

1. INTRODUCTION

1.1. Background

Limnology (from Greek: limnee, "lake"; and logos, "knowledge"), also called freshwater science, is the study of inland waters. It covers the biological, chemical, physical, geological and other attributes of all inland waters (running and standing waters, both fresh and saline, natural or man made). This includes the study of lakes and ponds, rivers, springs, streams and wetlands.

The term limnology was coined by Forel (1901) who established the field with his studies of Lake Geneva. Forel's original definition of limnology, "the oceanography of lakes", was expanded to encompass the study of all inland waters, and influenced Benedykt Dybowski's work on Lake Baikal.

As a field of study limnology is "a multidisciplinary field that involves all sciences that can be brought to bear on understanding the nature of such waters: the physical, chemical, earth, biological sciences, and mathematics" (Edmondson, 1994).

Limnology, as a field of study, integrates the examination of the physical, chemical, and biological components of inland aquatic ecosystems, as well as how the surrounding landscape and atmosphere impact those waters and their biological components (ASLO, 2012).

Limnological studies are highly interdisciplinary, with a great deal of overlap with other disciplines such as aquatic science, aquatic ecology, oceanography, ichthyology (study of fish), and wetland ecology, and can be considered a subfield of ecology (Arlinghaus et al., 2008). Today, limnology can be understood as a science that takes a holistic approach to understanding the structure and function of these bodies of water (Arlinghaus et al., 2008).

Studying inland waters, which are continental bound, have significant importance to humankind. We human being much dependent on this relatively small amount of water and understanding it will help to maximize the utilization of resources and minimizing impact on it. . The limnological discipline integrates the functional relationships of growth, adaptation, nutrient cycles, and biological productivity with species composition,

and describes and evaluates how physical, chemical, and biological environment regulate these relationships. Understanding of the causal mechanisms operating in and controlling our natural world is a primary objective of limnology because of the premier importance of fresh water for the well being of humankind. The greater our understanding, the higher the probability to predict accurately patterns of events within aquatic ecosystems in response to human manipulations and disturbances. The objectives of carrying out the physico-chemical and biological analyses of water bodies are as follows-

- a) Limnological evaluation for wetlands leads to information about their misuse by indicating the pollution status.
- b) Since the quality of aquatic life depends on the water quality, a thorough assessment of the water quality is an integral part of wetland evaluation.
- c) The assessment of the chemical criteria of the water body helps in evaluating the chemicals that cause toxicity to aquatic life and long-term effects on the ecosystem.
- d) Designate uses that protect the structure and function of wetlands for protection of fish, birds, wildlife, and recreation.
- e) Analyse the qualitative and quantitative aspects of plankton population of the water bodies.

Water quality is determined by physical, chemical, biological and microbiological properties of water. These water quality characteristics throughout the world are characterized with wide variability. Therefore the quality of natural water sources used for different purposes should be established in terms of the specific water-quality parameters that most affect the possible use of water. The physicochemical parameters of water bodies influence directly or indirectly the number, varieties, distribution, metabolic activities, growth etc. of the aquatic organisms in various ways. Functioning of the aquatic ecosystem is regulated by the interaction among the physicochemical and biological components of the system. Hence, it is essential to have the knowledge of physicochemical parameters of water bodies for aquaculture.

The physicochemical parameters of a water body change due to seasonal change, diurnal changes and pollutants. These bring significant seasonal and diurnal change in abundance of aquatic organisms. Among the physicochemical parameters air temperature, water

temperature, transparency, pH, dissolved oxygen, free carbon dioxide, alkalinity, hardness, chloride and BOD mainly determine the hydrological condition of water body. The physical, chemical and biological parameters that has been considered in this thesis work has been discussed below.

a) Colour

Colour of water is primarily a concern of water quality for aesthetic reason. coloured water give the appearance of being unfit to drink. On the other hand, colour can indicate the presence of organic substance, such as algae or humic compounds. Colour in natural waters may occur due to the presence of humic acid, falvic acids, metallic ion such as iron and manganese, suspended matter, phytoplankton, weeds and industrial wastes etc. Colour due to organic acids may not be harmful as such, but highly coloured waters are objected on aesthetic grounds. Colour has been used as a quantitative assessment of the presence of potentially hazardous or toxic organic materials in water

b) Depth

Depth of water body has important bearing on the physical and chemical qualities of water. It determines the temperature, circulation pattern of water and the extent of Photosynthetic activity.

c) Temperature

Impinging solar radiation and atmospheric temperature brings about spatial and temporal changes in temperature, setting up convection currents and thermal stratification. Temperature plays a very important role in wetland dynamism affecting the various parameters such as alkalinity, salinity, dissolved oxygen, electrical conductivity etc. In an aquatic system, these parameters affect the chemical and biological reactions such as solubility of oxygen, carbon dioxide, carbonate-bicarbonate equilibrium, increase in metabolic rate and physiological reactions of organisms, etc. Water temperature is important in relation to fish life. Fish are poikilothermic so, the rates of biochemical processes are temperature dependent. A 10°C rise in temperature doubles the reaction rate.

d) Transparency

Solar radiation is the major source of light energy in an aquatic system, governing the primary productivity. Transparency is a characteristic of water that varies with the combined effect of colour and turbidity. It measures the light penetrating through the water body.

e) pH:

The effect of pH on the chemical and biological properties of liquids makes its determination very important. It is one of the most important parameter in water chemistry and is defined as $-\log [H^+]$, and measured as intensity of acidity or alkalinity on a scale ranging from 0-14. If free H^+ are more it is expressed acidic (i.e. $pH < 7$), while more OH^- ions is expressed as alkaline (i.e. $pH > 7$).

In natural waters pH is governed by the equilibrium between carbon dioxide/bicarbonate/carbonate ions and ranges between 4.5 and 8.5 although mostly basic. It tends to increase during day largely due to the photosynthetic activity (consumption of carbon-di-oxide) and decreases during night due to respiratory activity. Waste water and polluted natural waters have pH values lower or higher than 7 based on the nature of the pollutant.

In aquaculture, the direct effects of high or low pH usually are less important than indirect effects of pH. In many low alkalinity waters, pH is not low enough to harm fish, but it is low enough to reduce the amount of dissolved inorganic phosphorus and carbon dioxide available for plankton. Liming is employed to improve productivity in low alkalinity waters.

f) Dissolved oxygen

Oxygen dissolved in water is a very important parameter in water analysis as it serves as an indicator of the physical, chemical and biological activities of the water body. The two main sources of dissolved oxygen are diffusion of oxygen from the air and photosynthetic activity. Diffusion of oxygen from the air into water depends on the solubility of oxygen, and is influenced by many other factors like water movement, temperature, salinity, etc. Photosynthesis, a biological phenomenon carried out by the autotrophs, depends on the plankton population, light condition, gases, etc.

The dissolved oxygen content of warmwater fish habitats shall not be less than 5 mg/liter during at least 16hr of any 24-hr period. Although fish can survive low concentration of dissolved oxygen, particularly if free carbon dioxide concentrations are low, prolonged exposure to low dissolved oxygen is harmful.

g) Free carbondioxide

The important source of free carbon dioxide in surface water bodies is mainly from respiration and decomposition by aquatic organisms. It reacts with water partly to form calcium bicarbonate and in the absence of bicarbonates gets converted to carbonates releasing carbon dioxide.

