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Research Article

Gut Content Analysis of *Oreochromis niloticus and Chanda nama* of Begnas Lake, Kaski, Nepal

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Abstract

Gut content analysis provides insights into fish species' feeding preferences, tropical interaction, and geographical and seasonal variations in their diets. Guts of *Oreochromis niloticus* and *Chanda nama* were analyzed and index of preponderance (IP) was calculated. IP depicts that in *O. niloticus*, phytoplankton (61.92%) was the most commonly consumed food followed by zooplankton (24.39%), debris, sand (12.57%), nematodes (0.7%) and hatchling (0.42%), In *C. nama*, micro-crustaceans (48.82%) contributed the most for total volume of food items, followed by scales (37.76%), insects (13.53%), and phytoplankton (0.29%). Begnas lake supports 28 phytoplankton and 12 zooplankton species out of which 24 phytoplankton, seven zooplankton species and seven phytoplankton and four zooplankton species were found in *O. niloticus* gut and *C. nama* gut respectively. *O. niloticus*'s diverse feeding habits and *C. nama*'s lepidophagous behavior have caused challenges for native fish, which may be reason for decline in native fish population.

Keywords: Gut analysis; *Oreochromis niloticus*; *Chanda nama*; Index of preponderance; Phytoplankton; Zooplankton.

Introduction

Gut content analysis is the scientific examination of gut content in order to identify and quantify the sorts of food items ingested (Amundsen and Sánchez-Hernández, 2019). Researchers are increasingly using stomach content analysis to determine feeding habits and diets of fish species (Baker *et al*., 2014; Bernal, 2015; Manko, 2016; Zacharia *et al*., 2004). Gut content analysis provides information on fish health conditions, eating habits, tropical interaction and competition, fish species-specific feed preferences, and geographical and seasonal variations (Manko, 2016; Mishra, 2020; Martins *et al*., 2021; Igejongbo and Esther, 2022; Pirroni *et al*., 2021; Zacharia, 2017). In fisheries, it helps to ascertain the feeding patterns and diets of fish species as well as provide information about the impact of introduced or invasive species to native fish species for fisheries management (Fritts and Pearsons, 2004; Hanson and Chouinard, 2002; Manko, 2016).

Begnas lake is the second largest lake after Phewa situated in south-east of the Pokhara valley. It is located at an altitude of 650m and lake water covers an area of $17.96 \times 10^6 m^3$ (Khadka and Ramanathan 2013). This lake, together with eight other lakes on its periphery (in Pokhara and Lekhnath), was designated as Nepal's 10th Ramsar site on 2nd February 2016 (DNPWC, 2016). Begnas Lake has been found to be home to twenty-six different fish species, out of which twenty fish species are native and six are exotic (Husen *et al*., 2019).

The exotic fish species O. *niloticus* and *Chanda nama* have been reported from catches of Begnas Lake. Husen *et al*., (2019) claimed that *Chanda nama* made its first appearance in the Begnas catches in July of 2018 while *O. niloticus* appeared in lakes of Pokhara in catches during 2003 (Nepal, 2008). Many authors have been reported the gut content analysis of *O. niloticus* (Abdelghany 2020; Engdaw 2023; Genanaw *et al*., 2021; Handago *et al*. 2024; Oso *et al*., 2006; Shalloof and Khalifa 2009; Wagaw *et al*., 2021; Temesgen *et al*., 2022) and *Chanda nama* (Bhuvaneswari and Serfoji 2018; Grubh and Winemiller 2004; Khoso *et al*., 2018) from different region of world. This is the first study on the gut content analysis of two exotic fish species of Begnas Lake to ascertain the feeding habits of these exotic species and also to explore what could be the effects of these two-exotic species on native fish species.

