

**PREVALENCE OF *ARGULUS* SPECIES IN CULTIVABLE FISH  
IN CHHAPIYA VILLAGE OF RUPANDEHI DISTRICT, NEPAL**



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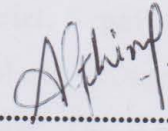
**Kirtipur, Kathmandu**

**Nepal**

**October, 2022**

## DECLARATION

I, hereby declare that this thesis entitled “**Prevalence of *Argulus* species in cultivable fish in Chhapiya village of Rupandehi district, Nepal**” is a thesis report based on primary work and all the sources of information used are duly acknowledged. This work has not been submitted to any other university for any academic award.



.....  
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### RECOMMENDATIONS

This is to recommend that the thesis entitled “**Prevalence of *Argulus* species in cultivable fish in Chhapiya village of Rupandehi district, Nepal**” has been carried out by Mrs. Ajita Ghimire for the partial fulfillment of Master’s Degree of Science in Zoology with Special Paper “Fish Biology and Aquaculture”. This is her original work and has been carried out under our supervision. To the best of our knowledge, this work has not been submitted for any other degree in any institutions.

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### LETTER OF APPROVAL

On the recommendation of supervisor, Prof. Dr. Kumar Sapkota this thesis submitted by Mrs. Ajita Ghimire entitled “**Prevalence of *Argulus* species in cultivable fish in Chhapiya village of Rupandehi district, Nepal**” is approved for the examination in partial fulfilment of the requirements for Master’s degree of science in Zoology with special paper Fish Biology and Aquaculture.

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### CERTIFICATE OF ACCEPTANCE

This thesis work submitted by Mrs. Ajita Ghimire entitled “**Prevalence of *Argulus* species in cultivable fish in Chhapiya village of Rupandehi district, Nepal**” has been accepted as a partial fulfillment for the requirements Master’s Degree of science in Zoology with Special Paper “**Fish Biology and Aquaculture**”.

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## ABBREVIATION

Cm	=	Centimeter
DAP	=	Di- ammonium Phosphate
DO	=	Dissolved Oxygen
EUS	=	Epizootic Ulcerative Syndrome
Ha.	=	Hectare
M	=	Meter
Mg / l	=	Milligram per litre
pH	=	Potential of Hydrogen
Sq. km	=	Square kilometer
TU	=	Tribhuvan University
US	=	United States
Yr	=	Year

## ABSTRACT

*Argulus* also known as fish lice is a hazardous ectoparasite infecting many freshwater fish species. This research was conducted to determine the prevalence, abundance, and intensity of *Argulus* in five different carp species (*Labeo rohita*, *Cirrhinus mrigala*, *Hypophthalmichthys molitrix*, *Cyprinus carpio*, *Ctenopharyngodon idella*) from the different ponds of Siyari rural municipality. The data was collected from Chhapiya village which is highly known by the name of the fish village and the fish pond was selected randomly. A total of 250 fishes were examined from different ponds for species-wise, sex-wise, and length-wise study of ectoparasite infestation. Species-wise maximum prevalence (50%), abundance (1.4) and intensity (2.8) respectively were found in *L. rohita* followed by *C. mrigala* [prevalence 40%, abundance (1) and intensity (2.5)], *H. molitrix* [prevalence 25%, abundance (0.5) and intensity (2.0)], *C. carpio* [prevalence 15%, abundance (0.25) and intensity (1.667)] and lastly *C. idella* [prevalence 10%, abundance (0.15) and intensity (1.5)]. Sex-wise prevalence, abundance, and intensity of *Argulus* species were found higher in female carp than in male carp. Similarly, length-wise highest prevalence, abundance, and intensity of *Argulus* species occurred in a small sized group (10-20 cm) of carp than in the largest group (30-40 cm). Lack of awareness about preventive measures and management practices are some of the factors responsible for *Argulus* infestation which has badly impacted farmers. To eradicate these parasites, efficient curative and effective preventive measures should be applied.

**Keywords:** *Abundance, Argulus, Ecto-parasite, Infestation, Intensity*

# CHAPTER-I

## 1. INTRODUCTION

### 1.1 Background

Aquaculture is one of the most economically important applied strategies all over the globe. Fishes have great economic, nutritional, and recreational benefits to humans on a global scale. Like humans and other animals, fish suffer from diseases and parasites. Along with the rise of aquaculture, there has been also a rise in fish disease which has somehow declined the fish market. The prevalence of fish disease has a negative impact on fish production. There is a deterioration in food value which may even result in their mortality. Several pieces of literature are available on fish mortality and morbidity due to bacteria, parasites, fungus, nutritional deficiencies, metabolic disorders, and faulty environmental conditions like oxygen deficiency, high or very-low water temperature, chemical pollutants, overcrowding, mal-nutrition, algal blooms (Mukherjee, 2002). It is estimated that the annual loss in production due to diseases ranges from 25 to 30% (Anon, 1996). The intensification and expansion of fish culture are facing a severe threat from pathogens as they are the major cause of huge mortalities and poor growth affecting the production and market value.

Parasites in fish are common natural occurrences. Most parasites are opportunistic and may be omnipresent time in a culture unit or on the fish hosts in low numbers and only cause disease when fish is stressed. Parasites multiply rapidly under favorable conditions, distressing fish health, causing high mortality, and inflicting economic losses (Snieszko, 1974; Klesius and Rogers, 1995). Unmanaged fish farming and poor water quality are the major cause of parasitic disease. Parasites and diseases constitute important factors in regulating the population in the natural environment and fish abundance (Tripathi, 1969). Parasitic infection and disease outbreaks occupy peak levels to prove themselves as the major constraints to aquaculture production that may cause heavy losses to aqua-pond farmers. Such loss affects the livelihood of people involved in the fishery and the community in which they occur through reduced food availability and loss of income as well as other related social consequences (Singh et al., 2001).