High concentrations of carbon dioxide have a narcotic effect on fish and even higher concentration may cause death. In water used for intensive fish culture, free carbon dioxide levels typically fluctuate from 0mg/liter in the afternoon to 5 or 10mg/liter at daybreak with no obvious ill effect on fish.

h) Alkalinity

Alkalinity commonly means concentration of carbonate, bicarbonate and hydroxide ions in water expressed as CaCO_3 . In alkaline waters essential nutrients are found in higher quantities and this is the most important reason for the higher biological productivity in alkaline waters than in acidic waters. But highly alkaline condition is not favorable for biological production. Hydroxide alkalinity indicates water pollution. Total alkalinity generally ranges from 0 to several hundred ppm in natural water bodies.

i) Total hardness

Hardness is the property of water which prevents the formation of lather with soap and increases the boiling point of waters. Principle cations imparting hardness are calcium and magnesium. However other cations such as strontium, iron and manganese also contribute to the hardness. The anions responsible for hardness are mainly carbonates, bicarbonate, sulphate, chloride and silicates, etc. total hardness may vary from 0 to several hundred ppm. Hardness of pond depends on physicochemical conditions of basin soil, watershed, climate, liming, fertilization etc.

j) Biological parameters:

All natural waters contain a variety of organisms, both plants and animals as the natural flora and fauna. The term plankton was firstly proposed by Victor Hensen (1887), to describe the free floating and drifting microscopic organisms having almost neutral buoyancy. The locomotion power of plankton is feeble and the current and waves move them from one place to other. Planktons are classified on the basis of size, habitat, origin, quality and life history. On the basis of size planktons are three different types:

- a) Macroplankton- planktons, which are visible to naked eye.
- b) Mesoplankton- planktons, which can be secured by net of no.25 bolting silk cloth .
- c) Nanoplankton- these are the plankton of very small size and are not secured by no. 25 bolting silk.

Broadly the planktons are classified as:

- a) Phytoplankton
- b) Zooplankton

a) Phytoplankton:

- b) Phytoplankton are the autotrophic components of the plankton community. The name comes from the Greek words *phyton*, meaning "plant", and *planktos*, meaning "wanderer" or "drifter". Most phytoplankton are too small to be individually seen with the unaided eye. However, when present in high enough numbers, they may appear as a green discoloration of the water due to the presence of chlorophyll within their cells (although the actual color may vary with the species of phytoplankton present due to varying levels of chlorophyll or the presence of accessory pigments such as phycobiliproteins, xanthophylls, etc.). The green phytoplanktons are responsible for primary productivity and forms the primary trophic level in an aquatic ecosystem. They are known as producer. Some of phytoplanktons are non photosynthetic, eg: bacteria and fungi. Phytoplankton are a key food item in both aquaculture

and mariculture. Both utilize phytoplankton as food for the animals being farmed.

c) **Zooplankton**

Zooplankton are heterotrophic (sometimes detritivorous) plankton. Plankton are organisms drifting in oceans, seas, and bodies of fresh water. The word "zooplankton" is derived from the Greek *zoon*, meaning "animal", and *plankton* meaning "wanderer" or "drifter". Individual zooplankton are usually too small to be seen with the naked eye, but some, such as jellyfish, are large. They form the link between the primary trophic level and tertiary level of the aquatic ecosystem.

1.2. Justification of the study:

The following points have justified the purpose of study:

- a) Though Nepal has great water resources but very little work has been done in the field of limnology.
- b) The zooplankton forms the major food for fry and fingerlings to grow. So, the knowledge about them is prerequisite for pisciculture.
- c) The baseline data obtained from the study will be useful for the management committee to use the resource in the efficient way.

1.3. Objectives of the study:

The objectives of the present work are as follows:

General objective:

- a) To investigate some physicochemical water parameters and zooplankton of the pond.

Specific objectives:

- b) To explore the diversity of zooplankton of the pond.
- c) To determine the effects of physicochemical parameters on the zooplankton.

1.4 Limitation of the Study

Every research work has its own limitations. The present study also has some limitations, which are as follows:

- a) The study has covered only three stations of the pond.
- b) The researcher being student could not venture for extensive study in order to cope up with the limited financial resources.
- c) Time limitations was also a major factor for the researcher since, she had to complete the work within 6 months. Therefore, the researcher is not able to deal with many other factors related to ecosystem of the pond.

2. LITERATURE REVIEW

The science of limnology had a steady improvement since time immemorial. Although, limnological observations have long history but they evolved as a distinct science during the last two centuries after the invention of microscope, silk plankton net, thermometer etc.

Interest in the discipline rapidly expanded, and August Thienemann (a German zoologist) and Einar Naumann (a Swedish botanist) co-founded the International Society of Limnology (SIL, for originally Societas Internationalis Limnologiae) in 1922. Forel's original definition of limnology, "the oceanography of lakes", was expanded to encompass the study of all inland waters, and influenced Benedykt Dybowski's (Polish naturalist and physician) work on Lake Baikal.

The development of the science of limnology in North America is considered to be entwined with the careers of Edward Asahel Birge and Chancey Juday (Beckel 1988). It is their partnership at the University of Wisconsin-Madison that substantially laid the foundations of limnology in North America (Beckel 1988). Their first paper together was published in 1900, but their first major paper came in 1901 with a classic publication on dissolved gases, "The inland lakes of Wisconsin: The dissolved gases of the water and their biological significance" (Beckel 1988).

Kavita and Sheela (2010), studied on Physico-chemical Parameters of Bharawa ponds, Rewari, Haryana and found that most of the physicochemical parameters viz. temperature, transparency, EC, free carbon dioxide, DO, chloride, carbonate, bicarbonate, alkalinity, hardness, calcium, magnesium, salinity, TDS and phosphate were beyond the permissible limits. pH and nitrate were found within the range. Indicating that the pond is highly polluted due to discharge of uncontrolled dairy effluents leading to eutrophication.

Mishra et al (2009-2010) conducted an assessment of the physicochemical parameters of Bhamka pond, Hanumana, Rewa district, India to determine the water quality of pond, for Agricultural, and Drinking and fish production. Unacceptable, high levels of assessment

parameters were observed in many cases for other Indian freshwater bodies except for turbidity, dissolve oxygen, Alkalinity, pH, nitrogen and phosphate which were found in higher concentration above freshwater limits. The cattle's, agricultural inputs, washings and other pollution creating activities have enhanced the heavy metals and altered the physicochemical and biological characteristics of the pond water.

Tidame and Shinde (2010-2011) provided quantitative information on the correlation of zooplankton with physico-chemical factors from man-made reservoir in the Nashik district and indicated that the distribution and density of zooplankton species influenced by physical and chemical factors of the environment.

Hamaidi-Chergui et al. (2006) carried out to determine the monthly variations of physico-chemical parameters in water samples from Chiffa river at Blida, North West of Algeria from April to August 2006. The various physicochemical characteristics of Chiffa river as temperature, pH, electric conductivity, chloride, calcium, nitrate, and inorganic phosphorous have been compared with the trophic status as suggested by various authors, then this river can safely be placed under the category of mesotrophic water bodies with moderate quantity of nutrients to support relatively good biota in the river.

Rahman and Hussain (2004-2005) studied on the abundance of zooplankton of a culture and a non-culture pond of the Rajshahi University campus which indicated that the culture pond showed better result than that of the non-culture pond regarding zooplankton production. Total zooplankton showed positive correlation with pH, carbonate alkalinity (CO_3) and bicarbonate alkalinity (HCO_3) in both ponds and DO, carbon dioxide (CO_2) in pond-1.

Rajashekhar et al. (2010) provided quantitative information on the seasonal variations of zooplankton and selected physicochemical variables of a large man-made reservoir in the Gulbarga district, India, which showed that rotifera was the dominant group throughout the study period and highest count was recorded in the summer season while low incidence was observed in southwest monsoon season. Zooplankton community was correlated with physicochemical parameters. The results indicated that the distribution and density of zooplankton species influenced by physical and chemical factors of the environment.