Materials And Method

Description of Study Area

This study was carried in Begnas Lake of Pokhara valley (Fig. 1). This lake has geographical grid coordinates 28.1621-28.2167°N latitude and 84.0814-84.1332°E longitude (MoFE, 2018). It is fed by a perennial stream with a catchment area of 19 km^2 and an average depth of 6.6 m (Husen *et al*., 2019).

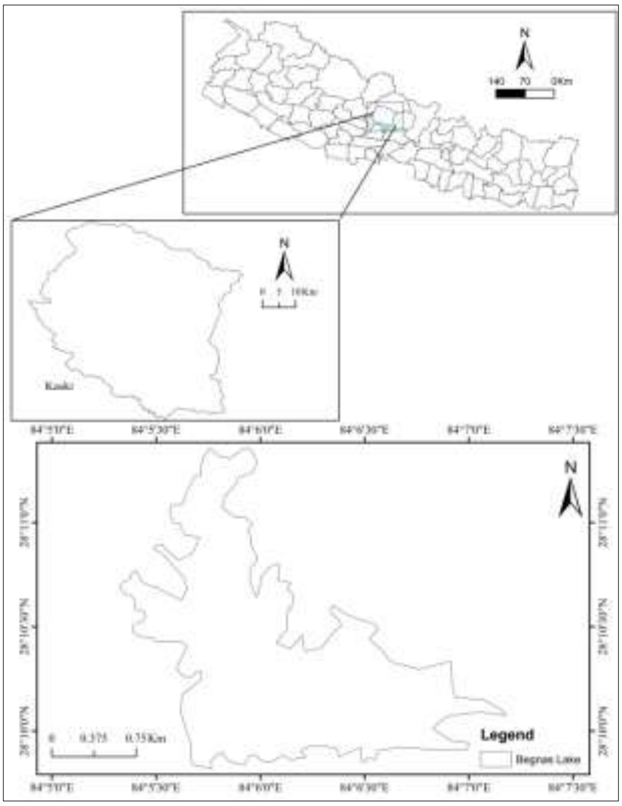

Fig. 1: Map of Nepal showing Study Area

Sampling of Planktons

For the collections of water samples to know the plankton's richness of Begnas Lake, pre-determined ten sampling locations were used. For zooplankton, water samples were collected by vertically hauling a plankton net (<75 micrometers) from the bottom to the surface at a constant speed of 0.5 meter per second. Concentrated samples were preserved in 5% neutral buffered formalin as methods described by Gifford and Caron, (2000) and finally identified and counted under microscope as methods of Johnson and Allen, (2012). One-liter water sample was collected from 10 locations and was preserved with Lugol's iodine solution for phytoplankton as methods of Kumari *et al*., (2018). Likewise, phytoplankton was identified by using book of Bellinger and Siegee, (2010).

Fish Gut Collection and Grading

A total of 36 fish of each species (*O. niloticus* and *C. nama*) were collected (December 2023 to March 2024) from the fish landing site of Begnas Lake. Total length and total weight were measured by use of digital Vernier caliper (Mituotyo, Japan) and electronic balance (Libror ED 3200D, SHIMADZU corporation Ltd.). Longitudinal cut was made on the ventral side of the fish from just behind the isthmus of the gills reaching to the anal fin by using scissors and scalpel. The gut was carefully removed and graded as empty, full, ¾ full, ¼ full by visual observation as methods of Sivadas and Bhaskaran, (2009). The stomachs extracted from the live fish were preserved immediately in 5% formaldehyde solution for later analysis.

Gut Content Analysis

To remove excess formalin, gut was placed out of formalin for 5-10 minutes. The stomach or gut segment was opened with the help of fine scissors or a scalpel. Larger prey items were directly taken out and other gut contents were dissolved in distilled water. In case of *O. niloticus* gut contents were dissolved in 60 ml of water whereas those of *C. nama* were dissolved in 10ml and observed under microscope. The gut content was analyzed using frequency of occurrence and volumetric (point method) based on methods of Windell and Bowen (1978).

