

1. INTRODUCTION

1.1. General Background

Only a tiny fraction of the planet's abundant water is available to us as fresh water. About 97.4% by volume is found in the oceans and is too salty for drinking, irrigation, or industry. Most of the remaining 2.6% that is fresh water is locked up in ice caps or glaciers or is ground water too deep or salty to be used. Thus, only about 0.014% of the earth's total volume of water is easily available to us as soil moisture, usable groundwater, water vapour and lakes and streams (Miller, 2002). The water that is not locked up as permanent ice is continually moving through various pathways in the atmosphere, biosphere and lithosphere, and this set of natural flows is called hydrological cycle (Simmons, 1981).

In ancient time the basic need of the people was water for drinking and later on for irrigation of their fields, and they utilized first visible water i.e. river and rain and later on they found the invisible water from spring and wells (Sharma, 1983). Freshwater is a finite and vulnerable resources needed by multiple stakeholders for a range of purposes like domestic water supply, irrigation, hydropower, and industrial production. The Hindu Kush-Himalaya (HKH) region is one of the largest storehouses of freshwater in the world and their mountains are the sources of major river systems that serve some 500 million people in South-Asia (Shrestha and Shilpakar, 2004).

In the case of Nepal with an area of 1, 47,181 sq. km has comparatively vast number of water resources. About 5.05% of total area is covered by rivers, lakes, ponds and reservoirs (Sharma, 1977). There are over 6,000 rivers and rivulets, which exceed a total length of 45,000km (CBS, 1989). It is drained by numerous rivers and these rivers depending on their sources and discharge can be classified into three types: major, medium and small rivers. The major rivers like Mahakali, Karnali, Narayani and Koshi are permanent and promising sources of water suitable for large scale irrigation and hydropower development. Medium rivers including the Babai, the West Rapti, the Bagmati, the Kamala and the Kankai are also perennial, but the seasonal discharges fluctuates widely and are originate in the mid hills. Small rivers are seasonable and have little or no flow during the dry season without surface; they are

suitable neither for year around irrigation nor for hydropower generation. Nepal is agriculture country and has few industries or industrial areas so approximately 80% of all people are engaged in subsistence agriculture.

Regarding the potable water the Shivapuri National Park (ShNP) is one of the sources of Kathmandu Valley. Shivapuri, located at the northern side of Valley established initially for watershed management in 1976, was later declared as Shivapuri National Park in 2002. It lies within three district of Central Development regions including the northern parts of Kathmandu, the southern parts of Nuwakot and the western parts of Sindhupalchowk. Twelve VDCs are of Kathmandu districts adjacent to ShNP. Similarly nine VDC's of Nuwakot district and two VDC's of Sindhupalchowk district which lies between 27°45'-27°52' N latitude and 85°15'-85°30' S longitude (Shrestha 2005). GIS shows that the ShNP covers an area of 95.22 km² (Amatya 1993 and HMG/FAO 1996). However, the Department of National Park and Wildlife Conservation (DNPWC) has shows it as 144 km² (DNPWC 2005). It stretches 8 to 10 km from north to south and 20 to 24 km from east to west. The highest point is the Shivapuri peak, which is 2,732 m above the sea level, which represent the second highest peak of Kathmandu valley.

Shivapuri National Park (ShNP) lies in the central-mid hills region of Nepal and climate ranges from subtropical to warm temperate, which are given in three climatic periods:

-) Pre-monsoon season; hot and dry season (mid February to mid June)
-) Monsoon season (June to September)
-) Post monsoon season; cold and dry season (October to mid January).

There are three climatologically stations at ShNP at Kakani, Sundarijal and Budhanilkantha. Rainfall of this region is more or less similar to Kathmandu valley in southern slope of Shivapuri hills. Nevertheless, it receives more rainfall than Kathmandu valley (Shrestha, 2005).

The area of Shivapuri covers the upper watershed of the Bagmati and Bishnumati River, which are the main water systems for Kathmandu valley. At Bishnudwar to Panimuhan site, Bishnumati and small streams of Panimuhan collect water into Panimuhan reservoir, located at Bishnudwar and Panimuhan respectively. The

reservoir tapped approximately 35.6 million liter of water per day (Shakya and Rai 1997). Beside that river has been considered as an open ecosystem, and generally the quality of water is defined by physical, chemical and biological. The physical and chemical quality of water depends on natural (geologic and environmental) and man-made factor (Sharma, 1997). Many physical and chemical variables of the streams are reflected in terms of the quantity and quality of benthic fauna (Sheng, 1990). Some of the important physical and chemical qualities of freshwater ecosystem are temperature of water, turbidity, pH, total dissolved solids (TDS), hardness of water, dissolved oxygen, free carbon dioxide, alkalinity etc. The abundance, diversity and distribution of benthic fauna are further related to various physicochemical and biotic factors (Dutta and Malhotra, 1986).

Therefore, water quality also involved the biological aspect like present of micro and macro organism in the water bodies'. Benthic-macro invertebrates or fauna are those fauna, which live their whole or part of life dwelling the bottom of the streams. Benthos means bottom dwelling and macro mean fauna, which can be seen with our naked eyes. They are important part of the water ecosystem and serve as link in food web decomposing leaves and algae, and fish and other vertebrates (Sharma *et al.* 2005). Whitten (1975) defined macro invertebrates as the invertebrates that range in size from 3-5mm and lived under log, rock, debris, sediments, and aquatic plants. Benthic invertebrates may offer a good opportunity to study the river water quality as this group confined to almost one part of the locality of river, they are easily sampled, manipulate, and relatively quick react to certain change in water environment (Kusza, 2004).

The aquatic insects like Placopteran, Ephemeropterans, and Tricopterans are generally taken to pollution intolerant fauna (WHO, 1976) so richness of these invertebrates indicates the better water quality but when population of non-biting midges and worms are higher which indicates the water is likely to be polluted. But Selwin (1974) on his own study and on the basis of reports of others concluded that Ephemeroptera, Tricoptera, and also the Coleoptera, Diptera and Odonates are tolerant to pollution and are found in wide range of water quality. He also considered planarians and Placopteran as pollution tolerant fauna. However, Mandaville (2002) reported that benthos had also disadvantages like they do not react to all impact and disturbance and their abundance and distribution are seasonally vary.

2. LITERATURE REVIEW

2.1. Water Quality

The study of freshwater habitat is known as Limnology. It is derived by a Greek word “Lime” means pool, marsh, or lakes so Limnology can be divided into two groups, i.e. “Lentic” or standing water habitats and “Lotic” or running water habitats. Adoni *et al.* (1985) explained Limnology as a new interdisciplinary science with multifarious dimension, which deals with the study of structural and functional attributes of the lentic freshwater environment and problem associated with them. In Nepal, very little attention has been paid on Limnology although a large number of freshwater bodies’ viz. lakes, reservoir, ponds, and rivers are present in the country (Yadav and Prasad, 1980).

Similarly, as above mentioned in chapter 1 the quality of water is determine by physical, chemical and biological parameters. The physical parameter like the temperature is one of the important parameter of water in river and temperature of cool water is generally more palatable than warm water. High water temperature enhances the growth of microorganisms and may increase taste, odour, colour, and corrosion problem (WHO, 1993). Temperature also affects the dissolved concentration and the response of aquatic organism to chemical toxins. Most aquatic animals are able to cope with the decrease availability of dissolved oxygen in the absence of additional stress but the occurrence of chemical pollution may have serious consequences (Trevlin and Burgis, 1979). Another parameter, pH is also one of the important operational water quality parameters. The optimum pH required will vary in different supplies according to the composition of the water and the nature of the construction material used in the distribution system but it is often in the range 5.0-8.5 (WHO, 1993). Water velocity or current is also factor of major importance in running water, which controls the deposition of organic matter at the substrate of the stream, oxygen concentration, abundance of planktons and other ionic composition of water. Many macro-invertebrates possessed an inherent need for current, either because they relied on it for feeding purpose or respiratory requirements (Hynes, 1970).

Total dissolved solids (TDS) as major parameter can have an important effect on the taste of drinking water. The palatability of water with a TDS level of less than 600 mg/L is generally considered good (WHO, 1993). The major effect of suspended solids on benthic communities is when the solids settle to blanket the stream bed, when this occur the lithophilous fauna such as the mayflies e.g. *Ecdyonurus*, stoneflies and caddiesflies is seriously affected and may be replaced by a silt community including Oligochaetae, Pulmonate, snails and chironomid larvae (Hawkes, 1979). On other hand, hardness is also an important chemical parameter. Hardness of water is due to the presence of Ca^{++} and Mg^{++} . The taste threshold for the Ca^{++} is in the range of 100-300 mg/L, depending on the associated anion, and the taste threshold for Mg^{++} is probably less than that for Ca^{++} . According to W.H.O., consumers tolerate water hardness in excess of 500 mg/L.

