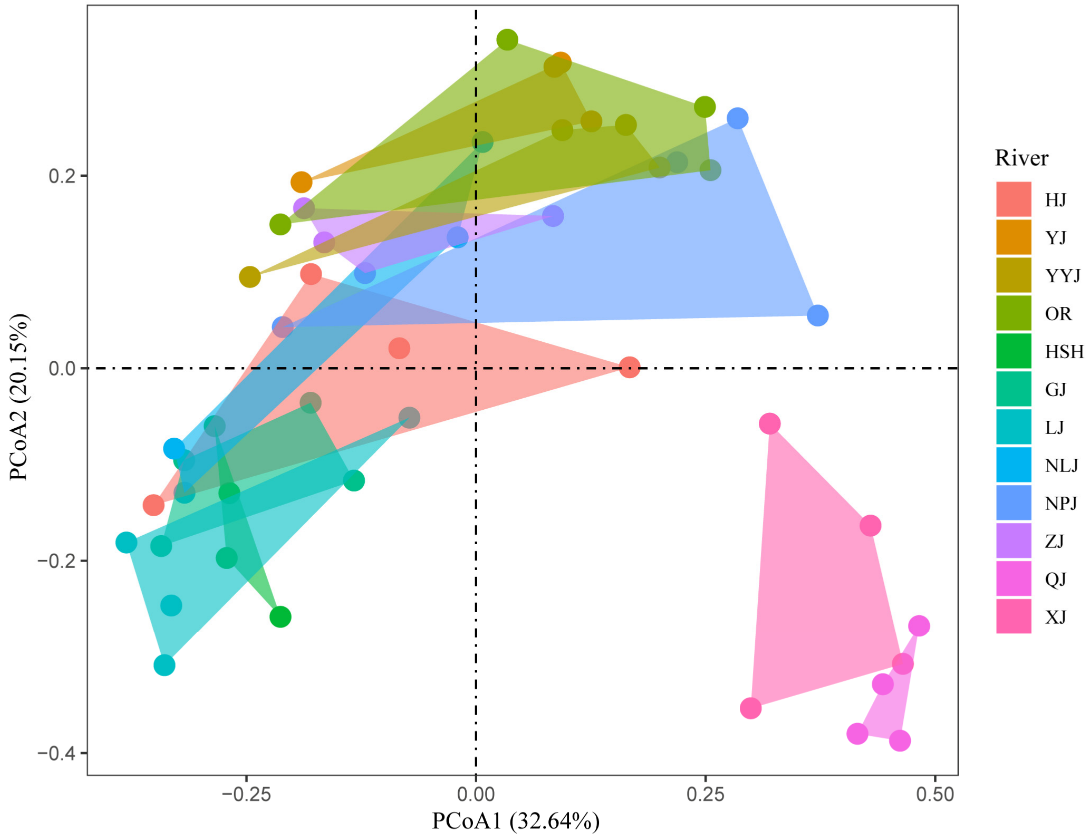

Figure 6. Principal coordinates analysis of non-native fish in each river system in inland waters of Guangxi.

4. Discussion

Non-native fish are widespread in the inland waters of Guangxi and have been recorded in all 12 water systems (Table 1). Although not all of the 23 species can be classified as invaders, all the river systems were occupied by multiple non-native species. In terms of large-scale research on non-native fish in the inland waters of Guangxi, He Jiayang et al. found that a total of 18 non-native fish species were recorded in the inland waters of Guangxi based on historical survey data and literature collection up to 6 November 2021 [28]. Shuai Fangmin et al. collected 11 non-native fish species through sampling from 2014 to 2016. In this study, 23 non-native fish species belonging to 7 orders, 13 families, and 20 genera were collected from the inland waters of Guangxi [29]. Compared with previous studies, the number of collected non-native fish species was increased because: (1) in this study, more sampling sites and a wide sampling area were arranged, with the increase in sampling times and the expansion of sampling range, fish species diversity would increase [30]; (2) there is a direct correlation between the amount of aquaculture and the amount of non-native fish species [31], aquaculture is an important medium for non-native species to invade, and at the same time, it is possible to escape non-native species with a high invasive potential [32]. So as the market demand for aquatic food has increased in recent years, the introduction of more and more aquaculture species and the expansion of aquaculture areas have increased the opportunities for non-native fish invasions.