Frequency of occurrence: The relative importance of food items to the diet of different fish species were analyzed using the frequency of occurrence.

The frequency of occurrence was computed as:

$$\%\ \text{Frequency of occurence of food items(Oi)} = \frac{\text{Number of fish containing food items } (Ji)}{\text{Number of fish with food in their stomach } (Pi)} * 100$$

Volumetric method: It can be calculated by using formula:

$$\%\ \text{of Volume of food item i (Vi)} = \frac{\text{Point allocated to food item i } (Vi)}{\text{Total Point allocated to subsample}} * 100$$

Index of preponderance: Index of preponderance provide clear and measurable basis of grading food items and calculated based on Natarajan and Jhingra (1961).

$$Index\ of\ Prepoderance(IP) = \frac{\%O(\text{Frequency of occurrence})x\%V(\text{\% of volumetric contribution})}{\sum(\%O + \%V)} x100$$

Length weight relation: It was calculated by using formula W = aL^b (Pauly, 1983). Where W is weight, L is the standard body length (in cm), a is the intercept and b is the slope.

Impacts of Exotic Fish on Native Fish

The purposive survey was conducted during the entire period of study to know the impacts of exotic fish on native fish species. The 40 Jalari fishermen of Begnas Lake were interviewed with structured questionnaire to share experience of impacts of exotic fish species on native fish species.

Statistical Analysis

The data were summarized with the help of Microsoft excel and analyzed with SPSS version 25.

Results

Length - Weight Relationship

The results of 36 fish of *O. niloticus* and *C. nama* assessed for the length weight relationship is presented in the Fig. 2 and Fig. 3. The average size of *O. niloticus* and *C. nama* was 16.61±5.16cm (226.07±186.72gram) and 3.96±1.05cm (3.48±1.57gram) respectively. The length weight relationship obtained for *Oreochromis niloticus* and *Chanda nama* was $y = 0.0348x^{3.0283}$ and $y = 0.2955x^{1.7571}$ respectively. The R^2 value for both fish species was positive, which indicate positive correlation between length and weight. The b value was 3.0283 in case of *Oreochromis niloticus* whereas in case of *Chanda nama*, it was 1.7571 as shown in Fig. 2 and Fig. 3.

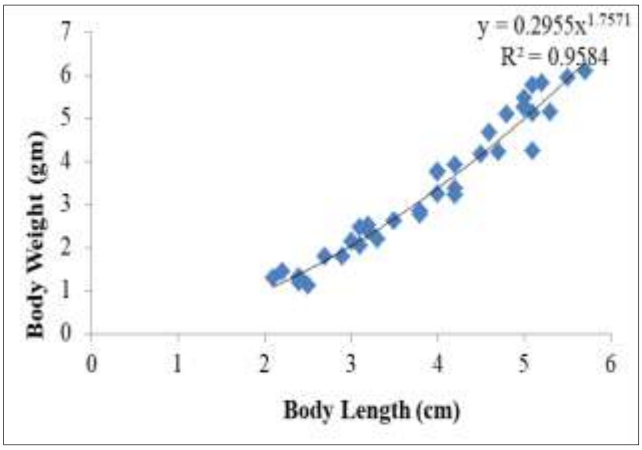

Fig. 2: Length-Weight relationship of Nile tilapia (*O. niloticus*)

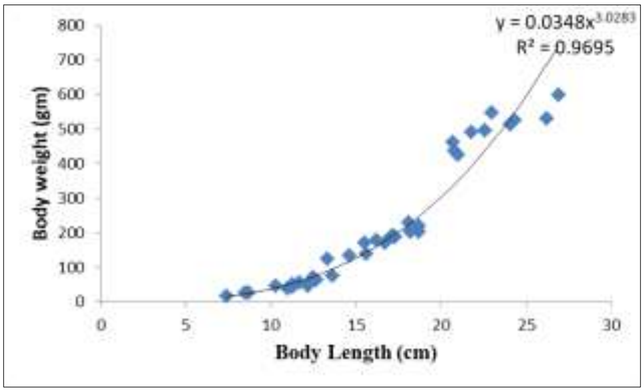

Fig. 3: Length-Weight relationship of Elongated glass perchlet (*C. nama*)