Sitre (2011) conducted studies on Ambazari lake of Nagpur city located in western part of Nagpur city, India, to assess the type of forms present and analyze their seasonal variation through quantitative estimation and noticed that number of zooplankton is relatively high during winter and summer season in the Ambazari lake water.

Shrivastav (2011-2012), recorded monthly variation in the number of zooplankton with the communities occupying higher trophic levels in Ramgarh Lake, Gorakhpur, U.P. The zooplanktons were represented by three groups of organisms in order Crustacea > Rotifera > Protozoa. In general, the production of zooplankton in the Lake was minimum during monsoon season and maximum during June-July months. The fluctuation in the number of zooplankton was discussed in relation to the physico-chemical and other environmental condition of the Lake. Dominance of Rotifers and Crustaceans indicate the eutrophic status of pond.

Jemi and Balasingh (2009-2010), attempted an investigation to study the physico-chemical characteristics of water in the two permanent temple ponds Kanyakumari district. A total of 15 parameters were analysed and their seasonal variations were discussed.

Paulose and Maheshwori(2005-2006) studied the seasonal variation in zooplankton community structure of Ramgarh Lake, Jaipur, Rajasthan. They found that copepod dominated for 6 months.

Bera et al. (2014) made the correlation study on zooplankton availability and Physicochemical parameters of Kangsbati reservoir. They reported the positive correlation with pH,DO. Alkalinity and negative correlation with water temperature.

Rahman et al.(2008) carried out the seasonal study on seasonal abundance and diversity of zooplankton in a semi-intensive prawn farm of Bagerhat district. They observed the positive correlation with alkalinity, hardness and negative with salinity.

Acharya et al. (2010), evaluated the monsoon and post monsoon water quality of Phewa lake in Pokhara, Midwestern Nepal. There was no significant difference in DO level, pH, TSS

whereas EC, TN and water temperature showed significant difference with respective seasons.

Sharma (2004), investigated the physico-chemical parameters and zooplankton of Kamalpokhari. He recorded that copepods dominated the pond and found positive correlation between zooplanktons and pH, DO, alkalinity and hardness.

Vaidya and Yadav (2000-2002), carried out investigation zooplankton of some fresh water bodies of Kathmandu valley with reference to water quality. Only three groups of zooplankton viz: Rotifera, Cladocera and Copepoda were collected from the lotic and lentic water bodies. A total of seventy one species of zooplankton were recorded during that period. The lentic water bodies supported a higher species richness constituting seventy species whereas the lotic water bodies constituted only seven species of zooplankton. The water quality of the investigated water bodies were found to be deteriorated due to discharge of untreated effluents, solid wastes and poor conservation practices.

Pal et al. (2002-2004) studied the physicochemical properties of seepage stream at Shripur area, eastern Nepal. The maximum air temperature was recorded in rainy season during first year (July, 2002 to June, 2003) and second year study period (July, 2003 to June, 2004). Water temperature was maximum in summer and lowest in winter season. Transparency, total alkalinity, total hardness, chloride were maximum in winter season. Free CO₂ and BOD was maximum in summer season. The minimum transparency, total alkalinity, total hardness and chloride were recorded in rainy season. DO and pH were maximum in winter and minimum in summer season during the whole study period.

Pal and Thapa (2010), investigated the general properties of water of Baidhya fish pond, Tankisinwari, Nepal in which ranges and correlation among different physico-chemical parameters were taken into consideration. The water quality of Baidya fish pond was normal except high fluctuation of chloride 1 ± 0.241 to 29.84 ± 0.260 mg/l and ammonia 1.55 ± 0.088 to 18.7 ± 0.061 mg/l during manuring period and casual addition of wastes like toilet cleaners, caustic potash etc.

3. MATERIALS AND METHODS

3.1. Study area

The study site, “Kamal Pokhari”, at Kamalvinayak, Bhaktapur, is located approximately 15 km away from Kathmandu. This place is situated at the verge of township and rural areas of Bhaktapur district, in ward no.4. the pond “Kamalpokhari” is a religious one surrounded by picturesque natural beauty with a temple at the side. In Newari, it is known as “Lamga Pukhu” and it is the fourth largest pond in the Bhaktapur district. The pond was made by the local people for different purposes. It is rich in aquatic vegetation as well as with fishes and number of faunas.

Its nomenclature as Kamal Binayak *Pokhari* is associated with an interesting event. In 1923, land survey team then headed by army personnel came to this area and asked the locals about the name of this pond. Locals gave account of this pond with historical connection, "Charumati, sister of Samrat Ashok had constructed three Chaitaya in the precinct of this pond. So, it is called Yathau Bahal or Yathau Bare." But the army-men couldn't pick the local Newari accent and observed around. They saw lotus leaves and flowers floating over the pond and termed the name of the pond as Kamal Binayak *Pokhari*. In short, it is called Kamal *Pokhari*. Due to the abundance of *Kamal* (lotus) in this pond, it was earlier known as Kamal Binayak *Pokhari*. However, lotus in this pond has disappeared since many years.

The pond has larger space which has been managed as picnic spot and the fish culture done in this pond has attracted many people for enjoyment. This has promoted internal tourism benefiting the local people to run small business enhancing their economic status

Three stations were selected in the pond for the convenient study, namely station ‘A’, ‘B’ and ‘C’.

Station ‘A’: This station was selected at the southwest corner of the pond.

Station ‘B’: This station was selected at the eastern side of the pond.

Station ‘C’: This station was selected at northeast corner of the pond.

Table 1: Morphometric data of “Kamal Pokhari” Kamalvinayak, Bhaktapur, Nepal.

Location:	
Latitude: 27 degree 39’N.	
Longitude: 85 degree 22’	
Pond surface above sea level	1338.0m
Area of the pond surface	10442.9m ²
Maximum length	117.6m
Maximum width	88.8m
Maximum depth	179m
Pond volume	1258367.04m ³

3.2 Study period

This study was performed for the total period of six months i.e. from August 14, 2012 to February 12, 2013. The water sample was collected fortnightly from each station for the analysis of chemical parameters like pH, DO, free CO₂, alkalinity and hardness. The reading of physical parameters such as depth, temperature, transparency, watercolor and nature of the day was observed on the field itself. A total of twelve observations were recorded during the period of six months of study.

3.3 Materials

1. pH meter
2. Sechhi disc
3. Standard mercury thermometer
4. Microscope
5. Plankton net
6. Chemicals

3.4. Methods

The parameters of the water sampling station were worked out in the field following Trivedi and Goel (1984) and AAPHA (1989).