Parasitic infections often indicate the quality of water since parasites generally increase in abundance and diversity in more polluted water (Poulin, 1999; Avenant-Oldewage, 2002). Fish parasitic diseases are a result of an imbalance in the interaction between fish and parasite factors and aquatic environment conditions. The occurrence and magnitude of these diseases are closely related to sanitary conditions in water (Hossain et al., 2007). The characteristic of water quality is therefore the most important key to successful fish production. Any characteristic of water that affects the survival, reproduction, growth, or management of fish is a water quality variable (Noga, 2010). Water pollution influences the pathogenic potential of ectoparasites and endoparasites (Khan and Thulin, 1991). In fish, the same poor water quality increases disease susceptibility (Biswas and Pramanik, 2016), by lowering the defense and immunogenic status of the fish (Noor El-Deen et al., 2015). Environmental factors such as water temperature, dissolved oxygen, salinity, hydrogen ion concentration, and eutrophication have a positive influence on the occurrence of parasitic populations and communities (Ali et al., 2004; Lagrue et al., 2011; Zargar et al., 2012; Ojwala et al., 2018).

Along with carp, all the freshwater species found more or less suffer from different types of disease while 15% - 20% are associated with parasite infestation. Indian major carps are highly susceptible to disease in comparison to Chinese and European carp (Lilley et al., 1992). The infection of parasites is greatly influenced by season, i.e., fish suffer more from parasitic disease in the winter season than in the summer season and very less in the rainy season. Parasites interfere with the nutrition of hosts, disrupting the metabolism and secretory function of the alimentary canal and damaging the nervous system (Farhaduzzaman et al., 2010). All these effects may reduce the normal growth of fish and finally induce host mortality. The parasitic infection tends to decrease the growth rate resulting in the stunting of fish (Cross, 1993). Intense parasitic infection can cause ulceration and upset the normal course of reproduction (Rahaman and Jahan, 2002). The most commonly reported parasitic disease in fish culture are Argulidae (Carp lice), *Dactylogymus* (Gill fluke), Ichthyophthiriasis (White spot), Trichodiniasis, Gyrodactylosis (Skin fluke) and Myxosporidiasis (Mishra et al., 2014).

The heavily infected fish shows physical injury and abnormal swimming behavior and blood ooze out from the skin thereby increasing susceptibility to secondary infection,

resulting in the nutritive devaluation of fish and subsequent economic losses (Onyedineke et al., 2010). Parasites are responsible for the direct mortality of farmed fish stock, retarding growth rate, feed conversion ratios, treatment cost incurred, and rejection of product during processing. *Argulus* (fish louse) and *Lernaea* (anchor worm) are the parasites on the external surface. However, some parasites are restricted to the internal organs causing serious disease and harm to host fish.

The success of fish culture depends on proper management which relates to understanding fish biology and the aquatic environment in which they survive. Fish health management begins with prevention rather than treatment which can be accomplished through good water quality management, nutrition, and sanitation. It is impossible to prevent an outbreak of opportunistic disease. Parasitic invasion of fish may cause high mortality, weight loss, and reduced fecundity (Grabda, 1991). For the prevention of diseases, it is important to study the nature of the fish disease. Several parasites including protozoan, crustaceans, copepods, trematodes, and nematodes can cause infection to fish.

Crustacean copepods *Argulus* and *Lernaea* are the most widely spread skin parasite. Those closely associated with fish can cause disease problems. Parasitic crustaceans are among the most serious gill and skin parasites of fish worldwide. They may be found attached to the external surfaces of both marine and freshwater fish. Almost 150 *Argulus* species are known from marine, brackish, and freshwater habitats (Kabata, 1985). They damage and kill small fingerlings as well as large fish, especially in crowded populations. Parasites from the genus *Argulus* are regarded as the most widespread and problematic parasites in freshwater fish culture (Kearn, 2004; Walker et al., 2008).

Argulids commonly known as “fish lice” are obligatory parasites causing problems in fisheries and aquaculture worldwide. *Argulus* species are known from marine, brackish, and freshwater habitats (Kabata, 1985). It has become a major problem in the carp culture industry causing mortality, morbidity, and growth loss (Rahman, 1996 and Ahmed, 2004). They are large parasites visible with naked eyes, dorsoventrally flattened and covered dorsally by a round or horseshoe-shaped carapace, ventrally positioned head appendages developed for attachment, and four thoracic segments each bearing a pair of bifid swimming legs. They have four stage life cycles (egg, nauplius,

meta-nauplius, and adult) and require a fish host for their survival. They complete their life cycle within 30-100 days depending on the water temperature. They are transmitted via the free swimming meta-nauplius stage (Shimura, 1981). Lice survive only a few days without host fish and juveniles for even shorter period (Kollatsch, 1959). The lice attach themselves to the skin of the host with their strong suckers and then feed on its host's blood or mucus by using a modified disk that possesses piercing and sucking mouthparts. During feeding, argulids inject cytolytic enzymes and then draw off the blood (Shimura and Inoue, 1984). In the early stage of infection, fish shows jumping behavior and a reduction in feeding. Further infection leads to bacterial and fungal infection and fish exhibits shoaling behavior. The pathogenic city of *Argulus* can kill larval cells by injecting a cytoplasmic toxin into the host. It is reported that this parasite creates ulceration and hemorrhagic changes to the host skin causing secondary infection by other parasites, bacteria, fungi, and viruses (Hoffman, 1977).

Fish lice, *Argulus*, are a hazardous ectoparasite for farmers. Argulosis has become a major threat that causes growth loss underweight, immature mating, and mortality of fish. It causes intense irritation and tissue damage. In addition to physical injury, affected fish are subjected to severe stress, which often leads to parasitic infestation from bacteria, fungi, and viruses. Opportunistic bacteria such as *Aeromonas* or *Pseudomonas* can sometimes infect damaged areas leading to skin ulceration (Abele, 1982). These all activities decrease the market value of fish and loss in production.