Fullness Index

The results of feeding intensity of *O. niloticus* and *C. nama* are recorded as full, ¾ full, ½ full, ¼ full and empty on the basis of food contents of stomach of fishes are presented in the Fig. 4. The results of fullness index from the total observed fish sample showed that, ¾ full of *O. niloticus* percent was found high while in the *C. nama* ½ full percent was found high. The fishes having empty stomach among the studied sample was low in the both two fish species during the studied period.

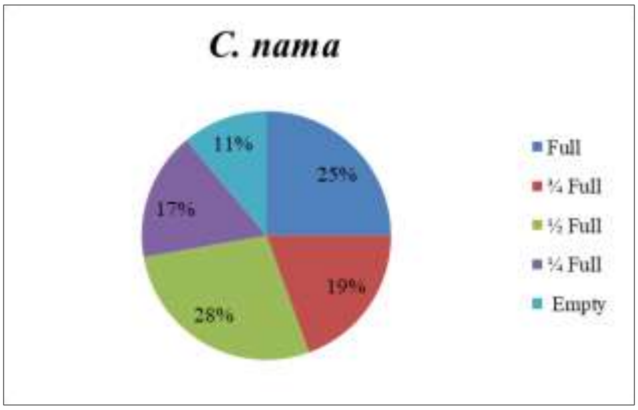

Fig. 4: Fullness index of *O. niloticus*

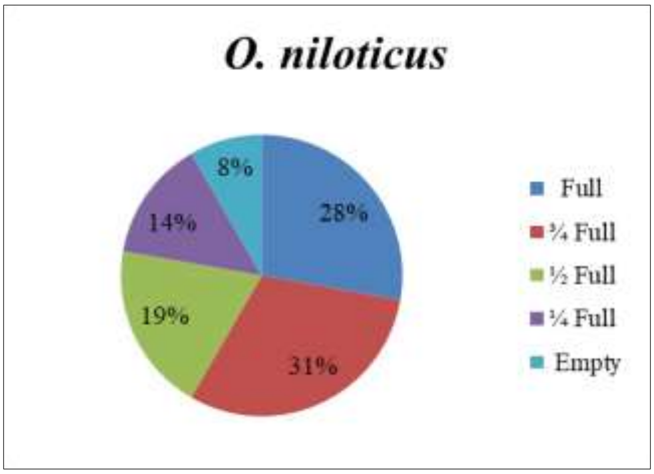

Fig. 5: Fullness index of *C. nama*

Plankton Diversity and Gut Content of Fish

The plankton present in the Begnas lake and food component present in the gut contents of different fish species are presented in the Table 1. A total of twenty-eight different genera of phytoplankton were recorded in the Begnas Lake, among which twenty-three were found in the stomach of *O. niloticus* and eight in the in the stomach of *C. nama*. Likewise, a total of twelve genera of zooplankton were recorded in the Begnas lake, among which in the gut contents of *O. niloticus* and *C. nama* seven and four genera recorded respectively.

Table 1: Plankton diversity of Begnas Lake and Gut content analysis of *O. niloticus* and *C. nama*