3.4.1. Physical parameters

The following physical parameters were experimented for the present study:

- a. **Nature of the day:** The nature of the day was recorded at the spot during working hours by looking around the surroundings.
- b. **Colour :** A small amount of water was taken in a petridish for the judgment of color of the pond water. The petridish with water was kept in a white paper and then the color of the water was examined.
- c. **Depth:** The depth of the pond was measured by using a long nylon rope, which was tied with appropriate weight at its one end. The nylon rope along with weight was lowered in the water body till it touched the bottom of the pond. The length of the rope, which was just inside the water, was measured with the help of the measuring tape (cms). The depth of the three different stations was recorded and the mean depth was found out.
- d. **Temperature:** The temperature of water was measured by using standard mercury thermometer graduated up to 50 degree centigrade. The temperature of the surface water was measured simply by dipping the bulb of the thermometer into water body of pond.
- e. **Transparency:** The transparency of the water measured with the help of a Secchidisc. The Secchidisc is a circular metallic plate of 20cm diameter painted with four alternative black and white quadrants on the upper surface and a hook in the center to a graduated rope. The secchidisc was immersed in water and noted the depth in cm at which it just disappeared and then gradually the disc was pulled upward and noted the depth at which it just reappeared. Now, the average value of these two readings was noted as transparency in centimeter.

$$\text{Transparency (T)} = \frac{\text{depth disappear} + \text{depth reappear}}{2}$$

2

The extinction coefficient was calculated with the help of a standard formula,

$$K = 1.7/D$$

Where, D = Secchidisc reading, k= Extinction Coefficient and 1.7 = Constant factor

3.4.2 Chemical parameters

Chemical proerties of water influences the physical properties as well as biological activities of the living organisms. The important chemical parameters analyzed in the

present study were pH, DO, Free CO₂, total alkalinity and hardness. Water samples from the sampling sites were taken in the sterilized plastic bottles of 1,000 ml. These sampling bottles were rinsed with respective water samples at the respective points before filling the water and filled airtight. All the sampling bottles were marked with respective sampling sites. The above mentioned process was strictly followed to collect the water samples from other two points throughout the experimental period. The chemical parameters of water during the study period were experimented as follows:

- a. **pH:** The P^H of any water body indicates the extent of acidity or alkalinity. The P^H of water was determined by using automatic digital p^H meter (HANNA) by dipping the P^H meter in water for 2 minutes. Before taking the readings, P^H meter was calibrated with distilled water (P^H 7).
- b. **DO:** The DO was determined by the standard Wrinkle's method. The sample water was filled in the BOD bottle (glass stopper bottle) of 300 ml volume avoiding bubbling and trapping of air bubbles in the bottle after placing the stopper. In this sample water 2 ml of each MnSO₄ and Alkaline KI solution were poured and formed the precipitation. Now placing the stopper, the BOD bottle was shaken so that the contents would invert the bottle repeatedly. The bottle was kept for sometimes to settle down the precipitate and then 2 ml of conc. H₂SO₄ was added to dissolve the precipitate by shaking the content well. Now, 50 ml of content of BOD bottle was titrated with standard Sodium Thiosulphate (0.025 N) using starch as an indicator. At the end point, initial dark blue colour changes to colourless. The DO can be estimated by using the formula,

$$\text{DO (mg/l)} = \frac{(\text{ml} \times \text{N}) \text{ of titrant} \times 8 \times 1}{V_2}$$

Where, N = Normality (strength of Sodium Thiosulphate)

V₂ = Volume of content titrate

V₁ = Volume of sample bottle (BOD) bottle

V = Volume of MnSO₄ and KI added

- c. **Free Carbon dioxide:** Free Carbon-dioxide can be determined by titrating the sample using a strong alkali (such as carbonate free NaOH) to P^H 8.3. At this P^H, all the free CO₂ is converted into bicarbonates. The end point was appeared as pink colour. The Phnophthalein was used as indicator. Free CO₂ can be calculated as,

$$\text{Free CO}_2 \text{ (mg/l)} = \frac{(\text{ml} \times \text{N}) \text{ of NaOH} \times 1000 \times 44}{V}$$

Where, V = Volume of water sample taken (ml)

- d. **Total alkalinity:** Total Alkalinity is the measure of the capacity of the water is to neutralize the strong acid. The alkalinity in the water is generally imparted by the salts of Carbonates, Bicarbonates, Phosphates, Borates, Silicates etc., together with Hydroxyl ion in free state. Total alkalinity of water was determined by titrametric method. A100 ml of sample in a conical flask with 2-3 drops of methyl orange was titrated against standard, 0.02 N H₂SO₄. At the end point, yellow colour was changed to pink colour.

$$\text{Total Alkalinity (mg/l)} = \frac{A \times N \times 1000 \times 50}{V}$$

Where, A = Volume of Standard H₂SO₄ used in titration

N = Normality of H₂SO₄ used

V = Volume of water sample taken (ml)

- e. **Hardness:** Hardness is caused by the Calcium and Magnesium ions present in water. Total Hardness of water was determined by EDTA method. First 50 ml of water sample was taken in a conical flask and 1 ml of buffer solution with Erichrome Black- T indicator was added. Then, it was titrated against standard EDTA. The solution was changed wine red to blue at the end point. The working formula is,

$$\text{Total hardness as CaCO}_3 \text{ (mg/lit)} = \frac{\text{ml of EDTA used} \times 1000}{V}$$

Where, V = Volume of water sample taken (ml)

3.5. Sampling of water for the identification of Zooplanktons

For the estimation of zooplanktons, 10 litres of surface water was collected by using plastic bucket. The surface water was filtered through plankton net having bolt-silk no.30. The planktons that remained at the cap of the plankton net was collected and stored in a bottle. The sample for the zooplankton study were preserved by adding lugol's solution and 5 percent formalin solution and were brought to the laboratory . The quantitative and qualitative analysis of the zooplankton was done with the help of a Sedgwick- Rafter cell under a compound microscope. The calculation was done by using the following formula.

$$N = \frac{A \times 1000 \times C}{V \times F \times L}$$

Where, N= No. of plankton cells or units per litre of original

A= total no. of plankton counted

C= Volume of final concentrate of the sample in ml

V= Volume of field (1 cumm)

F= No. of fields counted

L = Volume of original water in litre

3.6 Statistical analysis

The statistical analysis to determine the coefficient of correlation between the different physico-chemical parameters and zooplankton was done by using the method adopted by “Karl Pearson” and the significance of the coefficient of correlation was tested.

The formula adopted for statistical analysis is as follows:

$$a) \text{ Coefficient of correlation}(r) = \frac{N \cdot \sum XY - \sum X \cdot \sum Y}{\sqrt{N \cdot \sum X^2 - (\sum X)^2} \times \sqrt{N \cdot \sum Y^2 - (\sum Y)^2}}$$

$$b) \text{ Probability error (P.Er)} = 0.6745 \times \frac{1-r^2}{n}$$

4. RESULTS

The observations of physico-chemical and biological parameters of “Kamal pokhari” were done fortnightly from each station and data was collected for a total period of six months i.e. from 14th August 2012 to 6th Feb 2013. The data thus obtained within six months were discussed below.

4.1 Physico-chemical parameters:

- Nature of the day:** The nature of the day i.e. sunny, cloudy, bit sunny, partly cloudy was observed in the field. Among the twelve observations, most days are sunny i.e. on 13th September, 29th September, 13th and 30th October, 17th November and 3rd December (Table -1).
- Colour:** During the study period the color of the water was changed from brownish in August to greenish in September and October and then light green in November and December to colorless January and February (Table-1).
- Depth:** The minimum depth recorded was 98cm on 6th February and maximum depth was recorded was 157cm on 30th August.(Table-1)

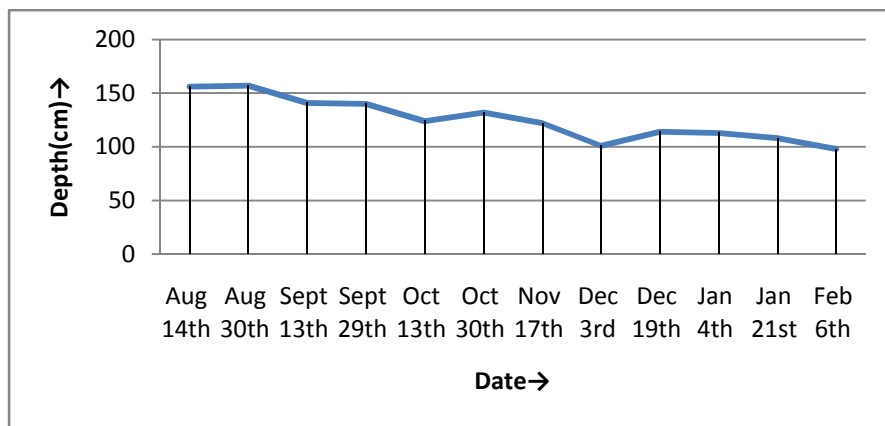


Figure 1: Fortnightly variation of Depth (cm)