This research is intended to investigate the prevalence intensity and abundance of *Argulus* species in the aquaculture pond of the Rupandehi district. It is located in the southern plain area of Nepal and has great potential for fish farming. Because of parasites and disease, rural farmers are not getting satisfactory results. So there is a great loss in fish farming. Various research has been carried out related to disease and parasites and the economic loss imposed by their infestation. But in the context of Nepal very little research has been carried out related to this topic. This research will account for the prevalence, abundance, and intensity of *Argulus* species in different 5 species of carps *Labeo rohita*, *Cirrhinus mrigala*, *Hypophthalmichthys molitrix*, *Cyprinus carpio*, and *Ctenopharyngodon idella* based on species, sex, and length. This research will help the rural farmer to know the present status of Argulosis.

## 1.2 Objectives

**General:** The general objective of this research was to determine the prevalence of *Argulus* species in cultivable fish in Chhapiya village of Rupandehi district, Nepal.

**Specific:** The specific objectives are:

- i. To identify the parasite from the fish body.
- ii. To investigate the species-wise, sex-wise, and length-wise prevalence, abundance, and intensity of *Argulus* species.

## 1.3 Rationale/ Justification

Fisheries in the Rupandehi district is a progressively growing sector with varied resources. People at the primary level are directly or indirectly dependent on this sector for their livelihood. It has a total water surface area of 751 ha and 3581 numbers of ponds. It has a total fish yield of 4500 metric tons/ha/yr. Fisheries Centre Bhairahawa and Mandal hatchery Patthardada have a hatchery system for the production of hatchlings, fry, fingerlings, and adults of different Indian and Chinese carp. Similarly, the people of Siyari rural municipality are mostly engaged in fish farming. This sector has shown significant growth from traditional cultural practices to commercial methods of culture, enhancing fish production. There is no satisfactory result in terms of carp health and growth in aquaculture ponds which may be due to infestation of parasites and diseases. Very little work was carried out on this type of study and no attempt was introduced to eradicate such emerging problems of aquaculture. Therefore the present study has been done to gather information regarding parasitic diseases like Argulosis. This work also highlights the study of water quality (pH, DO, and temperature) and the prevalence, abundance, and intensity of the parasite. Hence, this will be helpful for any further studies, research work on fish health management, and developmental plans on fishery sector upliftment.

#### **1.4 Limitation**

- i. The findings of this research may have limitations to generalize as the study was carried out in Chhapiya village only.
- ii. This study doesn't include the study of other fish parasites and diseases such as fungal and bacterial.

## CHAPTER - II

### 2. LITERATURE REVIEW

Most parasite species rarely cause problems in the natural environment but in aquaculture, parasites often cause a serious outbreak of disease. These diseases in fish are caused by protozoans, crustaceans, copepods, helminths, and fungi. They play an important role in determining the sustainability and economic viability of aquaculture. The ectoparasites form the largest group of pathogenic organisms.

Parasites cause deterioration in the food value of fish and may even result in their mortality. The losses due to Epizootic Ulcerative Syndrome in several Asian countries before 1990 exceeded US 10 million (Chainabut, 1994).

Banerjee and Bandyopadhyay (2010) researched on the prevalence of ectoparasites in carp fingerlings in two districts of west Bengal and reported the highest prevalence (51.66%) of infection in ciliophorans and the lowest in crustaceans (17.5%) where highest ectoparasite prevalence (36.85%) was in the winter season and lowest (9.16%) in the rainy season. They concluded that the biological factors of the host as well as water quality are responsible for infestations.

Alam et al. (2012) researched on the intensity of parasitic infestation in silver carp (*Hypophthalmichthys molitrix*) and revealed nine different parasite species (*Trichodina pediculatus*, *Dactylogyrus vastator*, *Ichthyophthirius multifiliis*, *Gyrodactylus elegans*, *Learnea* species, *Apiosoma* species, *Myxobolus rohita*, *Camallanus ophiocephali*, and *Pallisentis ophiocephali*) were recorded from the gill, skin, stomach, and intestine of the host fish. The highest level of infection was found for the host skin, while lower levels were observed for the host gill, stomach, and intestine. The result also revealed that the intensity of parasite infection in different organs of *H. molitrix* varied with the season.

Otachi et al. (2013) worked on parasites of commercially important fish from lake Naivasha, Rift valley, Kenya in which ten taxa of parasites were recovered from *C. carpio* dominated by monogenean *Dactylogyrus minutes*, thirteen taxa of parasites were from *Oreochromis leucostictus* dominated by monogeneans *Cichlidogyrus* species and *Tilapia zillii* harbored nine taxa of parasites dominated by digenean *Tylodelphys* species whereas *Barbus paludinosus* harbored eleven taxa of parasites which were dominated

by an unidentified monogenean of genus *Dactylogyrus*. Similarly, they found that *C. carpio* had the lowest helminth species diversity.

Akter et al. (2007) studied five exotic carp species viz. *Hypophthalmichthys molitrix*, *Cyprinus carpio* var. *specularis*, *Cyprinus idellus*, *Cyprinus carpio* var. *communis* and *Puntius gonionotus* of Bangladesh for parasitic diseases and reported 3 protozoans (*Trichodina* species, *Ichthyophthirius* species and *Chilodonella* species), 2 monogeneans (*Gyrodactylus* species, *Dactylogyrus* species), 2 trematode (*Gorotrema* species, *Metadena* species), 4 Cestoda (*Rhopalothyra* species, *Marsipometra* species, *Lytocestus* species, and *Senga* species), 2 Nematoda (*Camallanus* species, *Procamallanus* species), 3 crustacean (*Argulus* species, *Alitropus* species, and *Lamproglana* species) and one insects (Dipteran larvae) parasitic species. These entire parasites were isolated from the body slime, gills, and intestine of the infected fish.