The dissolved oxygen (DO) is another controlling parameter in water. Fresh water always contains some dissolved oxygen. It is this dissolved gas which most of the aquatic animal breathe. Fresh water streams (springs, rivulets, creeks, brooklets etc) and rivers change over their course from being narrow, shallow, and relatively rapid to become increasingly broad, deep and slow moving. Consequently, chemically the upper reaches of lotic environment are rich in oxygen. But as the water moves downstream and becomes more sluggish, the oxygen level tends to drop (Kormondy, 2002). Depletion of DO in water can encourage the microbial reduction and reduction of sulfate to sulfide, giving rise to odour problem. It can also cause an increase in the concentration of ferrous iron in solution (WHO, 1993). Because of the small depth, large surface exposed to the air, and constant motion, streams generally contain an abundant supply of oxygen, even when there are no green plants. For this reason, stream animals generally have narrow tolerance and are especially sensitive to reduced oxygen. Therefore, stream communities are especially susceptible to and quickly modified by any type of organic pollution, which reduces the oxygen supply (Odum, 1971). On other hand, cold water, which is well stirred up by passing over waterfalls, contains much oxygen. Warm and stagnant water contains less. Different animals have different oxygen requirement, which often helps to explain why any particular species is only found in one definite type of fresh water.

Another parameter of water include biological parameter which involve the benthic-macro invertebrates include aquatic insects like Ephemeroptera, Placoptera, Tricoptera, Diptera, Odonata etc., and other like clams, snails, crayfishes, and aquatic worms. They help to maintain the health of the water ecosystem by eating bacteria and dead and decaying plants and animals (Kaspal *et al*, 2005). Some like beetles and bugs also acts as scavengers in keeping the water fresh by removing the decayed leaves and other faecal materials. As some of them are fairly short lived and remain in one area during their aquatic phase, they are particularly good indicators of localized condition of river for short period time (RHP, 2004) and are also important in increasing the oxygen percentage of water. The total eleven insects order are aquatic of which 4% of the total insect fauna pass their life in water (Ward and Whipple, 1966). The large number of aquatic animals belongs to phylum arthropod. The young stages in some orders have the adults, and differ from them only in their smaller size and lack of complete wings, and genital organs, they called nymphs like stoneflies, mayflies and some orders have aquatic larvae like caddis flies (Macan and Worthington, 1972).

Another important feature of benthic organism is a dominance of one group adversely affecting abundance of the other taxonomic groups thus indicating a competition for food in the stream. Since cold-water streams are inherently poor in biological productivity the available quantity of food is limited and hence a keen competition (Sehgal, 1991). The various species of zooplankton are often confined to relatively restricted zones and so exhibit an uneven distribution in a vertical direction. The types of movement observed are numerous and depend not only on the species and locality under consideration but also on season, age, sex, etc (Hutchinson, 1967).

In flowing water, zooplanktons live on the bottom or else attached to a substrate, to which they cling fast in order to counter the drag of the current. For the same reason most flowing water animals in mountain streams inhabit the areas out of the main streams, that is dead water between the pebbles and river bed, plant clusters, or as far as the narrow passage allows, the cavity regions of the porous rock under the streambed. The attachment to the substrate occurs by means of sticky cell walls by the outgrowth of basal cell projections, gelatinous pads or branching of the basal cells

to a rhizoid cell, which can fasten itself to the small irregularity of stone (Schwoerbel, 1991).

2.2. Water Pollution

Water pollution may be defined as the presence of any foreign substance (organic, inorganic, radiological or biological) in water which tend to degrade its quality to constitute a hazard or impure the usefulness of the water (Camp and Meserve, 1963). Such contamination of water or such alternation of the physical, chemical or biological properties of water or such discharge of any sewage or trade effluent or of any other liquid, gaseous or solid substance into water (whether directly or indirectly) as may or likely to create a nuisance. Such water is harmful or injurious to public health or safety or to domestic, commercial, industrial and agriculture uses (Sapru, 1989).

The middle hills of Nepal are heavily populated compared to other parts; the quality of water deteriorates as river passes through the major settlements, whereas in sparse settlements water pollution is reasonably low. Solid Waste is not treated before they are drained it in the river-system and water quality is beyond the permissible limited (Sharma, 1997). Water pollution may be both by natural and/or anthropogenic factors, however human being is much more responsible for creating polluted water. The contamination of surface water with many pathogens (bacteria, viruses etc.) leading to chronic health problems like cholera, typhoid, amoebic dysentery and other intestinal tract diseases. Nearly 80% of the world's diseases more so in developing world, can be linked with water (Khoshoo, 1989).

2.3. Study reveals physicochemical parameters

Many research works has been done in the past relevant related to the present investigation of water quality. Brehm (1953) was the first to study benthic fauna from Kalipokhari in eastern Nepal.

Cairns and Dickson (1971), Learner *et al.* (1971), Olive and Damback (1973), and Hawkes (1979) concluded that some of the chironomids are the some inhabitants of

polluted waters, rich in nutrients and poor in oxygen content and are also used as biological indicators of the pollutions.

Upadhaya and Rao (1982) studied chemical parameters in six rivers and rivulets- Dhobikhola, Manohara, Nakkhukhola, Balkhukhola, Bishnumati and Bagmati through out one year. In their study a wide seasonal variation was noted in water chemistry. The Bagmati and Bishnumati River had higher total dissolved solids (TDS) values, specific conductance and concentration of Na^+ , K^+ and Cl^- .

Khadka (1983) studied on major ions in Bagmati River near Pashupatinath Temple and reported that concentration of major ions especially Na^+ and Cl^- was high in sample collected down stream of the temple than upstream.

Khatri (1986) studied on water pollution of Bagmati and Vishnumati River of Kathmandu and concluded that the moment Vishnumati join the Bagmati, it is more or less equally polluted like Bagmati River.

DISVI and RONAST (1988) made study on river had definite zones of pollution depending on proximity to urban area and industry. The Bagmati was classified into six zones- a healthy zone, in moderate up stream zone, pollution zone, polluted zone, moderate downstream polluted zone, recovery zone and clean zone.

ENPHO (1992) carried out water quality testing activity for installing shallow tabewell and gravity flow systems in some areas in Terai in 1990. Some tested physicochemical parameters like pH, conductivity, Mn, Fe, etc. and result showed that concentration of most of the chemical constituent in the analyzed samples were found to be within WHO permissible limit. Mn and Fe were found in excess of WHO permissible limit in 57 % and 44 % respectively.

Sharma (1998) studied the ecology of Koshi River in Nepal and India (North Bihar). He reported that river water was quite suitable and possessed high degree of ecological efficiency and enormous potential for biotic development. During study, he observed alkaline pH, low free carbon dioxide, high dissolved oxygen (5.88 to 7.33mg/l) and total alkalinity of 50 mg/l.

Ghimire (1999) evaluated water quality of Bagmati River of Kathmandu by investigating different physicochemical and heavy metal parameters. According to him, most of the pollution parameters were found to be increased towards downstream. Most of the heavy metals in the river water were well within threshold limit but Fe and Mn were present in significant amount. In this study the water quality in term of heavy metal was also assessed by examine macrophytic plants of the river.

2.4. Study reveals biological parameters

Atkinson (1882) made a comprehensive study in the form of 'Fauna of Himalayas' (North-West Panjab) and Mt. Makalu (Nepal). In same year, Atkinson described stoneflies of Bagladesh, India and South-East Asia.

Godwin-Austen (1910) was, perhaps the first man to make a report on mollusk from Nepal, collecting from Kathmandu Valley. A review of available literature on mollusks of Nepal that there exist scanty of sufficient information. However, contributions of Godwin-Austen (1910), Majupuria (1981-82), Yadav *et al.* (1980) and Pfeiffer *et al.* (1999) on mollusk of Nepal are few to be mentioned. Recently Subba and Ghosh (2000 and 2001), Subba (2003), have listed a number of freshwater as well as terrestrial mollusks collected from Eastern, Central, Far-Western Nepal.

Asahida (1952) studied 100 specimens of dragonfly nymphs belonging to 20 species from Nepal Himalays. He in 1963 made a good comprehensive study of the Odonata of Nepal and India collected by Yamada in 1961; in 1964 he studied the Odonata collection made by the Chiba University Rolwling Himal Expedition in 1983; in 1964 he studied more Nepalese Odonata collected by Botanical Expedition of Tokyo University.

Quentin (1970) studied the Odonata of Khumbu Himal in Nepal. Beside that Mishra (1976) presented a paper entitled 'Fauna of Nepal including insects' at Natural Science Seminar organized by Tribhuvan University, Kathmandu.

Dubey (1971) also studied about the mayflies of Nepal; Mc. Author and Barnas (1985) reported Baetis mayflies to be first organism to colonize bare or rocky substra.