- d. **Transparency:** Transparency in the present study ranged between 10.62 to 18.8cms of which higher value (18.8 cm) was reported in summer season while the lower value (10.62 cm) in rainy season. (Table -1)

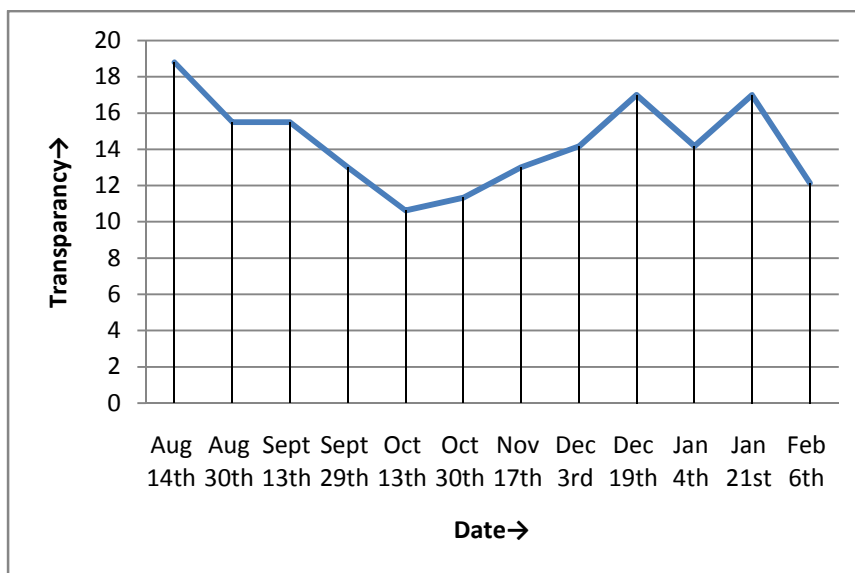


Figure 2: Fortnightly variation of Transparency (cm)

- e. **Temperature:** In the present study the average temperature is recorded to be ranging from minimum 11.2⁰C to maximum 27⁰C 4th January and 14th August respectively (Table -1).

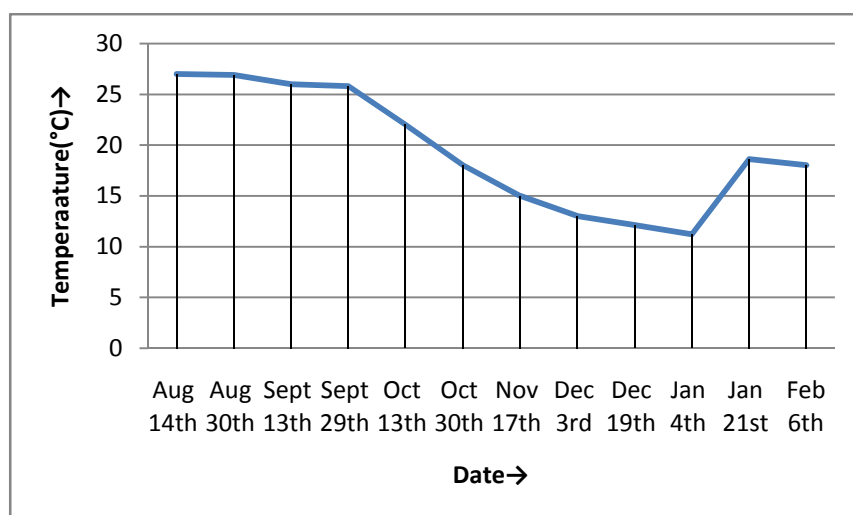


Figure 3: Fortnightly variation of Temperature

- f. **pH:** The pH of the water is alkaline and never recorded acidic. The minimum pH i.e. 7.2 was recorded on 3rd December and the maximum pH was recorded on 9.8 on 29th September (Table -1).

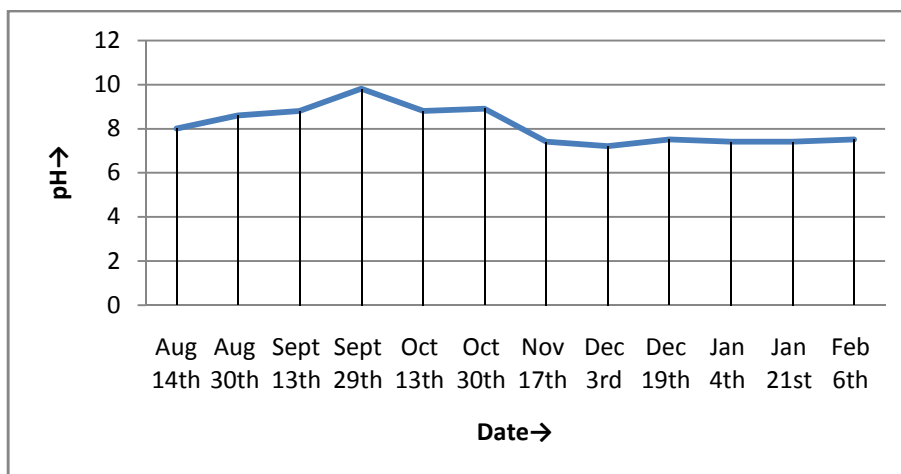


Figure 4: Fortnightly variation of pH

- g. **Dissolved oxygen:** The dissolved Oxygen content is recorded in the range of 3.3 mg/l to 7.5mg/l. The minimum value of DO is recorded in the month of February 6th and the maximum value of DO is recorded in the month of Sept 13th and October 3rd (Table1).

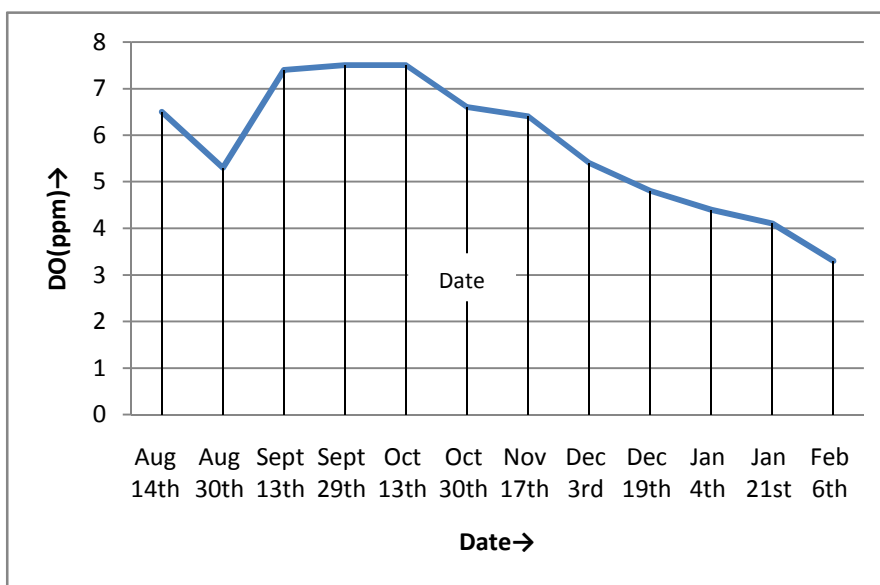


Figure 5: Fortnightly variation of DO

- h. **Free Carbon-dioxide (CO₂) :** The value of free CO₂ ranged between the 0 mg/l at the minimum to 8 mg/l at maximum. The minimum of CO₂ is recorded on 13th and 30th October and its maximum value is recorded on 6th February (Table -1).