Alom et al. (2019) conducted research on the status, occurrence intensity, and impact of Argulosis in different brood stock ponds and revealed that heavy infestation and intensity of *Argulus* affect fish health and cause secondary infection as well as mortality due to low growth, survival, and production of carp in captivity. They reported that the prevalence and intensity of infestation were higher in large-sized females than in smaller ones. Sex-wise infestation and intensity of *Argulus* species were higher in female carp than in males in all ponds and length-wise highest prevalence and intensity occurred in large-sized groups of carp.

Hasan and Ahmed (2002) worked on some issues of finfish hatcheries and nurseries in some selected areas of Bangladesh where they found that 69% of hatcheries and nurseries were affected by diseases though diseases were less prevalent in hatcheries than in nurseries. A white spot, tail and fin rot, EUS, sudden spawn mortality, gill rot, dropsy, and malnutrition were major diseases in nurseries, while the major diseases in hatcheries were sudden spawn mortality and fish lice.

Waruiru et al. (2020) carried out the research on prevalence, intensity, and influence of water quality on parasites of farmed fish in Kirinyaga County, Kenya, where they isolated 8 fish parasite species (*Diplostomum* species, *Clinostomum cutaneum*, *Gyrodactylus* species, *Dactylogyrus* species, *Acanthocephalus* species, *Piscicola* species, and *Pseudophyllidian cestodes*) in which most prevalent parasitic genera were *Diplostomum* and *Acanthocephalus* each at 6.5% with a mean intensity of 32.7 and 4.4,

respectively. The highest mean intensity was observed in *Neascus* species at 190 with an abundance range of 4-596 parasites. The majority of the parasites were recorded from the skin, while the rest were from the gills, intestine/gut, eyes, fins, and muscles. Parasitic prevalence was positively correlated with ammonia-free nitrogen, nitrates, and phosphates.

Yamin (2007) reported several diseases in brood fish and spawn in different hatcheries of Mymensingh (Jessore and Bogra district of Bangladesh) in which the most prevalent disease of brood fish was Argulosis (35%), followed by EUS (30%), gill rot (12%), dropsy (9%) and nutritional diseases (9%). The average prevalence of diseases in brood fish was about 15-20% which varied from hatchery to hatchery. The major disease problem of fish spawn as reported by the hatchery owner was a fungal infection in fertilized eggs, white spots inside the yolk sac, loss of slime, spinal deformities, enlarged head and stomach, blindness, and sudden spawn mortality due to unknown reasons.

Faruk et al. (2004) worked on the status of fish disease and health management practices in Rural freshwater aquaculture of Bangladesh in which they revealed a prevalence of fish disease varied with the district and size of farms. Small ponds suffer high disease prevalence followed by medium and large farms where the most prevalent disease was tail and fin rot (20.5%) followed by EUS (18.9%), nutritional disease (15.3%), red spot (13.0%), and gill rot (12.3%). Other diseases such as Argulosis, dropsy, and white spot were reported lower by farmers. They concluded that the knowledge of farmers on basic health management was very poor and 83.8% of farmers used treatment measures in response to particular disease problems and also identified some fish health management problems in rural aquaculture.

Saha and Bandyopadhyay (2015) carried out research in which they isolated and identified 3 species of *Argulus* namely *A. coregoni*, *A. japonicus*, and *A. foliaceus* from the skin or gills of the freshwater fish species. The crustacean parasites were prevalent during the pre-monsoon season but their intensity was very less during other seasons and they claimed that this was the first report of infection with 3 species of *Argulus* of Red can Oranda Goldfish in India.

Aalberg et al. (2016) experimented Common carp (*Cyprinus carpio*), Pike perch (*Sander lucioperca*), and Brook trout (*Salvelinus fontinalis*) in which they identified

*Argulus* species as *A. foliaceus* and *A. japonicus*. Common carp were identified as *A. japonicus* and Pike-perch encountered both *A. japonicus* and *A. foliaceus*. Fish lice from Brook trout were identified as *A. foliaceus*. They revealed that the prevalence of *Argulus* infection recorded from carp varied among sampled localities ranging from 6.3% to 38.5%. Pike-perch prevalence was found very high ranging from 80% to 100% whereas the prevalence of *A. foliaceus* in Brook trout was 100% with infection intensities ranging from 25-30 and the mean intensity of infection was 30 specimens per fish and the abundance was found to be 30.

Sahoo et al. (2012) studied the mixed infection of *Argulus japonicus* and *Argulus siamensis* and estimated the economic loss of US\$ 1428 per hectare per year was incurred due to the infestation of *Argulus japonicus* and *Argulus siamensis*.

Das et al. (2016) experimented to investigate the infection of *Argulus* of juvenile carps of the Mymensingh region. The *Argulus* were very common in the body surface, the base of the pelvic, pectoral, anal, caudal fin, muscle, and mouth cavity of all fishes. They revealed the risk of infestation by *Argulus* in carps significantly increased when the water quality parameters were maintained at the optimum level and prevalence (%) and mean abundance was found to be species-specific and also varied with seasons and management systems. *Labeo rohita* was more susceptible than *Labeo catla* and *Cirrhinus mrigala* by *Argulus*.