Kiauta (1972) made observations on title known dragonfly '*Macromia moorie*' from Nepal. Some species of dragonflies and nymphs of mayflies of Nepal are also reported by Kihara (1955).

Stanley (1975) has also worked on the stoneflies of North-West Himalays in India and Mt. Makalu in Nepal Himalays. Ueno (1952) described 24 nymphs belonging to 6 genera and 3 families of mayflies.

Malla *et al.* (1978) carried out the taxonomic studies on aquatic insects of the Kathmandu Valley and found 37 new generic and species records from Nepal.

Sharma (1988) study about the physicochemical parameters on distribution and abundance of zoo benthos in the river Bagmati and concluded the order dipterans are the most dominant group which are followed by Oligochaetes and Ephemeroptera.

Vaidhya (1990) and Tennyson *et.al.* (1990) studied the water quality of macro invertebrates of Bishnumati River. The stream after traveling through a length of 1.5 km receives different tributaries and flow as Bishnumati River from Budhanilkantha onward. The chemical quality of Bishnumati River was found quite better than other river in Kathmandu valley. It indicates that the chemical percentage of water flowing from the protected area of Bishnumati River is suitable for domestic use.

Basaula (2000) done his investigation on ecological study of Shivapuri Hill and observed Tricoptera, Ephemeroptera and Placoptera is the most dominant group.

Chakrabarti (2001) studied physicochemical parameters and abundance and distribution of zoobenthos in Bagmati River and concluded the deteriorated ecological balance of the river.

Vaidya, (2002) studied about the macro-invertebrates as measures of water quality of Vishnumati stream and concluded that Ephemeroptera, Tricoptera, and also the Coleopterans, Dipterans and Odonata are tolerant to pollution and are found in wide range of water quality and added planarians as pollution tolerant fauna.

Roy (2006) done her research work on aquatic insects of Shivapuri National Park as a tool for biological assessment of headwater streams of Sundarijal drinking water supply and report the 66 aquatic insects and found order Tricoptera is the most diversified insect followed by Ephemeroptera, Diptera, Placoptera, Odonata, Coleoptera, Hemiptera and Neuroptera.

3. METHODS AND METHODOLOGY

3.1. Study Area

Shivapuri National Park (ShNP) with an area of 144 km² is situated on the northern fringe of Kathmandu Valley and lies about 12 km away from the capital city. The area was gazette as the country's ninth national park in 2002. Shivapuri lies in a transition zone between subtropical and temperate climates. The vegetation consists of a variety of natural forest types including pine, oak, rhododendron etc, depending on altitude (DNPWC, 2006).

Water from the watershed is tapped from different streams and channeled in reservoirs. Shivapuri is one of the main sources of drinking water for Kathmandu Valley. Among them Sundarijal reservoir tapped maximum water for Valley. The area of Shivapuri covers the upper watershed of the Bagmati and Bishnumati River, which are the main water system for Kathmandu valley. Tributaries of the Likhu and Shindhu Khola, which drain the northern side, also fall within the park. There are many rivulets that are tributaries different spring fed rivers at different sites.

3.1.1. River System

) Kakani to Gorje site

Rivers, streams and reservoirs are at the western and northern sites of national park with Thule khola, Alle khola, Bounde khola and Balaju khola and Bhandre khola located at Panchmane, Allegaun, Boundegaun and Gahirigaun respectively. There collect water into Balaju reservoir that approximately 27.4 million liter of water per day.

) Bishnudwar to Panimuhan site:

Bishnudwar and Panimuhan or Charchhaare khola lies at this site. These khola collect water into Panimuhan reservoir, located at Bishnudwar and Panimuhan. The reservoir tapped approximately 35.6 million liter of water per day.

) Sundarijal site:

Bagmati, Shyalmati, Nagmati, Chilaune khola, Okherna khola are streams at Sundarijal site that collect water into Sundarijal reservoir approximately 163.8 million liter of water is tapped per day (Shakya and Rai 1997).

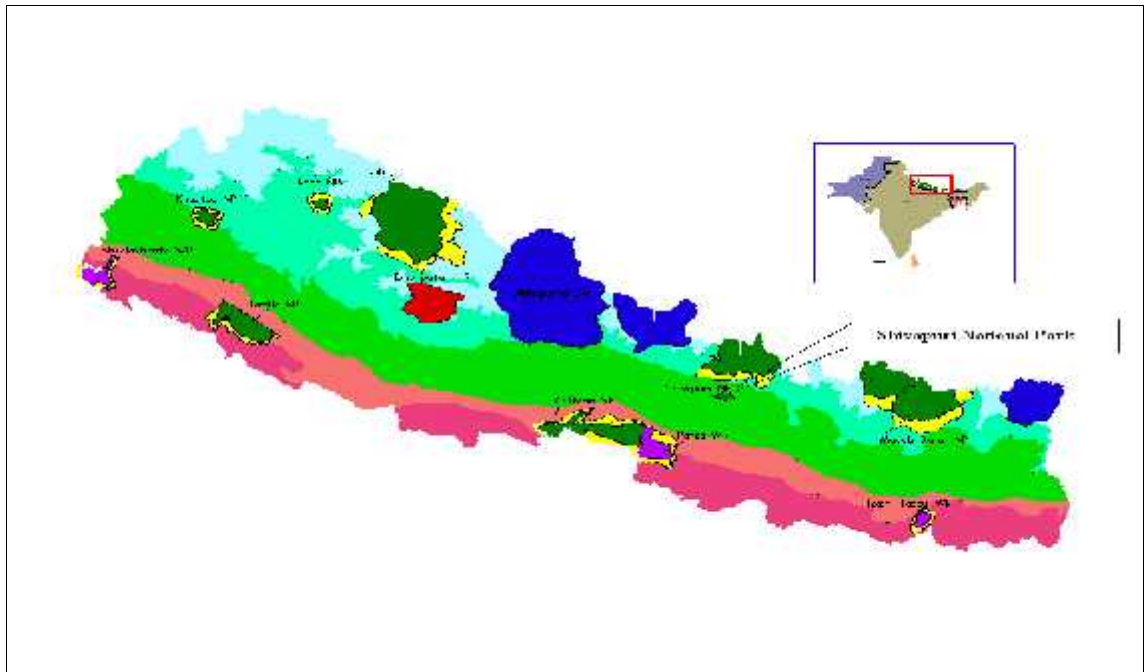


Figure 1: Location of Shivapuri National Park

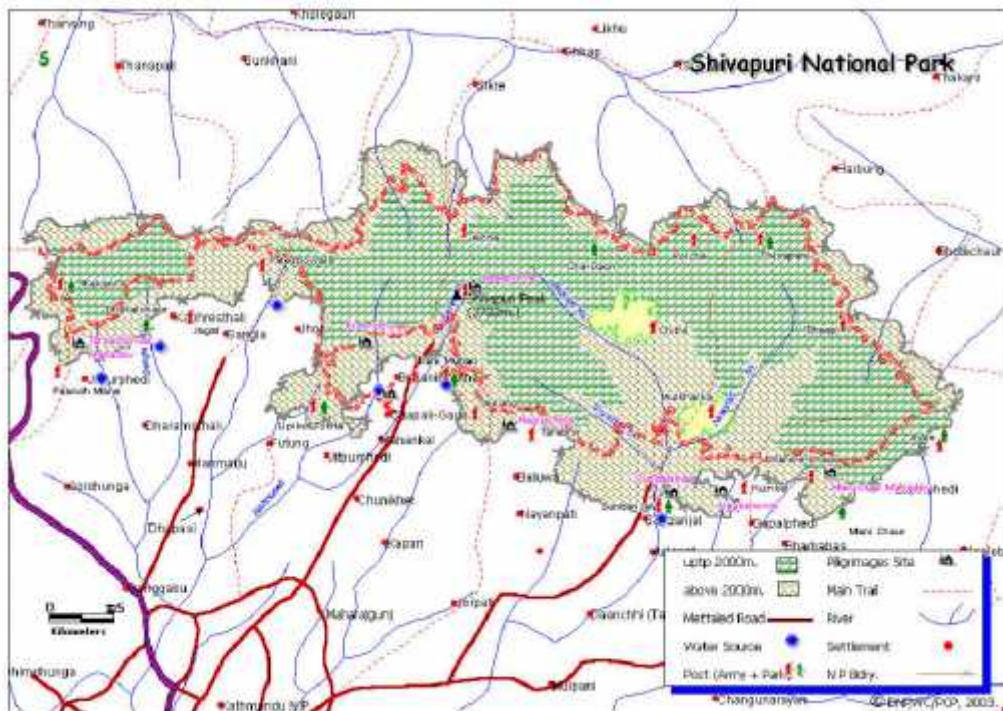


Figure 2: Shivapuri National Park



Figure 3: Study sites at Panimuhan stream

3.2. Site Selection

The study area was selected at the southern hill of ShNP i.e. Panimuhan and locally called it as Muhan pokhari which is upper side of Budhanilkantha and 12 km away from Ratna park. For this study, three sites were selected site 1 and site 2 was inside ShNP and site 3 was located outside the national park. Water qualities of Panimuhan were analyzed during the year of 2007 from the month of July to December over the period of six month. The area was divided into three different sites on the basis altitude.