- i. **Hardness:** The total hardness ranged from 5 mg/l at minimum to 23 mg/l at maximum. The minimum hardness is measured on 17th November and the maximum hardness is measured on 14th August (Table -1).

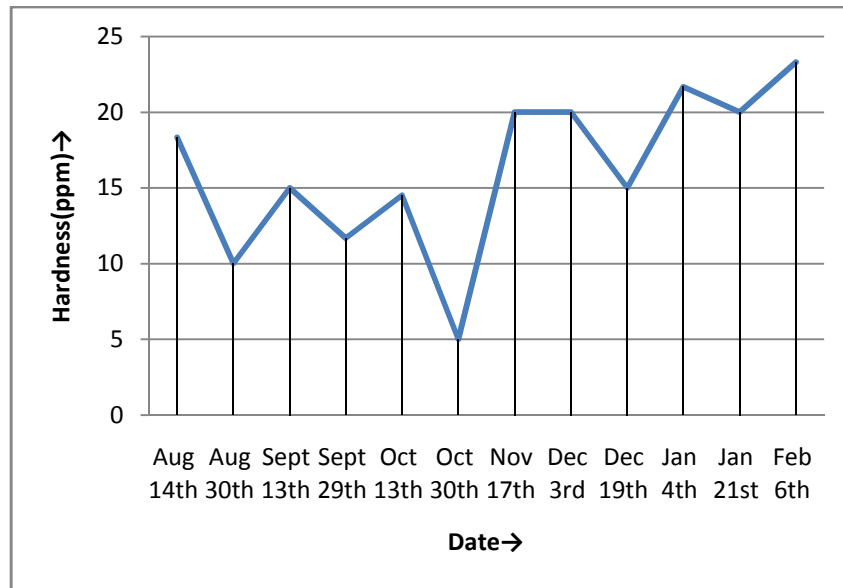


Figure 6: Fortnightly variation of hardness

- j. **Total Alkalinity:** The value of Total Alkalinity is measured to be ranged from 106 mg/l at minimum to 185 mg/l at maximum. The minimum value of Total Alkalinity is measured on 21st January while the maximum value is recorded on 30th October (Table -1).

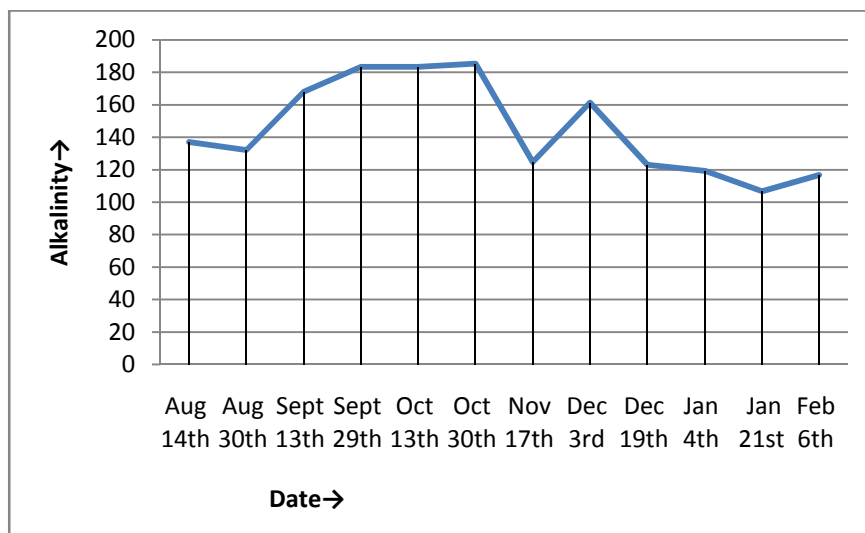


Figure 7: Fortnightly variation of alkalinity

4.2 Biological parameters:

- a. Zooplankton:** Zooplankton, the microscopic free swimming animal components of aquatic system are represented by wide array of taxonomic groups, of which the members belonging to Rotifera, Ostracoda, Copepoda and Cladocera were most common and often dominated the entire consumer communities. They are endowed with many remarkable features and are often armored with spines, which hamper their predation by higher organisms.

The zooplanktonic assemblage in this pond named “Kamal Pokhari” were contributed by two genera of rotifer (Plate I) i.e. Keratella (Plate I, fig.2), Brachionus (plate II, Fig. 1), two genera of Copepoda (Plate II) i.e. Cyclopes (Plate ii, fig. 1) and Diaptomus (Plate II, fig. 2) and one genera of Cladocera (Plate III) i.e. Daphnia (Plate III, fig.1) during the six months of study period i.e. 14th August 2012 to 6th February 2013.

The zooplanktons in the present investigation showed seasonal fluctuating pattern. The zooplanktons were less in the number in the month of January and February 2013. They were found maximum in the months of August, September and October 2012. The average number of zooplankton during the study period was 161.1 no/lit (Table 2) fortnightly. The total count of zooplanktons was found to be 1944 no/lit (Table 2).

The number of zooplanktons of sub-surface layer of water was identified both qualitatively and quantitatively. In Rotifera, Keratella was found varied from 27 to 107 no/lit. On 29th of September 2012 the no/ lit of Keratella was very high while on 4th January, 17th November and on 14th August it was minimum. Brachionus fluctuated from 4 to 46 no/ lit with an average value 20.41 no/lit. The minimum no/lit of Brachionus was seen on 4th January while the maximum was on 13th September (Table 2).

In case of Copepoda, Cyclopes varied from 14 to 254 no/lit. with an average value of 32.64 no/lit. The minimum was noted on 13th and 29th September, 30th October and on 4th January while maximum was noted on 30th August. The number of Diaptomus ranged from 14 to 94 no/lit with an average value of 22.5 no/lit. The minimum number

was found on,13th September and 13th October while maximum was found on 30th August (Table 2).

In Cladocera, Daphnia was found during the investigation period. Daphnia varied from 10 to 56 no/lit. with an average 25.4 no/lit. The minimum number per litre of Daphnia was seen in January 21st of 2013 while maximum was found on 13th October 2012 (Table 2).

The percentage of Rotifera, Copepoda and Cladocera was found to be 37.64%, 46.6% and 15.77% respectively.

Plate I : Rotifera



Fig 1: *Brachionus* (x 400)



Fig 2: *Keratella* (x 400)

Plate II: Copepoda



Fig 1: *Cyclops* (x 400)



Fig 2: *Diaptomus* (x 400)

Plate III : Cladocera



Fig 1: *Daphnia* (x 400)

4.3 Statistical analysis

The statistical analysis between physicochemical parameters and zooplankton was performed by Karl Pearson method.

1. Correlation between water temperature and zooplankton

Here, Co-efficient of correlation (r) = 0.5485

$$P.Er = 0.1949$$

There is positive correlation between water temperature and zooplankton. Hence, the correlation coefficient is positively significant.

2. Correlation between transparency and zooplankton

Here, Co-efficient of correlation (r) = 0.1763

$$P.Er = 0.1889$$

The correlation between transparency and zooplankton is positively significant.

3. Correlation between pH and zooplankton

Here, Co-efficient of correlation (r) = 0.427

$$P.Er = 0.1594$$

There is positive correlation between pH and zooplankton.