Food items	Class/Order	Genus	*Begnas Lake*	*O. niloticus*	*C. nama*
Phytoplankton	**Chlorophyceae**	*Ankistrodesmus*	+	+	+
		Staurastrum	+	+	+
		Chlorella	+	+	-
		Scendesmus	+	+	+
		Spirogyra	+	+	+
		Closterium	+	+	-
		Tetraedron	+	+	+
		Kirchneriella	+	+	-
		Crucigenia	+	+	-
		Selenastrum	+	+	-
		Coelastrum	+	+	-
		Chlamydomonas	+	-	-
		Pediastrum	+	+	+
	Dinophyceae	*Ceratium*	+	-	-
	Euglenophyceae	*Euglena*	+	+	+
		Phacus	+	+	+
	Bacillariophyceae	*Navicula*	+	+	-
		Melosira	+	-	-
		Pinnularia	+	+	-
		Nitzschia	+	+	-
		Surirella	+	+	-
		Fragillaria	+	-	-
		Tabellaria	+	+	-
		Cymbella	+	+	-
	Cyanophyceae	*Anaebena*	+	+	-
		Merismopedia	+	+	-
		Arthrospira	+	+	-
	Chrysophyceae	*Dinobryon*	+	-	-
Zooplankton	**Cladocera**	*Ceriodaphnia*	+	+	-
		Bosmina	+	+	+
		Alona	+	+	-
	Copepoda	*Diaptomus*	+	+	-
		Cyclops	+	+	+
	Rotifera	*Keratella*	+	+	+
		Polyartha	+	-	-
		Lecane	+	-	-
		Trichocerca	+	-	-
		Asplanchna	+	-	-
		Filinia	+	-	-
		Brachionus	+	+	+
Nematodes				+	-
Hatchling, Fish parts				+	-
Miscellaneous		Sand particles		+	-
		Detritus		+	-
Insects				-	+
Fish scales				-	+

[(+) = Present, (-) = Absent]

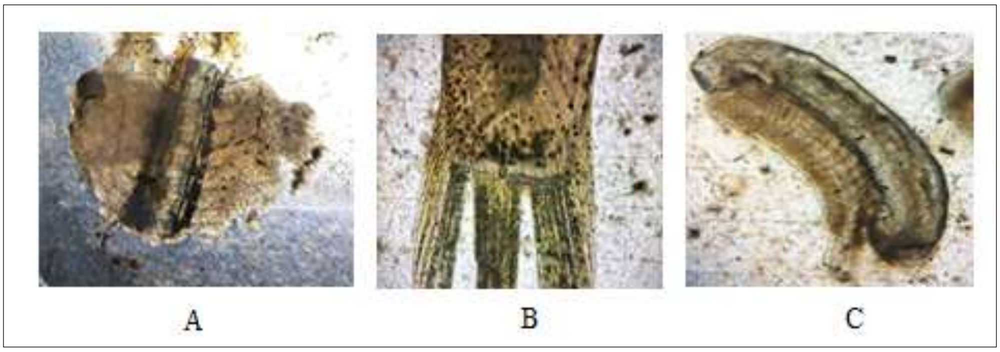

Fig. 6: (A, B) fish parts and (C) hatchling found in gut of *O. niloticus*

Fig. 7: Fish scale found in gut of *C. nama*

Table 2: %Oi, %Vi, and IP of food items of Nile tilapia (*Oreochromis niloticus*)

S.N.	Food items	% composition of food items by		Vi*Oi	% IP
		%Oi	%Vi		
1.	**Phytoplankton**	**48.09**	**55.28**	**12910.09**	**61.92**
	Chlorophyceae	12.38	10.44	627.35	10.22
	Bacillariophyceae	16.69	25.58	2073.12	33.77
	Cyanophyceae	12.76	16.33	1011.73	16.48
	Euglenophyceae	6.26	2.93	88.93	1.45
2.	**Zooplankton**	**32.49**	**26.87**	**1496.89**	**24.39**
	Cladocera	15.23	14.24	1052.79	17.15
	Copepoda	5.04	2.73	66.83	1.09
	Rotifera	8.55	8.47	351.76	5.73
	Ostracoda	3.67	1.43	25.51	0.42
3.	**Nematodes**	**5.71**	**1.55**	**42.84**	**0.7**
4.	**Miscellaneous (sand and debris)**	**11.14**	**14.26**	**771.72**	**12.57**
5.	**Fish part**	**2.57**	**2.06**	**25.75**	**0.42**