## **CHAPTER-III**

### **3. MATERIALS AND METHOD**

#### **3.1 Study Area**

##### **3.1.1 Site Selection**

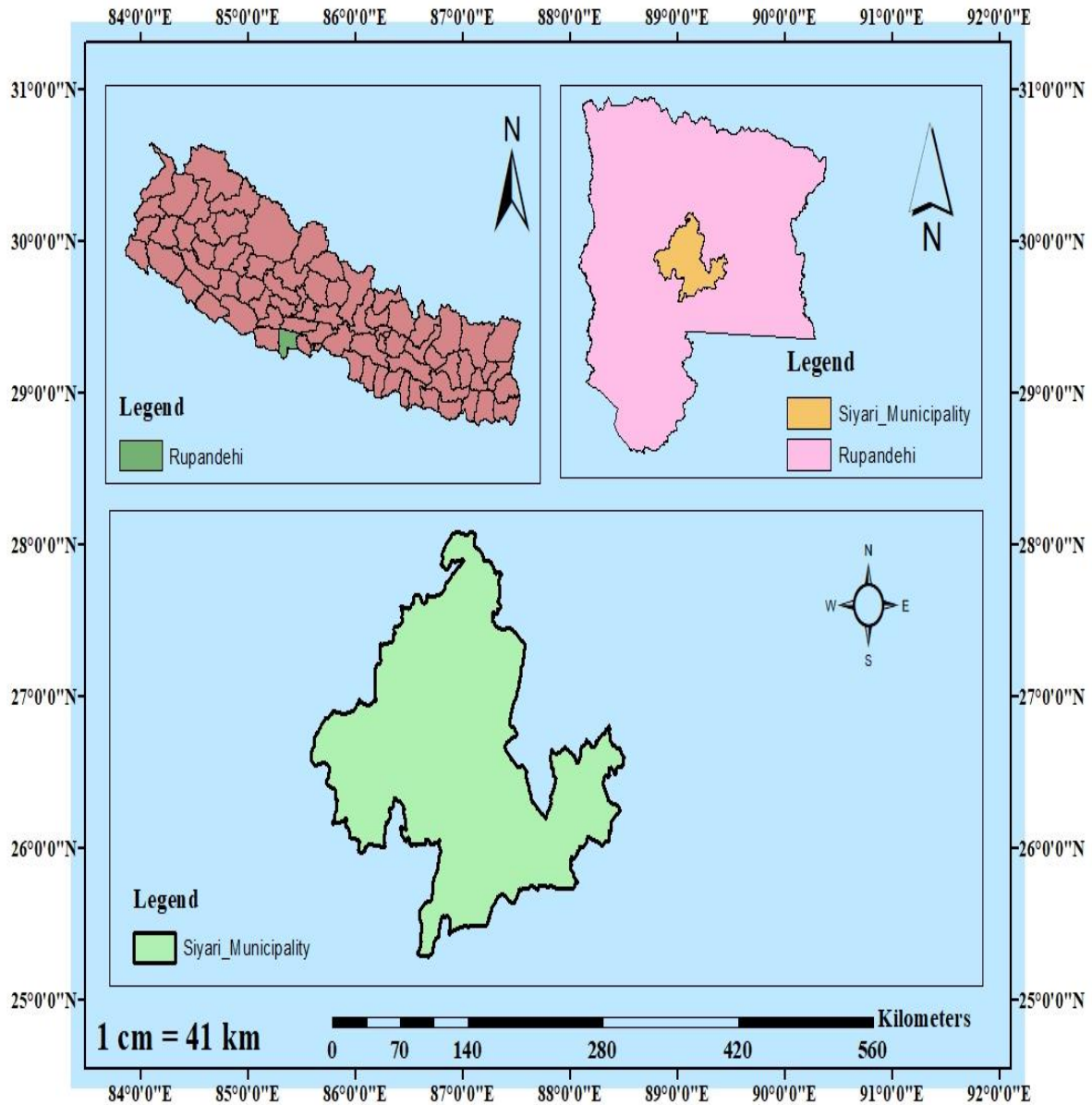
The study was carried out in the Siyari rural municipality of Rupandehi district, Lumbini province located in the south western part of Nepal. Siyari rural municipality lies 19.3 km east of Butwal which occupies a 66.17 sq. km area of the Rupandehi district. This area is considered a suitable area for fish culture. Most people in this area were found to have the aquaculture practice as their means of livelihood. So, this site was taken as my study area.

##### **3.2 General Description of Rupandehi District**

Rupandehi district is one of the seventy-seven districts of Nepal which lies in Lumbini Province. It lies in the southern and western parts of Nepal. On the East, it shares a border with Nawalparasi District, on the West with Kapilvastu District, on the North with Palpa District, and the South with India. The elevation of the district lies between 100m to 1229m from sea level. The total area of the district is 1,360 km<sup>2</sup> with 16.1% in Churia Range and the rest in the Terai region. The district headquarter is Bhairahawa. The population of the Rupandehi district was 11,18,975 as per the National Census 2078. About 1000 hectares of the land area of Rupandehi is covered with water of which the natural wetland/lake and aqua ponds share 125 hectares and 875 hectares, respectively. The community pond water surface area occupies 250 hectares and the owner's pond water area is 625 hectares. Similarly irrigated rice field area occupies 47617.0 hectares.

##### **3.3. General Description of Chhapiya Village**

Chhapiya village, which is very famous for aquaculture is located in Siyari rural municipality. Chhapiya is highly known by the name of a fish village. Most of the people of this area were found to have aquaculture practices rather than growing paddy as their means of living due to the productivity of fish culture. Farmers in this area are converting their crop fields into the fish ponds for aquaculture which has benefitted the farmer. It has changed their standard of living than raising crop fields.



**Figure 1:** Location Map of the Study Area

### **3.4 Data Collection**

Data were collected from the different fish ponds of Chhapiya village which was selected randomly.

#### **3.4.1 Measurement of Water Parameters (Dissolved Oxygen, pH, and Temperature)**

Dissolved oxygen, pH, and temperature were measured by using the pH meter, DO meter, and thermometer respectively.