Site 1

The first site was inside the national park and was 1 km away from the main entrance gate at the height of 1700 m. This area was small stream covered by dense forest of *Schima wallichiana*. The velocity of water was speed with clear and transparent. The substratum was composed of cobbles, gravels with big boulders numerous fallen decaying leaf litters.

Site 2

The second site was located in the main entrance gate of park at height of 1610 m. it was a reservoir gets poured water from site 1 was poured in this reservoir. The site 2 was open area exposed to direct sunlight and from this site drinking water draining into reservoir of Budhanilkantha and Pani Pokhari. Finally water supplies to Kathmandu valley. This site was surrounded by *Schima wallichiana* and *Alnus* sp. The substratum was composed of sandy bottom, no big boulders, and gravel with speed current of water and submerged aquatic plants.

Site 3

The third site was outside the national park at the height of 1500 m and 1 km south from the main entrance gate of the park. The site 3 was open place which run through the mid of residence area so the water get mild polluted. The sources of pollution were domestic wastes, tins, cans, bottles, papers and plastics. The water used as the source for washing clothes and bathing. The substratum was composed of sand, cobbles, and gravel with large boulders and submerged aquatic plants. The water was over flown of the site 2 and got semi dry during winter seasons.

3.3. Rationale of the study

Shivapuri Watershed Area is one of the main water resources of Kathmandu valley. All these water resources are passed through the mid of valley but due to countless anthropogenic activities make these potable water into unhygienic and unfit for used in any purposes. Due to all these activities water quality is degraded and causes many water borne diseases. In Panimuhan stream and Bishnuduwar, 35.6 million liter water is tapped per day for Kathmandu Valley. This water is collected in Panimuhan reservoir and first supplied to Budhanilkantha reservoir for purification and further and finally supplied to Panipokhari reservoir where final purification is done and supply it for Valley. From Panimuhan reservoir, everyday for four hours they supply water to local people living up to Budhanilkantha. They supply water to people by purifying chemical called potash without any long process of purification.

Beside that aquatic insects reflect the effects of physical and chemical impacts, where they are regarded as good indicators of pollution and used as bio-monitoring tools.

-) Benthic-macro invertebrates are highly affected by physical, chemical, and biological condition of streams.
-) Some of the benthos shows limited patterns of life, limit of pollution tolerant so shows the short term and long-term pollution condition.
-) They maintain the overall biological productivity and food web in the streams.
-) Their long life cycle allows studies conducted by aquatic ecologist to determine any decline environment pollution.

The physicochemical components of water are very crucial to analyzed before it is utilized and similarly biological aspects like benthos plays a fundamental role in maintaining health of water. Therefore, due to all these objectives it becomes necessary for analyzing drinking water supplies for valley and the study was undertaken.

3.4. Objectives

The prime objectives of this study were:

3.4.1. To assess the water quality analyzing basic physicochemical parameters including:

-) The velocity
-) Temperature
-) Hydrogen ion concentration (pH)
-) Total solids (TS)
-) Total dissolved solids (TDS)
-) Total suspended solids (TSS)
-) Dissolved oxygen (DO)
-) Free carbon dioxide
-) Total alkalinity
-) Total hardness (Calcium and Magnesium)

3.4.2. To explore the benthic macro-invertebrates as pollution indicator of three selected sites of Panimuhan stream.

3.5. Physical Parameters of water

The physical analysis of water was performed in the field.

3.5.1. Velocity (m/sec)

The surface float method was used to measure the velocity of the water current. The following formula was used for calculation:

$$\text{Velocity} = \frac{\text{Distance fixed (m)}}{\text{Time taken (sec)}} \quad (\text{Adoni } et.al. \text{ 1985})$$

The velocity was measured in meter/second (m/sec)

3.5.2. Temperature (°C)

The standard mercury thermometer (graduated to an accuracy of 0 ± 1 °C) was deep in the stream and temperature was recorded in Celsius scale.

3.5.3. Hydrogen ion concentration (pH)

Hanna instruments pH meter was used to measure the sample water collected in a 250ml beaker. For calibration, freshly prepared buffer was used. The pH meter was immersed in the buffer solution up to the maximum level in pH 7.0 and minimum level in pH 4.0 and the reading was allow to stabilized and finally by using a small screw driver to calibrated the trimmer to read pH 7.0 and pH 4.0.

3.5.4. Total solids (TS)

Evaporating dish made up of porcelain of at least 100 ml capacity was taken and kept in oven for an hour and for about half and hour was kept in desiccator for moisture absorbed. Then initial weight of evaporating dish was taken. After that 100 ml of unfiltered sample was evaporated in evaporating dish on a water bath having temperature not more that 90 °C. Residue was heated at 103-105 °C in an oven for an hour and the final weight after cooling in desiccator was taken.

$$\text{Total Solid (mg/L)} = \frac{A-B \times 1000 \times 1000}{V}$$

Where,

A= Final weight of the dish in gram

B= Initial weight of the dish in gram

V= Volume of sample taken in ml

3.5.5. Total dissolved solids (TDS)

A total dissolved solid was determined as the residue left after evaporating of the filtered sample.

$$\text{TDS (mg/l)} = \frac{\text{A-B} \times 1000 \times 1000}{\text{V}}$$

3.5.6. Total suspended solids (TSS)

A total suspended solid was determined by the difference between the total solids and total dissolved solids.

$$\text{Total suspended solids (TSS)} = \text{total solids (TS)} - \text{total dissolved solids (TDS)}$$

3.6. Chemical Parameters of water

Sample collection: The collection of samples was done very carefully as all interpretation were based on the analysis report. The sample water about 2 liters was collected in clean plastic bottles and was sealed in mouth. A very important point was that the water was poured in the plastic materials by the plastic cup. No floating materials was entered the bottles (Kudesia, 1985).

3.6.1. Dissolved oxygen (D.O.)

The dissolved oxygen content of river water was determined by using Winkler's method. For the determination of dissolved oxygen, BOD bottle of 300 ml was filled with the sample water and stopper was placed carefully without trapping air bubbles. The dissolved oxygen was fixed immediately by adding 2 ml of manganous sulphate (MnSO_4) and 2 ml of alkaline potassium iodide (KI) using separate pipettes. The stopper was then replacing taking care to exclude the air bubbles. The bottle was shake thoroughly for well mixing. A brown precipitate was seen which was allow to settle down. Half of this experiment was done in the field to trap the dissolved oxygen of water and was brought in the Natural Product Research Laboratory of Nepal Academy of Science and Technology (NAST). Next, the precipitate was allowed dissolving, by adding 2 ml of concentrated sulphuric acid (H_2SO_4). 50 ml of sample

water was taken in conical flask and was titrated against sodium-thio-sulphate solution (0.025 N) with few drops of starch solution (an indicator) and the sample was turn into ink-blue. Titration was continued till the blue colour disappears. The volume of titrant used in getting the end point was noted. D.O. was calculated in mg/l by using the following equation.

$$\text{Dissolved Oxygen} = \frac{(\text{ml} \times \text{N}) \text{ of titrant} \times 8 \times 1000}{V_1} \times \frac{V_2 (V_1 - V)}{V_1}$$

Where,

V_1 = Volume of BOD bottle

V_2 = Volume of titrated sample

V = Volume of MnSO_4 and KI added.