%Oi= frequency of occurrence of food item i, %Vi= Percentage of volume of food item i, IP= Index of preponderance

Gut Content Analysis

Nile tilapia (O. niloticus):

This study reveals that among different food items, phytoplankton was most frequently occurred food items followed by the zooplankton, sand and debris, nematodes and fish parts. The % IP illustrate that phytoplankton (61.92%) was dominant food item followed by zooplankton, miscellaneous bodies. Nematodes (0.7%) and fish parts (0.42%) were found to be consumed rarely (Table 2).

Elongated Glass Perchlet (Chanda nama):

The study reveals that micro crustaceans, scales and insects were found in 59.72%, 37.15% and 31.6% of gut and contributed to 48.42%, 37.76%, and 13.53% of total food items. Similarly, the only food of plant origin found in glass fish was phytoplankton which contributed to 0.29% of total diet and was found in 8.68% of gut. (Table 3).

Impacts Of Nile Tilapia and Elongated Glass Perchlet on Native Fish Species

The 95 % of respondents of survey of Jalari fishermen of Begnas Lake were stressed that Nile tilapia and glass perchlet impacts was negative on native fish species. They had agreed that after the introduction of these fish species in the Begnas Lake cage aquaculture have been declined. The native fish species such as *Barilius* sp. and *Putius* sp., catches have been declined in the Begnas Lake, they added. These respondents had added that silver carp and Bighead carp have been found to scale less sometimes in the catches of Begnas Lake. It is mainly due to facultative scale feeder nature of *C. nama* and leads to diseases and fish mortality. These fish species had showed food competitions with native fish species. As the source of water for FRS, Begnas ponds is from Begnas Lake, the Elongated glass perchlet and Nile tilapia have also affected the Fishery Research Station (FRS), Begnas research and production program specially in the nursing of hatchling to fry in the nursery ponds.

Table 3: %Oi, %Vi, and IP of food items of Elongated glass perchlet (*Chanda nama*)

S.N.	Food items	% composition of food items		Vi*Oi	% IP
		%Oi	%Vi		
1.	**Phytoplankton**	**8.68**	**1.95**	**9.34**	**0.29**
	Chlorophyceae	5.6	1.41	7.83255	0.24
	Euglenophyceae	2.78	0.54	1.506794	0.05
2.	**Microcrustaceans**	**59.72**	**51.49**	**1560.35**	**48.42**
	Cladocera	39.93	26.88	1073.29	33.31
	Copepoda	19.79	24.61	487.095	15.11
3.	**Insects**	**31.6**	**13.8**	**436.0187**	**13.53**
4.	**Scales**	**37.15**	**32.755**	**1216.663**	**37.76**

%Oi= frequency of occurrence of food item i, %Vi= Percentage of volume of food item i, IP= Index of preponderance

Discussion

The results of this investigation suggest that the *O. niloticus* exhibited isometric growth pattern since the value of growth coefficient (b) was 3.0283. Similar result was obtained for cage farmed tilapia and was demonstrated by the length-weight equation's b value of 3.064 (Silva *et al*., 2015). According to Wagaw *et al*. (2022a), Nile tilapia shows positive allometric growth, where the b value was 3.19. Likewise, the finding of Ubong *et al*. (2023) shows negative allometric growth with b value less than 3. In this study, growth coefficient of *C. nama* was 1.7571 indicating negative allometric growth. Our result was supported by finding of Bhuvaneswari and Serfoji (2018) and Sheikh *et al*., (2017), which also show negative allometric growth in *Chanda nama*. Whereas, Sangma *et al*. (2019), reported that the b value was 3.08 indicating isometric growth in *C. nama*.