### **3.4.2 Detection and Identification of Ecto-parasite**

#### **a. Host Fish Selection**

Five species of carp Rohu (*Labeo rohita*), Naini (*Cirrhinus mrigala*), Silver carp (*Hypophthalmichthys molitrix*), common carp (*Cyprinus carpio*), Grass carp (*Ctenopharyngodon idella*) were selected as the host species for the present study. A total of 100 fish were taken for the study of the prevalence, abundance, and intensity of the parasite. Twenty fish including 10 males and 10 females were taken in each host species. Further, for length-wise prevalence, abundance, and intensity of fish, a total of 150 fish were taken. The length of fish was categorized into three categories consisting of length groups 10-20 cm, 20-30 cm, and 30-40 cm with 10 fishes in each category making 30 fishes of each host species. Fishes were collected randomly from the farmer's pond and a total number of 250 fish were examined during the study.

#### **b. Collection of Specimen**

The live fish were collected directly from the pond of the study area using a seine net. The data was collected from Falgun of 2077 to Baisakh of 2078 taking 3 months period.

#### **c. Identification of Male and Female Fishes**

The male and female fish were identified considering the following points:

- i. The male fish are slimmer and smaller whereas the female is larger and rounder.
- ii. Males have long pointed anal fins whereas females has shorter triangular anal fins.
- iii. Males have external sexual organs.
- iv. Females have a swollen abdomen.

### **3.5 Observation of Ecto-parasite**

As ectoparasites are present on the external surface of the host fish and can be seen easily with necked eyes, a magnifying glass was used for the clear observation of parasites present on the skin, scales, fins, and operculum area. The number of the parasite was recorded. Further, other physical abnormalities like ulcers, lesions, raised scales, cysts, and injuries resulting from physio-chemical agents were also recorded.

Parasites were collected with the help of a fine brush and blunt forceps and kept in a petri dish for counting.

### **3.6 Estimation of Prevalence, Abundance, and Intensity**

Prevalence, abundance, and intensity were determined according to the formulas used by Margolis et al. (1982) as follows:

$$\text{Prevalence (\%)} = \frac{\text{Total number of infected fish} * 100}{\text{Total number of fish examined}}$$

$$\text{Abundance} = \frac{\text{Total number of parasite}}{\text{Total number of fish examined}}$$

$$\text{Mean intensity} = \frac{\text{Total number of parasites collected}}{\text{Total number of fish infected}}$$

A total number of the parasite was determined directly by numerical count. The number of fish sampled, prevalence, abundance, and intensity were analyzed and interpreted according to Margolis et al. (1982).

## CHAPTER-IV

### 4. RESULTS

#### 4.1 Water Quality Parameters

The major water quality parameters such as pH, DO, and temperature was recorded. These parameters were found to be slightly varied during each measurement. The maximum pH recorded was (8.7) and the minimum was (8.1) with a mean pH of  $8.35 \pm 0.246$ . Similarly, the dissolved oxygen content was recorded maximum of (8.9 mg/l) and a minimum of (7.3 mg/l) with a mean of  $8.29 \pm 0.587$  mg/l whereas the maximum temperature was found to be  $31^{\circ}\text{C}$  and a minimum  $21^{\circ}\text{C}$  with a mean temperature of  $27.28 \pm 3.385$  °C.

**Table 1:** PH, DO and Temperature recorded in different fish ponds.

S.N.	Visit days	Measurements		
		pH	DO (mg/l)	Temperature (°C)
1	1 <sup>st</sup>	8.2	8.9	21
2	2 <sup>nd</sup>	8.3	8.9	23.3
3	3 <sup>rd</sup>	8.5	8.7	25.5
4	4 <sup>th</sup>	8.1	7.9	27
5	5 <sup>th</sup>	8.6	8.6	28
6	6 <sup>th</sup>	8.4	7.4	26
7	7 <sup>th</sup>	8.7	7.3	30
8	8 <sup>th</sup>	8.6	8.1	30
9	9 <sup>th</sup>	8.0	8.6	31
10	10 <sup>th</sup>	8.1	8.5	31
Mean $\pm$ SD		$8.35 \pm 0.246$	$8.29 \pm 0.587$	$27.28 \pm 3.385$

#### 4.2 Prevalence, Abundance, and Intensity of *Argulus* in Different Host Species

Out of 100 host fishes taken for the total estimation of infestation, only 28 fishes were found to be infected with *Argulus* species. Table 2 shows that the highest prevalence (50%), abundance (1.4), and intensity (2.8) was found in *L. rohita* followed by *C. mrigala* (prevalence 40%, abundance 1 and intensity 2.5 per fish), *C. carpio*

(prevalence 25%, abundance 0.5 and intensity 2 per fish), *H. molitrix* (prevalence 15%, abundance 0.25 and intensity 1.667 per fish) and lastly *C. idella* (prevalence 10%, abundance 0.15 and intensity 1.5 per fish) were recorded.

**Table 2:** Total Estimation of prevalence, abundance and intensity of *Argulus* species in five different host species

Name of species	No. of host fishes		No of <i>Argulus</i> species	Prevalence %	Abundance	Intensity
	Examined	Infected				
<i>Labeo rohita</i>	20	10	28	50	1.4	2.8
<i>Cirrhinus mrigala</i>	20	8	20	40	1	2.5
<i>Cyprinus carpio</i>	20	5	10	25	0.5	2.0
<i>Hypophthalmichthys molitrix</i>	20	3	5	15	0.25	1.667
<i>Ctenopharyngodon idella</i>	20	2	3	10	0.15	1.5
<b>Total</b>	<b>100</b>	<b>28</b>	<b>66</b>	<b>28</b>	<b>0.66</b>	<b>2.357</b>

### 4.3 Infestation of *Argulus* Species with Sex of the Fish

A hundred fishes were taken for the study of sex-wise infestation of species. As shown in Table 3 female carps were highly infected compared to males. The prevalence, abundance, and intensity were found higher in the female of *L. rohita* (prevalence 60%, abundance 1.8, and intensity 3) followed by *C. mrigala*, *C. carpio*, *H. molitrix*, and *C. idella*.