4. Correlation between D.O and zooplankton

Here, Co-efficient of correlation (r) = 0.4053

$$P.Er = 0.1629$$

There is positive correlation between D.O and zooplankton.

5. Correlation between alkalinity and zooplankton

Here, Co-efficient of correlation (r) = 0.2624

$$P.Er = 0.1815$$

There is positive correlation between alkalinity and zooplankton.

6. Correlation between hardness and zooplankton

Here, Co-efficient of correlation (r) = - 0.499

$$P.Er = 0.1464$$

There is positive correlation between hardness and zooplankton.

5. DISCUSSION

Water can be said as the cradle of life. In every water body, the quantity and quality of the biota is determined by the combined effects of physicochemical parameters. The interactions among these factors create favorable or unfavorable circumstances for the growth and development of any particular biotic element.

Water and air are the two vital fundamental media for life and it divides the world into two major divisions i.e. terrestrial and aquatic environments, but both are incompletely separated from each other. The study of fresh water ponds on all its varied aspects, such as physical, chemicals and biological aspects is termed as limnology. The particular biotic element is influenced by some climatic factors such as solar radiations, temperature, wind, water current, rainfall and some chemical factors such as acidity, salinity and then availability of inorganic nutrients needed by green plants for photosynthesis and primary production.

Therefore, it is necessary to study the physicochemical parameters of any aquatic body. The different parameters found in the present pond water during the study period are discussed below.

a) Colour

During the study period the color of the water was changed from brownish to greenish and then light green to colorless. The color of water mostly remained green, which may be due to swarms of planktons, algal blooms. When there is heavy rainfall the color of water was found to be brownish and it may be due to the deposition of surface runoff, suspended particles in the pond.

The fluctuation of water color affected the production of zooplankton. In the present study it has been observed that, when the water color was greenish to light green, the population of zooplankton was high but when it turned colorless the density of zooplankton decrease to minimum. Similar result was obtained by Sharma (2004).

b) Depth

Depth of water is an important physical parameter. It mainly affects the distribution of zooplanktonic population. The pond had a suitable depth averaging 125.7cm. The maximum depth was observed in the rainy season and minimum in the winter season.

The increase in depth in rainy season is due to rainfall and surface runoff. The depth of about 2 meter is considered congenial from the biological productivity point of view of a pond (Jhingram, 1985)

c) Temperature

Temperature is one of the most important factors of water, which varies at different times of the day and during different seasons of the year from place to place. The physical, chemical and biological parameters of the pond are directly or indirectly affected by temperature.

Variation of water temperature followed a similar trend to that of atmospheric temperature indicating the influence of seasonal and climatic conditions. Maximum water temperature (27°C), during August might be due to longer duration of days. This value did not deviate much from what had been reported earlier from Kamalpokhari (Sharma 2004). The temperature effects indirectly on the other factors of aquatic ecosystem like dissolved oxygen and free carbon dioxide. So, it indirectly influences on population supports this view. When the temperature was high, the population density of zooplankton was also high and vice-versa. The correlation coefficient between temperature and zooplankton was found positive. The value of correlation coefficient R was calculated to be 0.5, P.Er 0.2 which shows that there was positively significant correlation between temperature and zooplankton.

d) Transparency

Water is a transparent medium whose transparency enables the penetration of light to the depths, where it is ultimately absorbed. The transparency of pond water ranged from 10 to 18.8 cm. In the month of August, September and October, the transparency was low (Table:1). This may be due to swarms of planktons, disposal of sewage, suspended silt particles, etc. which reduced transparency. Transparency is inversely proportional to the turbidity of water.

Hutchinson (1957) found that, transparency of the pond depends upon the turbidity, which is caused by silting, microorganisms and suspended organic matters in the water.

During the study period it was observed that, when the transparency was low the population density of zooplankton was high and vice versa. Similar type of result was found by Sharma (2004). During the months of November and December 2013 transparency was high (Table:1), which may be due to scarcity of planktonic assemblage as well as by the settling down of suspended silt and solid particles.

e) The hydrogen ion concentration (pH)

The pH often becomes a determining factor for the biota by becoming a limiting factor in different habitats. The pH of chemically neutral water is 7. Water is acidic if the pH is below 7 and is alkaline if the value is above 7.

The pH value of the pond water was found alkaline throughout the study period. The pH value fluctuated from 7.2 to 9.8 with an average of 8.1. This pH value of pond water proved to be slightly alkaline and suitable for pisciculture. Ellis (1973) reported that pH value between 6.7 to 8.4 is suitable for aquatic life. Thus, it is nearly equal to the investigated value. From the present observation (Table: 1), it is seen that there is definite correlation between pH and zooplankton. The correlation coefficient (r), between pH and zooplankton was calculated to be positive, i.e. $r = 0.42$ and $P.Er. = 0.1$. The above result showed that when the pH value was high the population density of zooplankton was also high and vice versa. The finding resembles the work of Rahaman et al.(2013).

f) Dissolved oxygen

Dissolved oxygen is the most important factor for animal and plant life in the aquatic environment. It is available by absorption from the surface and from the photosynthesis of aquatic plants as well. The animal community, residing in the pond requires dissolved oxygen for respiration and releases carbon dioxide as a catabolic product both during day and night. Oxygen consumption in body of water occurs by the respiration of animals and aquatic plants, as well as by the putrefaction of organic matter and other causes. The oxygen available in the water at a given time is the balance of the above process.

Ideally, the oxygen producing and oxygen consuming process in natural water should be balanced so as to keep the dissolved oxygen concentration within a range congenial

to all organisms. Photosynthesis depends on the presence of sunlight; the depth up to which plant life may exist is limited by factors, which effect light penetration in the water. The concentration of DO becomes highest during the afternoon to evening and lowest at dawn. The concentration of dissolved oxygen decreases with the rise of temperature.

In the present investigation, the dissolved oxygen ranged between 3.3ppm to 7.5 ppm with the average of 5.7 ppm. (Table 1). Ellis (1937), pointed out that the dissolved oxygen content in water for maintaining aquatic life in healthy condition must be 5mg/ltr. (5ppm) at 20°C.

The density of zooplankton population was high when the value of dissolved oxygen was also high. The positive correlation coefficient was found between dissolved oxygen and zooplankton from the statistical analysis ($r=0.4053$). This result have similarity with the findings of Bera et al. (2014).

g) Free Carbon dioxide

Free carbon dioxide plays an important role in photosynthesis for chlorophyll bearing organism. It provides the source of carbons, which is necessary for the growth of all green plants and indirectly for all other organisms. Hence, it performs a significant role in the food cycle of an aquatic ecosystem. High value of carbon dioxide was recorded during February 6th, which was a cloudy day causing lack of photosynthesis process and due to the respiration process of zooplankton the carbon dioxide content may have been high.

In the present study, carbon dioxide showed an inverse relation with D.O. When free carbon dioxide was high, the dissolved oxygen was found low and vice-versa. This may be due to the high amount of oxygen consumed by the animals in the pond, which in turn releases high amount of carbon dioxide.

Free carbon dioxide was absent during the months of October (Table: 1). Nasar (1977), in his investigation in a fresh water pond of Bhagalpur, India, free carbon dioxide was found to be mostly absent. Since, carbon dioxide was present irregularly; its correlation with zooplankton has not been worked out.

h) Alkalinity

As carbonate was absent in the pond water, the total alkalinity was only due to bicarbonate. When the pH value is in between 8.3 to 4.5 partially no carbonate is present, but bicarbonate may be present (Jhingran 1975). The present study was in accordance to it as the average pH value was 8.1. More or less similar behavior of alkalinity was reported by (Kushlan and Hunt 1979), in an alligator pond.