According to the 2023 China Fishery Statistical Yearbook, the output of freshwater aquaculture in Guangxi increased by 44,232 tons in 2022 compared with 2021. And the freshwater aquaculture area increased by 3165 hectares in 2022 compared with 2021. Among the 23 non-native fish species collected in this study, 19 are cultured fish, accounting for 82.61% of the total number of non-native fish species, including *P. spathula*, *N. taihuensis*, *P. lineatus*, *P. brachypomus*, *L. rohita*, *C. mrigala*, *M. pellegrini*, *C. carpio specularis*, *L. longirostris*, *I. puncta-*

tus, *C. lazera*, *M. salmoides*, *O. niloticus*, *T. galilaea*, *C. zillii*, *P. managuensis*, *O. mossambicus* × *O. niloticus*, *O. marmorata*, and *L. macrochirus*.

The breeding mode of ornamental fish is similar to that of aquatic fish, but the breeding equipment of ornamental fish lack measures to prevent escape, which leads to the water change in the fish pond, and the ornamental fish will enter the natural water with the drainage ditch when the flood occurs [33]. In addition, fish release and abandonment are also important ways for fish to enter natural waters [34]. The *L. cyanellus* collected in this study were mainly distributed in GJ (Guijiang River and Lijiang River), but whether they formed viable populations needs further investigation. At the same time, there were no records of collection in the inland waters of Guangxi. The *L. cyanellus* was introduced to China in 1999 as an ornamental and swimming fish. It is now one of the most important species of freshwater fish to have been introduced into China, and it has been cultivated in Guangdong Province and even throughout the country [35]. *Hypostomus* sp. was introduced into Guangdong Province in the 1990s as an ornamental fish. Due to it being highly adaptable to the environment [36], it is now widely distributed in various rivers in Guangdong Province and has been shown to have established self-sustaining populations [37].

In terms of seasonal distribution, the seasons had similar species composition, with *C. zillii* being dominant in each season, which is followed by *O. niloticus*. In terms of spatial distribution, the 12 river systems could be divided into three assemblages according to the cluster result based on the Bray–Curtis similarity measure: lower reaches of mainstems (QJ and XJ), northern tributaries (LJ, HJ, and GJ), and southern river systems (OR, YJ, HSH, YYJ, NPJ, ZJ, and NLJ) (Figure 4). The PCoA results were similar, QJ and XJ were clearly distinguished, and the structure of the non-native fish composition was not consistent and there were significant differences ($p < 0.001$). QJ and XJ are located in the easternmost part of the inland waters of Guangxi downstream of the main stream. The two rivers are closely connected with QJ being the upper reaches of XJ. In addition, they have similar climatic conditions and geographical features with an average precipitation of 1418 mm and an average temperature of 21.6 °C [38]. The reason that LJ, HJ, and GJ form northern tributaries together may be that the three river systems are all located in the northern part of Guangxi, and the minimum temperature in winter can reach below 0 °C, which is different from other river systems. *O. niloticus* and *C. zillii* were found in all river systems (Table 3). Tilapia is one of the most common invaders in the world and has become a dominant species in many rivers in southern China [39]. Tilapia is the common name for the African cichlids that include the mouthbrooding *Sarotherodon*, *Oreochromis*, and substrate-spawning tilapia [40,41]. Tilapia (*Oreochromis mossambicus*) was first introduced to southern China in 1957; and later, more than seven species of tilapia were introduced into China [42]. Five species of tilapia have been identified in southern China rivers, including *C. zillii*, *O. aureus*, *S. galilaeus*, *O. niloticus*, and a hybrid of *O. mossambicus* and *O. niloticus* [43]. Among them, *C. zillii* and *O. niloticus* are the most common, both of which were introduced into the mainland China in 1978 and have a wide distribution.

5. Conclusions

Our study revealed that a total of 23 non-native species were identified, most of which were introduced for aquaculture purposes. Compared with historical studies, the number of non-native fish species increased. Non-native fish were abundant in summer and winter, and the composition was similar in each season. The Hongshuihe River had the highest number of non-native fish species with 16 species, and the Npanjiang River had the lowest number of non-native fish species with seven species, the spatial distribution showing differences in species composition. *O. niloticus* and *C. zillii* were found in all river systems, and they have become the main invasive non-native fish species in the inland waters of Guangxi. Therefore, it is necessary to strictly control the artificial introduction and breeding and make good efforts to control the release and escape of non-native and ornamental fish. There are already a lot of non-native fish in the main river system of the Hongshuihe River, Xunjiang River, and Qiangjiang River, so prevention and control measures such as early warning and the monitoring of non-native fish should be strengthened.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/w16213062/s1, Table S1: The PERMANOVA result of the overall difference at all river system grouping levels; Table S2: The PERMANOVA result of the pairs of river systems.

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