**Table 3:** Total sex-wise Prevalence, Abundance and Intensity of *Argulus* species in the host species

Name of species	Sex	No. of host fishes		No of <i>Argulus</i> species	Prevalance %	Abundance	Intensity
		Examined	Infected				
<i>Labeo rohita</i>	M	10	4	10	40	1	2.5
	F	10	6	18	60	1.8	3
<i>Cirrhinus mrigala</i>	M	10	3	8	30	0.8	2.667
	F	10	5	12	50	1.2	2.4
<i>Cyprinus carpio</i>	M	10	2	4	20	0.4	2.0
	F	10	3	6	30	0.6	2.0
<i>Hypophthalmich thys molitrix</i>	M	10	1	2	10	0.2	0.5
	F	10	2	3	20	0.3	1.5
<i>Ctenopharyngodon idella</i>	M	10	1	1	10	0.1	1
	F	10	1	2	10	0.2	2
Total		100	28	66	28	0.66	2.357

\*M = Male and F= Female

Table 3 shows that female carp of all species were highly infected as compared to that males. The highest rate of infestation was found in females of *L. rohita* (Rohu) followed by females of *C. mrigala*, *C. carpio*, *H. molitrix*, and *C. idella* respectively.

#### 4.4 Infestation of *Argulus* Species with Length of the Fishes

During the study, the host fishes which were taken for the study of length-wise prevalence, abundance, and intensity of *Argulus* species were categorized into three major groups based on different sizes as shown in tables (4, 5, 6, 7, and 8), where the highest record was from small sized fishes which shows small sized fishes were more susceptible to Argulosis.

#### 4.4.1 Length-wise Prevalence, Abundance, and Intensity of Infestation of *Argulus* Species on *Labeo rohita*

From Table 4, the maximum prevalence of *Argulus* (50) was observed in the smallest length group (10-20 cm), and the minimum prevalence (20) was found in the (30-40) cm length group of *L. rohita*.

**Table 4:** Length-wise prevalence, abundance and intensity of infestation of *Argulus* species on *Labeo rohita* (Rohu)

Length group cm	No of host fish		No of <i>Argulus</i>	Prevalence	Abundance	Intensity
	Examined	Infected				
10-20	10	5	12	50	1.2	2.4
20-30	10	3	7	30	0.7	2.333
30-40	10	2	4	20	0.4	2.0
Total	30	10	23	33	0.76	2.3

#### 4.4.2 Length-wise Prevalence, Abundance, and Intensity of Infestation of *Argulus* Species on *Cirrhinus mrigala*

Table 5 shows that the highest prevalence (40), abundance (0.8), and intensity (2) were recorded in the length group 10-20 cm in *C. mrigala* (Naini) and the lowest prevalence and abundance were found in the (30-40) cm group.

**Table 5:** Length wise prevalence, abundance and intensity of infestation of *Argulus* species on *Cirrhinus mrigala* (Naini)

Length group cm	No of host fish		No of <i>Argulus</i>	Prevalence	Abundance	intensity
	examined	infected				
10-20	10	4	8	40	0.8	2
20-30	10	3	5	30	0.5	1.67
30-40	10	1	2	10	0.2	2.0
Total	30	8	15	26.6	0.5	1.875

#### 4.4.3 Length-wise Prevalence, Abundance, and Intensity of Infestation of *Argulus* Species on *Cyprinus carpio*

Table 6 shows that the highest prevalence (50), abundance (0.8), and intensity (1.6) was recorded in the length group 10-20 cm in *C. carpio* (Common carp) with the lowest prevalence, abundance, and intensity in the length group of 30-40 cm.

**Table 6:** Length wise prevalence, abundance, and intensity of infestation of *Argulus* species on *Cyprinus carpio* (common carp)

Length group cm	No of host fish		No of <i>Argulus</i>	Prevalence	Abundance	intensity
	examined	infected				
10-20	10	5	8	50	0.8	1.6
20-30	10	2	5	20	0.5	2.5
30-40	10	2	3	20	0.3	1.5
Total	30	9	16	30	0.53	1.78

#### 4.4.4 Length wise prevalence, abundance, and intensity of infestation of *Argulus* species on *Hypophthalmichthys molitrix*

Table 7 shows that the highest prevalence (30), abundance (0.6), and intensity (2) was found in the length group 10-20 cm in *H. molitrix* (Silver carp) with the lowest prevalence, abundance, and intensity in the length group of (30-40) cm.

**Table 7:** Length wise prevalence, abundance, and intensity of infestation of *Argulus* species on *Hypophthalmichthys molitrix* (silver carp)

Length group cm	No of host fish		No of <i>Argulus</i>	Prevalence (%)	Abundance	intensity
	examined	infected				
10-20	10	3	6	30	0.6	2.0
20-30	10	2	3	20	0.3	1.5
30-40	10	1	2	10	0.2	2.0
Total	30	6	11	20	0.36	1.83

**4.4.5 Length wise prevalence, abundance, and intensity of infestation of *Argulus* species on *Ctenopharyngodon idella***

Table 8 shows that the highest prevalence (20), abundance (0.4), and intensity (2) was found in the length group 10-20 cm in *C. idella* (Grass carp) with the lowest prevalence, abundance, and intensity in the length group of 30-40 cm.

**Table 8:** Length wise prevalence, abundance, and intensity of infestation of *Argulus* species on *Ctenopharyngodon idella* (grass carp)

Length group cm	No of host fish		No of <i>Argulus</i>	Prevalence	Abundance	intensity
	examined	infected				
10-20	10	2	4	20	0.4	2.0
20-30	10	2	3	20	0.3	1.5
30-40	10	1	1	10	0.1	1.0
Total	30	5	8	16.6	0.26	1.6

## CHAPTER-V

### 5. DISCUSSION

Regarding water quality parameters, the Mean  $\pm$  SD of pH, DO and temperature was found to be  $8.35 \pm 0.246$ ,  $8.29 \pm 0.587$ , and  $27.28 \pm 3.385$  respectively which is almost similar to the study performed by (Das et al., 2016) in which the Mean  $\pm$  SD of pH, DO and temperature was found to be  $7.2 \pm 0.08$ ,  $5.3 \pm 0.22$  and  $26.3 \pm 2.02$  respectively. Comparing both data the mean DO is slightly higher in my study which is good for fish culture.