3.6.2. Free Carbon dioxide (mg/l)

The sample of 50 ml was taken in a clean conical flask and a few drops of phenolphthalein as indicator was added to it. The sample does not become red; it was immediately titrated against sodium hydroxide (NaOH-0.05 N) solution stirring with a glass rod until pink colour appeared. The mean volume of three readings at each side was taken and converted later on into mg/l by using following equation:

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

3.6.3. Total alkalinity (mg/l)

For total alkalinity, 50 ml of sample was taken in a conical flask and 2 drops of phenolphthalein indicator was added to it. The sample remained colourless which indicated that the phenolphthalein alkalinity was zero. Immediately 2 or 3 drops of methyl orange was added to the sample and titrated against HCl (0.1 N) until the yellow colour changes to pink at end point. The volume of HCl consumed was noted which give the total alkalinity. Three readings were noted and the mean was taken which later was converted into mg/l by using following simple equation.

$$\text{TA as CaCO}_3, \text{ mg/l} = \frac{\text{B} \times \text{N of HCl} \times 1000 \times 50}{\text{ml. of sample used}}$$

Where,

B = ml of total HCl used with phenolphthalein and methyl orange

TA = Total alkalinity

3.6.4. Total Hardness

For this, 50 ml. of sample was taken in a conical flask in which 2 ml of ammonia buffer solution was added and stir for through mixing. To this 200 mg of Erichrome Black T as indicator was added and shaken well. Then the sample was titrated against standard Ethylenediamine tetraacetic acid (EDTA) solution of 0.01 N till the colour from wine red to blue. The volume of EDTA consumed reading at each stage was converted into mg/l by using following equation.

$$\text{Total hardness (mg/l)} = \frac{\text{ml. of EDTA used} \times 1000}{\text{ml. of sample used}}$$

3.6.5. Calcium Hardness

To determine calcium hardness, 50 ml of sample was taken in a conical flask and 2 ml of sodium hydroxide (0.01 N) solution was added to it. Next, 200mg of Mureoxide indicator was added and its colour change to pink. The solution was titrated against standard Ethylenediamine tetraacetic acid (EDTA) solution of 0.01 N till the pink colour changed to purple at the end point. The mean volume of EDTA consume in three readings were converted into mg/l by using the following equation.

$$\text{Calcium (mg/l)} = \frac{(\text{ml} \times \text{N}) \text{ of EDTA} \times 400.8}{\text{ml. of sample used}}$$

3.6.6. Magnesium Hardness

Magnesium hardness was determined by using the following formula:

$$\text{Magnesium (mg/l)} = \frac{(\text{Total hardness} - \text{Calcium hardness}) \times 400.8}{\text{Vol. of sample} \times 1.645}$$

3.7. Sampling of benthic macro-invertebrates

At each site, three samples were taken in each month. For the collection of macro-invertebrates a quadrat size of 20×20cm was used. Thus a total of 54 samples were taken during study period.

The pebbles and gravels were turned over by lifting vertically upward with hand and macro fauna were collected with the help of pointed forceps and brush where

precaution was taken without damaging the fauna. Small pebbles and mud were sieved with the help of a sieve of 500 μ . The larger sized stones were removed by hand. The sieved samples were transferred into marked polythene bags along with little amount of 80% alcohol. The collected samples were brought to the Natural Product Research laboratory of Nepal Academy of Science and Technology (NAST) and the sorting of samples were carried out in white enamel trays by using the fine forceps, brushes, droppers etc. Animals were identified into possible taxonomic level.

3.8. Statistical Analysis

3.8.1. Chi-Square (χ^2) test (variance to mean ratio)

The values of χ^2 were calculated by the following equation:

$$\chi^2 = \frac{S^2 (n-1)}{\bar{X}^2} \quad (\text{Brinkhurst, 1971})$$

Where,

S^2 = Variance

\bar{X} = Arithmetic mean

3.8.2. Variance (S^2)

$$S^2 = \frac{\sum x^2 - \frac{(\sum x)^2}{n}}{n-1} \quad (\text{Berg, 1938})$$

Where,

n = no. of samples

x = counts of the sampling units

3.8.3. Arithmetic Mean (\bar{X})

$$\bar{X} = \frac{\sum x}{n} \quad (\text{Bailey, 1959})$$

Where,

$\sum x$ = Sum of all observations

n = Total number of observations

3.8.4. Correlation Coefficient (r)

For correlation analysis of physicochemical of water and benthic fauna distribution methods described by Cox (1981) and Gupta (1982) were used.

$$r = \frac{N \cdot XY - X \cdot Y}{\sqrt{N \cdot X^2 - (X)^2} \sqrt{N \cdot Y^2 - (Y)^2}}$$

Where,

N = Number of observation

X = Summation of sampling unit X

Y = Summation of sampling unit Y

The value of 'r' lies between +1 to -1

r = +1, Perfect positive correlation i.e. increase in one variable is accompanied by the increase in the other

r = 0, No correlation

r = -1, Perfect negative correlation i.e. increase in one variable is associated by decrease in the other

3.8.5. Species Diversity (\bar{D})

For the species diversity, the most widely used index is Shannon-Wiener diversity index (Washington, 1984). Evenness is also measured.

$$\bar{D} = - \sum p_i \log_e p_i$$

Where,

\bar{D} = Diversity index

$$p_i = \frac{n_i}{N}$$

n_i = number of individual in species

N = total number of individual in the sample

3.8.6. Evenness index (e)

$$e = \frac{\bar{D}}{\log s}$$

The value of Evenness index (e) generally lies between 0-1. If the value lies near to 0, the diversity is said to be uneven and if value lies near to 1, the distribution is said to be even.

4. RESULTS

4.1. Physicochemical Parameters of water

Some of the physicochemical parameters of water like velocity, temperature, pH, total solids, total dissolved solids, total suspended solids, dissolved oxygen, free carbon dioxide, total alkalinity, total hardness, and calcium and magnesium hardness were analyzed over the period of six months from July to December 2007 in Panimuhan stream.

The overall average velocity of water current of all sites was found to be 0.58m/sec during the period of six months. The average temperature, pH, TS, TDS and TSS of water were recorded 16.26 °C, 7.22, 1282.33 mg/l, 1044.44 mg/l and 238.88 mg/l respectively. Similarly average DO was 7.49 mg/l, free CO₂ was 6.84 mg/l, and total alkalinity was recorded 48.33 mg/l. The average total hardness was 15.66 mg/l and 4.71 mg/l and 0.94 mg/l were the average Ca and Mg hardness of the water (Table 1).

Table 1: Average fluctuation on physicochemical parameters of three sites during the study period of six months.

S.N.	Parameters	Site 1	Site 2	Site 3	Average
1.	Velocity (m/sec)	0.515	0.79	0.44	0.58
2.	Temperature (°C)	14.68	16.1	18.01	16.26
3.	pH	6.83	7.11	7.13	7.22
4.	TS (mg/l)	1150	1383.33	1316.66	1282.33
5.	TDS(mg/l)	983.33	1133.33	1016.66	1044.44
6.	TSS(mg/l)	166.66	250	300	238.88
7.	Dissolved oxygen (mg/l)	7.36	7.76	7.36	7.49
8.	Free CO ₂ (mg/l)	8.06	6.6	5.86	6.84
9.	Total alkalinity (mg/l)	43.33	55	46.66	48.33
10.	Total hardness (mg/l)	13.66	13	20.33	15.66
11.	Ca hardness(mg/l)	4.26	3.6	6.27	4.71
12.	Mg hardness(mg/l)	0.72	0.97	1.13	0.94

4.1.1. Water velocity (m/sec)

The water velocity of 0.66 m/sec at site 1 was found highest in the month of July and found lowest in the month of December i.e. 0.35 m/sec (Table 24). Similarly at site 2 the water velocity of this site was found 1.0 m/sec as highest during the months of July and August and 0.62 m/sec lowest velocity of water during the month of December (Table 25). And at site 3 the velocity of water was highest during the month of July i.e. 0.66 m/sec and lowest during the month of December i.e. 0.22 m/sec (Table 26). The average water velocity of site 1 was 0.51 ± 0.03 m/sec during the study period and site 2 had 0.79 ± 0.07 m/sec. Similarly the average velocity of site 3 was found 0.44 ± 0.05 m/sec (Table 1). The average water velocity was highest at site 2 than site 1 and site 3 because as already mentioned on site selection that site 2 was without any big boulders and substratum for obstacles for the current of water than site 1 and site 3.

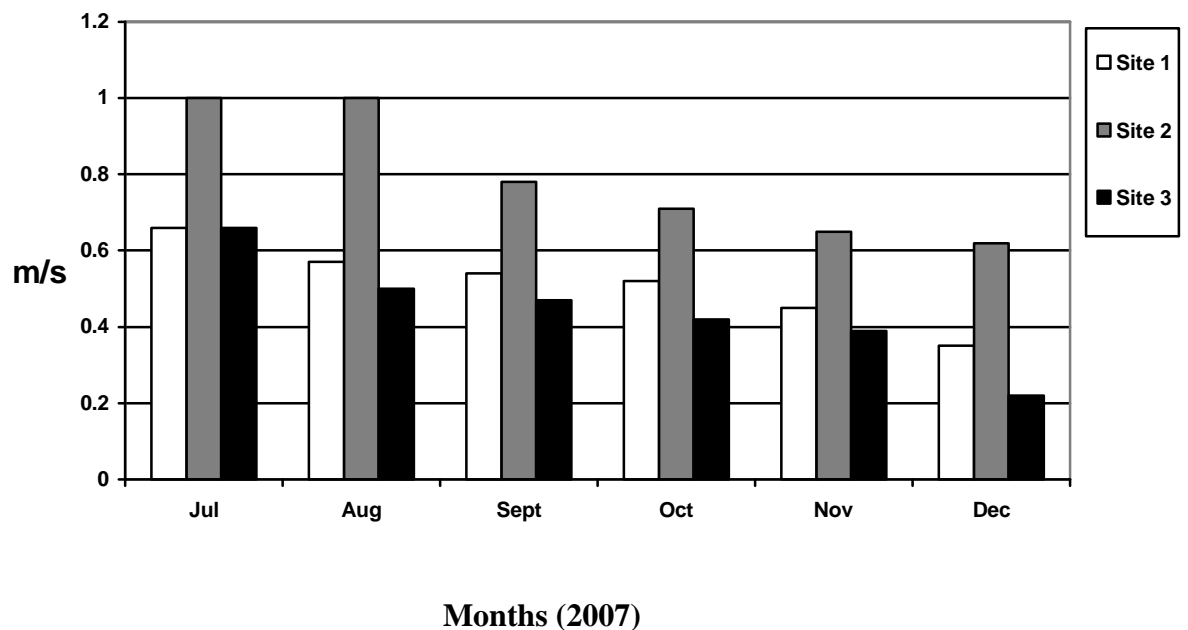


Figure 4: Variation in velocity of water during the study period of six months

4.1.2. Temperature (°C)

The water temperature at site 1 during the study period was found highest in August i.e. 18 °C and 9.9 °C as lowest in the month of December (Table 24). At site 2 the temperature of the water was found highest 20.5 °C in the month of August and lowest i.e. 10.9 °C in December (Table 25). The highest temperature of the water was