The stomach content analysis of *O. niloticus* of present study in Begnas Lake indicate that *O. niloticus* feeds on varieties of feed items including food from plant origin like phytoplankton as well as food of animal origin like zooplankton, nematodes and fishes suggesting it to be omnivore in nature. Previous studies suggested *O. niloticus* to be omnivore (Oso *et al*., 2006; Wagaw *et al*., 2022b). Present study shows that based on frequency of occurrence and volumetric contribution, phytoplankton was most dominant food, which is similar to the finding of Tadesse (1999) and Wagaw *et al*., (2021). The predominant phytoplankton present in gut of *O. niloticus* was of class Bacillariophyceae followed by Cyanophyceae, Chlorophyceae and Euglenophyceae in the present study. However, Mohamed *et al*. (2019) reported Cyanophyceae and Chlorophyceae were predominant over other phytoplankton. Nile tilapia has a versatile feeding behavior, characterized by generalist and opportunistic omnivorous feeding behavior (Canonico *et al*., 2005). This study revealed that the second most important food item was found to be zooplankton is supported by Abdulhakim *et al*., (2015), explained about the presence of zooplankton and insects in *O. niloticus* diets in various lakes and reservoirs.

The present study has shown gut of *C. nama* contained range of food items which includes zooplankton, insects, fish scales as well as phytoplankton and majority of food items of animal origin suggest it to be carnivore in nature. This present finding is comparable to the findings of previous studies such as Khoso *et al*. (2018), Bhuvaneswari and Serfoji (2018) and Grubh and Winemiller (2004).

Exotic fish species are one of the greatest threats to freshwater ecosystems globally. The present survey on the impacts of exotic fish species on native fish species has indicated towards the negative impacts on the native fish species. It is also supported by the results of present study on the gut contents analysis of *O. niloticus and C, nama*. The previous findings have also indicated that the native fish contribution declined and Nile tilapia increased in the catches from Begnas Lake (Husen *et al*., 2014; Husen *et al*., 2016). Nile tilapia (*Oreochromis niloticus*) contributed the biggest proportions (51.9 %) to the total exotic fish of Begnas lake in the FY 2016/17 (Husen *et al*., 2019). Previous findings also demonstrated that Nile tilapia could compete with native species for food and habitats, which reduces the native fish populations due to competitive displacement (Canonico *et al*., 2005; Martin *et al*., 2010). Further, Nile tilapia also cause the extinction of native fish species by preying on eggs, fry and small fish of other species (Russell *et al*., 2012). Additionally, one of the studies provide clear evidence that invasive Nile tilapia could destroy recipient ecosystem stability by disrupting the trophic structure and food chains of native communities (Shuai and Li, 2022). Likewise, *Chanda nama* is facultative scale feeding (*Lepidophagy*) and its feeds on fish scales, microcrustacea and aquatic insects with juvenile diets containing larger fractions of invertebrates, and cyprinids were found clearly anxious by repeated attacks(Grubh and Winemiller, 2004).Therefore, to control further expansion of Nile tilapia and elongated glass perchlet into other natural lakes, reservoirs, and rivers of Nepal, native fish conservation policy, laws, and protocols should be rigorously enforced (Husen *et al*., 2016).

Conclusion

The substantial populations of *O. niloticus* and *C. nama* presence in Begnas Lake has led to long-term competition with native fish species for food, space as well as their aggressive behavior and carnivore nature causes to decline of their population in Begnas Lake and it is proven by the present study of gut content analysis of these two fish species. It is urgent need to make awareness for restriction of invasive fish for further invasion in other natural resources. The government should take suitable action to implement the existing laws at local government level.

Authors' Contribution

M.A. Husen designed the research plan; A. Basnet, R. Shahi & N. Khanal performed experimental works, collected, and analysed the data; A. Basnet, R. Shahi, N. Khanal prepared the manuscript. M.A. Husen critical revised and finalized the manuscript. Final form of manuscript was approved by all authors.

Conflicts of Interest

The authors declare there is no conflict of interest regarding this study and publication of this paper.

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