The DO is regarded as the most important basis for aquaculture practice. If it drops below the normal range, it may directly influence the fish and might cause mass mortality of fish. It can create a negative impact on farmers. The DO was found to be in the normal range (7 to 9) in most of the ponds of the study area.

The maximum level of infestation was found in *L. rohita* and the minimum in *C. idella* i.e. 15.2 % and 12 % respectively. Similarly, the mean intensity of Argulus infestation was recorded maximum in *L. rohita* and *C. carpio* as  $0.84 \pm 0.22$  and  $0.84 \pm 0.24$  respectively (Gautam, 2018) which is similar to my findings.

Similarly, according to (Gautam, 2018) the abundance of the parasites in different fish species was found to be highest in *C. carpio* followed by *L. rohita*, *H. molitrix*, *C. mrigala*, and *C. idella* which is slightly different than my study. Because in my study the abundance is higher in *L. rohita* followed by *C. mrigala* and *C. carpio*. Seasonal variation in the water quality affects the abundance of pathogens and their ability to survive on a host.

During summer, fish are heavily infected by the intense attacks of parasites because of their hemorrhagic skin. Mostly parasite concentration appears heavier around the pectoral and pelvic fins. The increase in temperature was favorable for the multiplication of parasites and found maximum during the summer months. The prevalence abundance and mean intensity were maximum in these months. This result agrees with Koyun (2011) who noted that fish lice increase rapidly depending on water temperature.

In the present study, it was found that higher number of ectoparasite recorded on the body surface, the base of the pelvic, pectoral, anal, and caudal fin, and lowest in the intestine of the examined carp. This result strongly supported Das et al. (2016) who reported more parasites on the body surface and fins. In the same way, Delwer et al. (2010) also reported a higher number of parasites on the skin and the lowest in the intestine.

The result revealed that there was the highest parasitic infestation in Indian major carp's *Labeo rohita*, *Cirrhinus mrigala* followed by others. Indian major carp were more susceptible to the *Argulus* infestation which is similar to the result of Vijaysundardeva et al., 2018. (Vijaysundardeva et al., 2018).

Walker et al (2008) found a difference in louse burdens in different species of fish. They reported the highest prevalence of *A. foliaceus* on fish species dominated by larger individuals. *C. carpio* and *A. brama* had the largest mean louse intensities. They found a pattern of increasing infestation intensity with an increase in host body weight. This research is a little dissimilar from the result of the present study because small-sized fish were found more infected in this study. This may be due to influence in physiochemical parameters or may be due to lack of sufficient balanced diet for fish fingerlings and immature immune systems.

According to Alom et al. (2019) species wise highest infestation (65.71%) and intensity (25.86 per fish) of *Argulus* species, were found in *Labeo rohita*, followed by *Cyprinus carpio* (60.0% and 12.0 per fish) and lowest in *Cirrhinus mrigala* (38.6% and 5.02 per fish) which is also similar to my findings.

Overall sex wise infestation of *Argulus* was found higher in females in comparison to males (Rahman et al., 2019). The reason may be the weak body of the female due to reproduction. Within female fish, female *Labeo rohita* have high infestation which is similar to the present research. According to (Gautam, 2018) length-wise infestation was higher in the largest size group (30-40cm) than in smaller ones (10-20 cm) which contradict my study. It may be due to a faulty water environment, high stocking density, and introduction of infected fish fingerlings. But, according to Golder et al. (1987), small-size fishes were more susceptible to *Argulus* species which is similar to the present result. He observed that among the different size groups of fish, the prevalence was highest in the small length group due to less immune power.

## CHAPTER-VI

### 6. CONCLUSION AND RECOMMENDATION

#### 6.1. Conclusion

The study was carried out to find the *Argulus* infection in different five species of fish based on sex and body length (cm). Argulosis was found as a major disease affecting the fish species. The infestation rate of parasites varied with the species, sex, and body length of fish. Results suggest that *L. rohita* had the highest rate and *C. idella* had the lowest rate of prevalence, abundance, and intensity. Further, sex-wise data shows that female fishes were more susceptible to parasites than that males. Within the female fish, *L. rohita* females were highly susceptible. Similarly, length-wise infestation was found higher in small-sized (0-10 cm) fishes as compared to large (30-40 cm) sized.

The present study revealed that heavy infestation and intensity of *Argulus* can increase mortality due to low growth survival and production of carp fish in captivity. It can be concluded from the present investigation that *Argulus* species were found all over the body surface of the host but most preferred sites of *Argulus* were the base of pectoral and pelvic fins and anal fins. As infestation where found higher in small-sized fishes than the larger ones it may create an impact on the development of commercial hatcheries. In this research, it was found that female carps were more heavily infected than males which may create a negative impact on the production of healthy fish fries.

#### 6.2 Recommendations

From the overall study, I would like to recommend the following points to reduce parasitic infection in fish:

1. Proper management of ponds and maintenance of water quality.
2. Proper stocking density should be maintained to minimize the risk of parasitic disease.
3. Proper maintenance of the water exchange system.
4. Government should give high priority to improving the cultural practices and training must be given to aqua pond farmers.

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## PHOTOPLATES



**Photo 1: Pond in the study area**



**Photo 2: Researcher in the study area**



**Photo 4: Measuring the length of large fish**



**Photo 3: Measuring the length of small fish**



**Photo 6: Collection of live specimen using seine net**



**Photo 5: Researcher measuring water temperature**



**Photo 7: Detaching Argulus from the fish body**