21 °C at site 3 in August and lowest 14.5 °C during the month of December (Table 26). The average temperature of water at site 1 was recorded 14.68 ± 1.29 °C and site 2 water had the average temperature of 16.1 ± 1.56 °C similarly the average temperature of water of site 3 was found to be 18.0 ± 1.04 °C (Table 1). The overall average temperature of water was highest at site 3 than site 1 and site 2 because among three sites, site 3 was at the lowest altitude i.e. 1500 m than site 2 and 1 and was also pollutant.

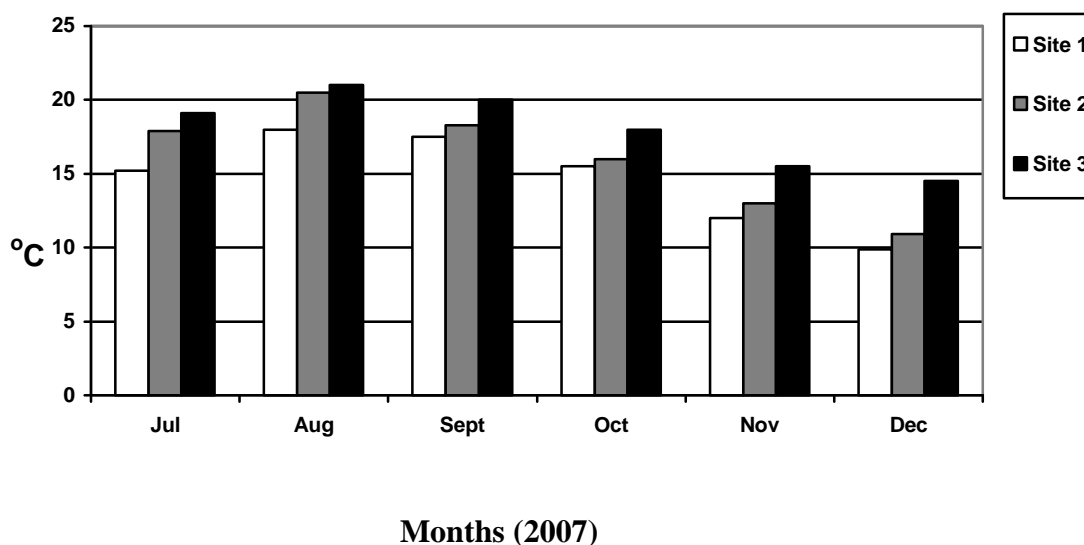


Figure 5: Variation in temperature of water during the study period of six months

4.1.3. Hydrogen ion concentration (pH)

The water of all the sites was found alkaline during whole study period. At all sites hydrogen ion concentration (pH) was highest during the month of July. At site 1 the highest pH of 7.2 was found during the month of July and lowest in the month of November i.e. 6.4 (Table 24). At site 2, 7.5 was highest pH during the month of July and lowest in the month of November i.e. 6.7 (Table 25) and 7.4 was highest pH in month of July and lowest 6.6 on the month of November (Table 26). The average pH of site 1 was found between 6.83 ± 0.01 , the average pH of site 2 was between 7.11 ± 0.137 and similarly 7.4 ± 0.12 was the average pH of site 3 (Table 1). The average hydrogen ion concentration was higher at site 3 than site 1 and site 2 may be due to the higher percentage of macro invertebrates' i.e. 37.05 % and higher temperature than other sites.

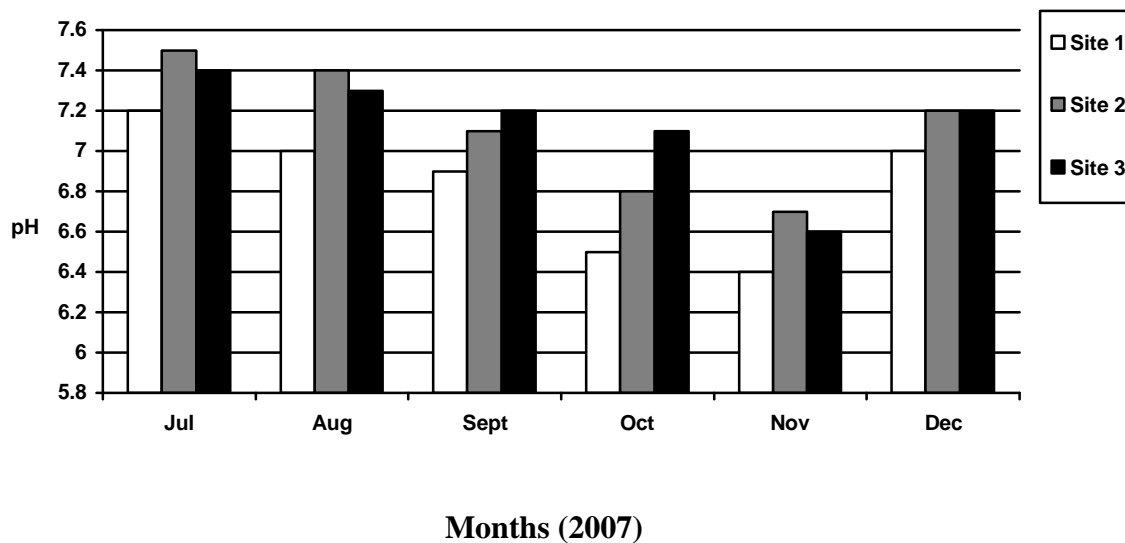


Figure 6: Variation in pH of water during the study period of six months

4.1.4. Total solids (TS) mg/l

The highest total solids (TS) at site 1 were obtained in site 1 i.e.1500 mg/l in the month of July and lowest of 500mg/l in the December (Table 24). At site 2, 1900 mg/l was maximum TS during the month of July and minimum TS was 1000 mg/l in December (Table 25). In July TS at site 3 was highest i.e. 1700 mg/l and 900 mg/l was lowest in December (Table 26). The average total solids (TS) of site 1 was found 1150 ± 143.73 mg/l and the average TS of site 2 was found in between 1383.33 ± 130.67 mg/l and similarly the average TS of site 3 was recorded between 1316.66 ± 125.43 mg/l (Table 1).The average total solids TS was highest at site 2 followed by site 3 and then site 1. This is may be due to that site 2 was composed of mostly sandy ground and less gravel than other sites.