Bicarbonate was found regularly during the study period and value was also high. The bicarbonate alkalinity ranged from 116.6 to 185.3 ppm. Alikunhi (1957) reported that in highly productive water, the alkalinity ought to be over 100mg/l and the total alkalinity of the present pond water in average being 145.02 ppm. The pond water of present study was also found to be best for fish production. From the statistical analysis positive correlation between alkalinity and zooplankton was found i.e. $r = 0.2624$, and $P.Er = 0.1815$, $r > P.Er$. which shows that the value of correlation coefficient (r) was positively significant. This result shows similarity with the result obtained by Paulose and Maheshowri (2008).

i) Total hardness

Total hardness is the sum total of soluble calcium and magnesium present in water. It also includes the sulphates and chlorides of calcium and magnesium. Hardness in water indirectly affects the population density of zooplankton. The total hardness was found regularly in the pond water in the study period, it ranged from 5 to 21.67 ppm. and the average value was 16.21ppm. It reached maximum during August and minimum on November.

According to Swingle (1967), pond water having hardness of 15ppm or above may be considered suitable for the growth of the fish. So, this water may also be suitable for the fish culture practices. When the total hardness was high the value of pH, DO content and Total alkalinity was also high. From the statistical analysis, the correlation (r) between total hardness and zooplankton was found to be negative ($r = -0.499$) as reported by Paulose and Maheshwari(2008).

Zooplankton

Zooplanktons are the free-swimming microscopic animal component of aquatic systems. Zooplankton is the connecting link between primary producer and consumer of higher order in aquatic food webs. The zooplanktons feed on phytoplankton and it in turn are consumed by tertiary level of organisms. The zooplankton oscillations thus influence the production of organisms at both primary and secondary trophic strata. They provide varieties of indicator in an aquatic ecosystem.

In the water of the present pond, low genera diversity was observed with the total of 5 genera belonging to rotifer, copepoda and cladocera.

The species diversity in zooplankton is related to the alkaline nature of the water body. Very low pH is known to reduce species diversity and abundance (Goldman and Horne 1983). The average pH value of the pond water was 8.1 which was alkaline and good for the growth of planktons.

The total zooplanktonic abundance in the pond was 1944 no./liter during the study period, which showed the fortnightly density of 161.1 no./liter. Of the total, Copepoda(901 no./liter) uniformly dominated Rotifera (728 no./liter) and Cladocera (315 no./liter). The zooplanktons were more densely found during summer season and their density was low in winter.

In the present investigation, two genus of rotifer i.e. Keratella, Brachionus, one genus of Cladocera i.e. Daphnia and two genus of Copepoda i.e. Cyclops and Diaptomus were recorded.

Statistically zooplankton shows positive correlation with temperature, transparency,pH, D.O. , alkalinity and negative correlation with hardness. This result resembles the work of Paulose and Maheshwari (2007)

6. CONCLUSION AND RECOMMENDATION

6.1 Conclusion

The color of water varies according to the season, indicating the swarms of planktons in the pond. The temperature varied due to seasonal change and weather condition and it was observed that in high temperature, the population density of zooplankton was also high and vice versa.

It was observed the transparency was inversely proportional to the zooplankton production.

The pH value obtained was suitable for the aquatic life.

The density of zooplankton was positively co-related with DO, Alkalinity, pH, transparency and temperature.

Three major groups of namely Rotifera, Copepoda and Cladocera contributed the zooplanktonic assemblage of this pond named “Kamal Pokhari”. The zooplanktonic mass was dominated by Copepoda (46.6%), which was followed by Rotifera (37.64%) and the last by Cladocera (15.77%)

6.2 Recommendation

Following measures have to be adopted for the proper utilization of the pond

1. The study should be well fenced as people use the water for on the site for cleaning purposes.
2. The study on biomass of phytoplankton and zooplankton of the water for complete one year should be done to know the seasonal and complete picture of the pond
3. The local people must be made aware regarding the importance of water body so that they come forward to protect the pond and the environment.

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Table 1: Fortnightly variation of Physico-chemical Parameters of “Kamal Pokhari” Kamalvinayak, Bhaktapur, from 14th August 2012 to 6th February 2013

s.n	Date	Aug 14	Aug 30	Sept 13	Sept 29	Oct 13	Oct 30	Nov 17	Dec 3	Dec 19	Jan 4	Jan 21	Feb 6	Max.
	Parameters													
1	Nature of day	Bit Sunny	Partly cloudy	Sunny	Sunny	Sunny	Sunny	Sunny	Sunny	Cloudy	Cloudy	Cloudy	Cloudy	
2	Temprature (.c)	27	26.9	26	25.8	22	18	15	13	12.1	11.2	18.6	18	27
3	Depth (cm)	156	157	141	140	124	132	122	101	114	113	108	98	156
4	Transparancy (cm)	14.17	14.16	13.01	13.01	10.62	11.33	15.5	15.5	17	18.8	17	12.14	18.8
5	Transparency coefficient	0.09	0.11	0.11	0.13	0.16	0.15	0.13	0.12	0.1	0.12	0.1	0.14	0.16
6	Color of water	Brownish	Brownish	Greenish	Greenish	Greenish	Greenish	Light green	Light green	Light green	Colorless	Colorless	Colorless	
7	pH	8	8.6	8.8	9.8	8.8	8.9	7.4	7.2	7.5	7.4	7.4	7.5	9.8
8	DO (ppm)	5.3	6.5	7.4	7.5	7.5	6.6	6.4	5.4	4.8	4.4	4.1	3.3	7.5
9	Free Co2	5.3	5.5	5.5	5.5	-	-	5	2.9	5.5	7.3	6.6	8	8
10	Hardness	23.33	21.67	20	18.33	20	20	5	14.5	11.67	15	15	20	21.67
11	Alkalinity	137	132	168	183.3	183.3	185.3	124.6	161.3	123	119.3	106.6	116.6	185.3

Table 2: Fortnightly variation of Zooplanktons of “Kamal Pokhari” Kamalvinayak, Bhaktapur, from 14th August 2012 to 6th February 2013.

S. N	Date	Aug 14	Aug 30	Sep 13	Sep 29	Oct 13	Oct 30	Nov 17	Dec 3	Dec 19	Jan 4	Jan 21	Feb 6	Mar	Apr	Average	Total	%
	Zooplankton																	
A	Rotifers																	
1.	Keratella	27	40	67	107	54	-	27	94	40	27	-	-	107	27	40.25	483	24.97
2.	Brachio	34	27	46	42	14	3	18	12	14	4	-	-	46	4	20.41	24	12.

	nus					4											5	67
	Total	61	67	11 3	14 9	68	3 4	45	10 6	54	3 1	-	-	14 9	31	60.67	72 8	37. 64
B	Copepo da																	
1.	Cyclops	40	25 4	14	14	80	1 4	54	40	67	1 4	4 0	-	25 4	14	52.58	63 1	32. 64
2.	Diapto mus	27	94	14	27	14	-	27	27	40	-	-	-	94	27	22.5	27 0	13. 96
	Total	67	34 8	28	41	94	1 4	81	67	10 7	1 4	4 0	-	34 8	14	75.08	90 1	46. 6
C	Cladoce ra																	
1.	Daphnia	54	44	54	18	56	3 7	14	14	14	-	1 0	-	56	10	25.4	31 5	15. 77
	Total	54	44	54	18	56	3 7	14	14	14	-	1 0	-	56	10	25.4		15. 77
	G. total	18 2	45 9	19 5	20 8	21 8	8 5	14 0	18 7	17 5	4 5	5 0	-	45 9	45	161.1	19 44	