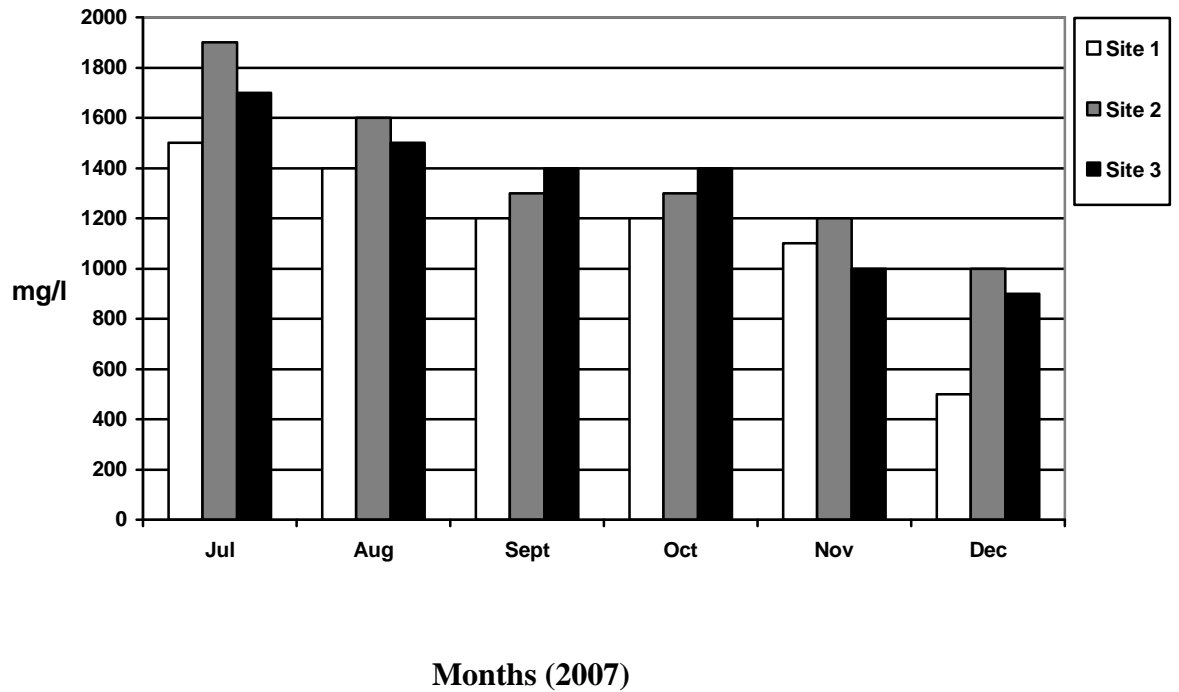


Figure 7: Variation in total solids of water during the study period of six months

4.1.5. Total dissolved solids (TDS) mg/l

The maximum total dissolved solids (TDS) at site 1 were 1200 mg/l in the months of July and August and minimum TDS was 400 mg/l during the month of December (Table 24). Site 2 had 1500 mg/l as a highest TDS in the month of July and lowest during the month of November and December i.e. 900 mg/l (Table 25). Site 3 had 1500 mg/l as maximum TDS in the month of July and 600 mg/l was minimum TDS in November and December (Table 26). The average TDS of site 1 was found in between 983.33 ± 122.72 mg/l, followed by TDS of site 2 i.e. 1133.33 ± 95.82 mg/l and the average TDS of site 3 was 1016.66 ± 145.28 mg/l (Table 1). The average TDS was highest at site 2 followed by site 1 and 3. This is may be due to that site 2 was composed of mostly sandy ground and less gravel than other sites.

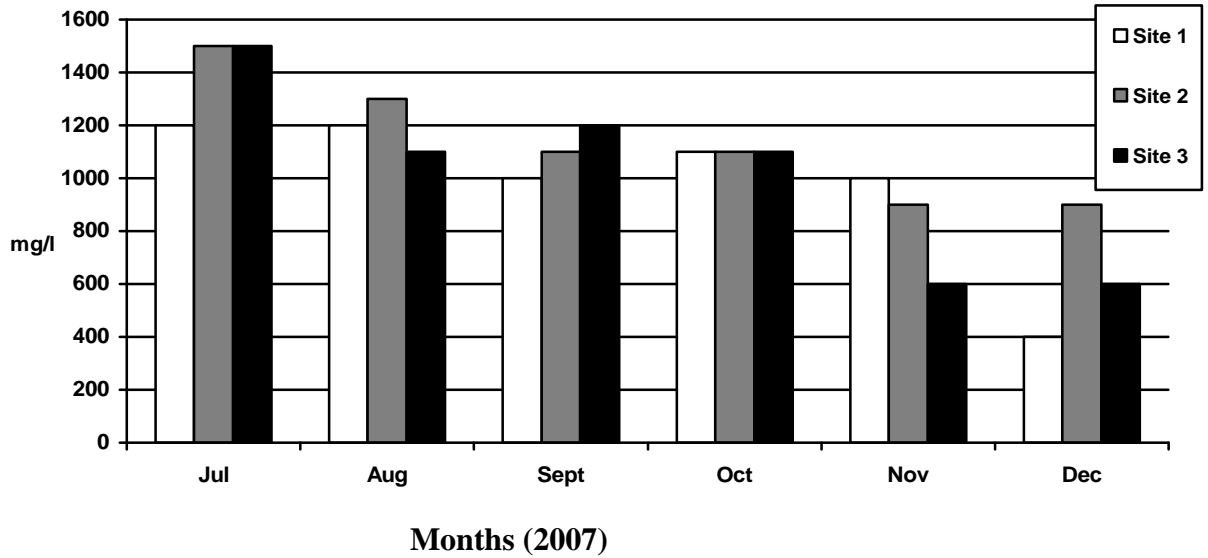


Figure 8: Variation in total dissolved solids of water during the study period of six months

4.1.6. Total suspended solids (TSS) mg/l

At site 1 the highest total suspended solids (TSS) were highest in the month of July i.e. 300mg/l and lowest TSS was 100 mg/l during the months of October, November and December (Table 24). At Site 2 highest TSS 400 mg/l was in July and lowest was 100 mg/l in the month of December (Table 25). The maximum TSS was 400 mg/l in the months of August and November and minimum during the month of July and September i.e.200 mg/l at site 3 (Table 25). The average TSS of site 1 was 300 ± 36.65 mg/l followed by the average TSS of site 2, 250 ± 42.98 mg/l and the average TSS of site 3 was 166.66 ± 74.82 mg/l (Table 1). So, average TSS was highest at site 3 which was followed by site 2 and site 1 i.e. it may be due to that the site 3 was mostly composed of much debris, substrums, and ground was sandy too with small pebbles.

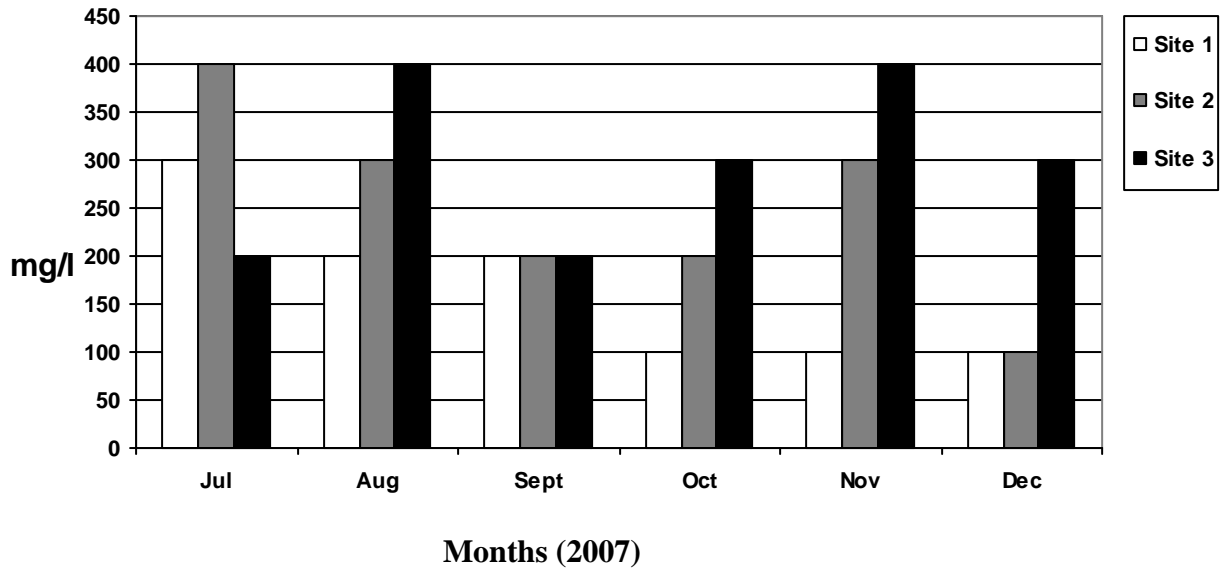


Figure 9: Variation in total suspended solids of water during the study period of six months

4.1.7. Dissolved oxygen (D.O.)mg/l

The maximum dissolved oxygen concentration (D.O.) was 8.10 mg/l in the month of November and minimum dissolved oxygen concentration was 6.89 mg/l in the months of August and September at site 1. At site 2 the highest D.O. concentration was 9.32 mg/l in November and lowest D.O. concentration was 6.89 mg/l during the months of August and September. 8.10 mg/l was highest concentration of D.O. in the month of November at site 3 and 6.89 mg/l was lowest during the months of August and September. The average D.O. concentration of site 1, site 2 and site 3 were 7.36 ± 0.19 mg/l, 7.76 ± 0.43 mg/l, 7.36 ± 0.19 mg/l respectively. Thus, average D.O. concentration was highest at site 2 than site 1 and site 3. This is might be due to that site 2 had higher water current and open area where oxygen can well mixed than other two sites.

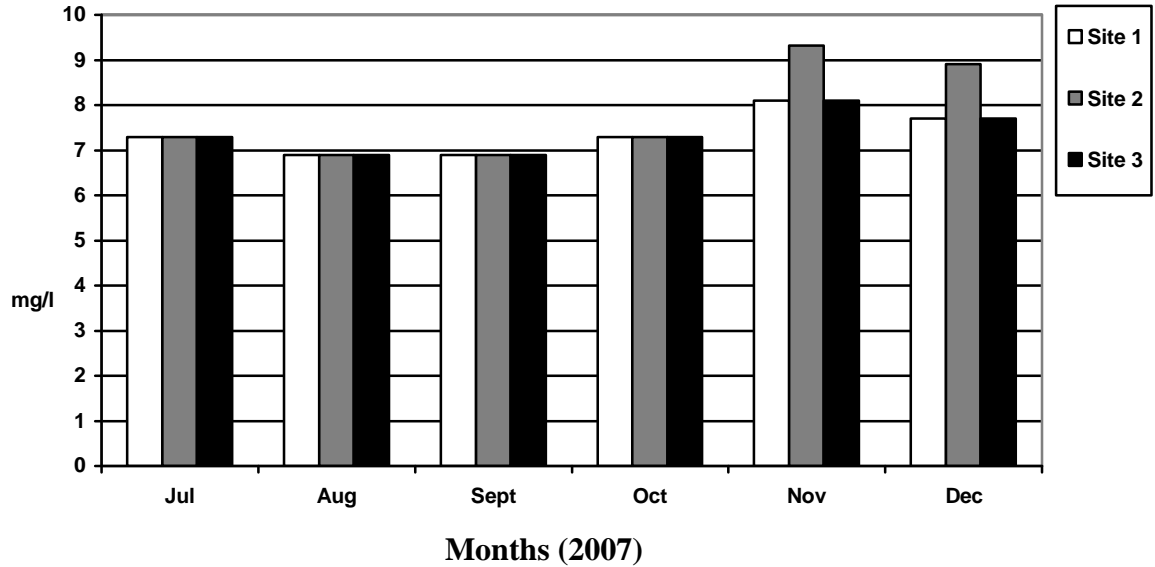


Figure 10: Variation in dissolved oxygen of water during the study period of six months

4.1.8. Free carbon dioxide (CO₂) mg/l

The highest free carbon dioxide was 13.2 mg/l in the month of July and lowest in the months of November and December i.e. 4.4 mg/l at site 1 (Table 24). At site 2, 8.8 mg/l was highest during the months of July, August and followed by the month September and lowest was 4.4 mg/l in the months of October, November and December (Table 25). The minimum concentration of free carbon dioxide was 4.4 mg/l in the months of July, August, November and December and maximum concentration of free carbon dioxide was 8.8 mg/l during the months of September and October at site 3 (Table 26). The average concentration of free carbon dioxide of water in site 1, 2 and 3 were 8.06 ± 1.36 mg/l, 6.6 ± 0.98 mg/l, 5.86 ± 0.93 mg/l respectively (Table 1). The average concentration of free carbon dioxide of water was highest at site 1 than site 2 and site 3 because lower the values of dissolved oxygen higher the value of carbon dioxide.

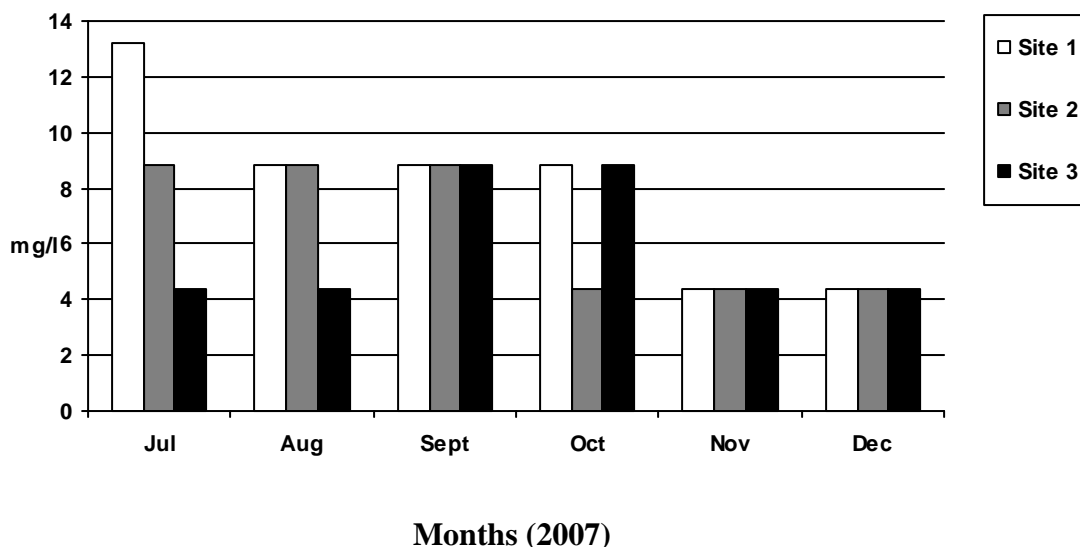


Figure 11: Variation in free carbon dioxide of water during the study period of six months

4.1.9. Total alkalinity mg/l

The total alkalinity was found highest in the month of July, September and October i.e. 50 mg/l and lowest 30 mg/l in the month of November at site 1 (Table 24). The highest content of total alkalinity was 70 mg/l in the month of July at site 2 and lowest content was 40 mg/l in the month of November (Table 25). At site 3 maximum content of total alkalinity was 70 mg/l in the month of October and minimum content was 30 mg/l during the month of September (Table 26). The average content of total alkalinity was 43.33 ± 3.34 mg/l, 55 ± 4.29 mg/l, 46.66 ± 5.57 mg/l respectively and overall the average content of total alkalinity was highest at site 2 than site 1 and site 3 during the study period this might be due to presence much aquatic phytoplankton at site 2 than other two sites.

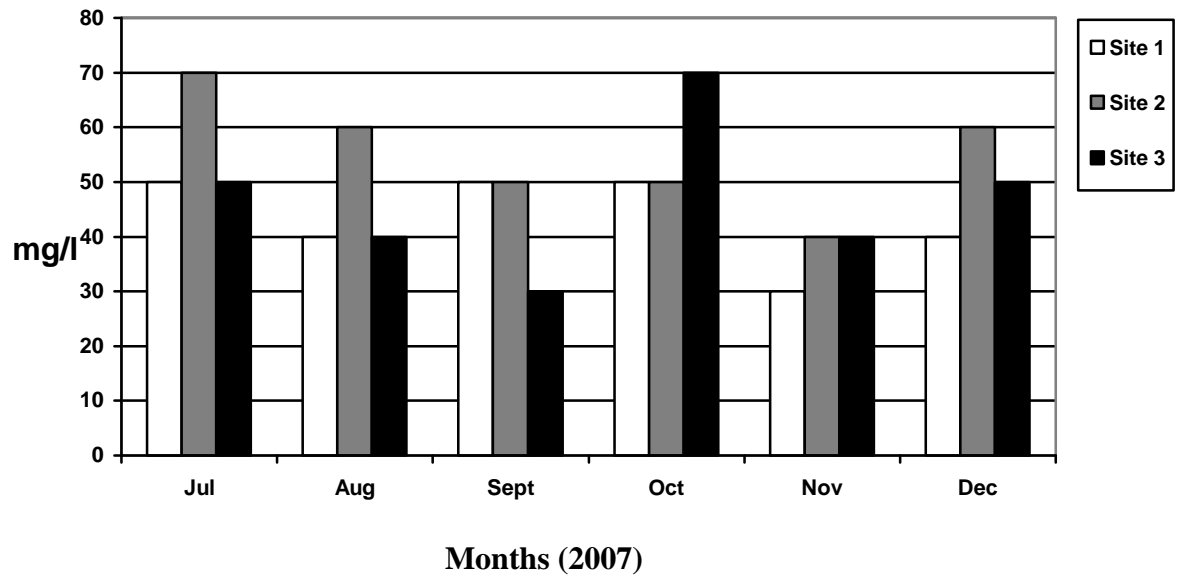


Figure 12: Variation in total alkalinity of water during the study period of six months

4.1.10. Total hardness mg/l

The total hardness of this site was found highest in the month of August i.e.20 mg/l at site 1 and lowest 10 mg/l in the months of September and November (Table 24). The maximum total hardness was 18 mg/l during the month of August and lowest content was 10 mg/l in the months of September and November at site 2 (Table 25). At site 3 highest total hardness of this site was 34 mg/l during the month of December and 10 mg/l was lowest during the months of July and September (Table 26). The average total hardness content of site 1, site 2 and 3 were recorded as 13.66 ± 1.58 mg/l, 13 ± 1.23 mg/l, 20.33 ± 3.94 mg/l respectively (Table 1). Thus, average total hardness was highest at site 3 than site 2 and site1. This reason might be due to pollutant and higher pH than other two sites.

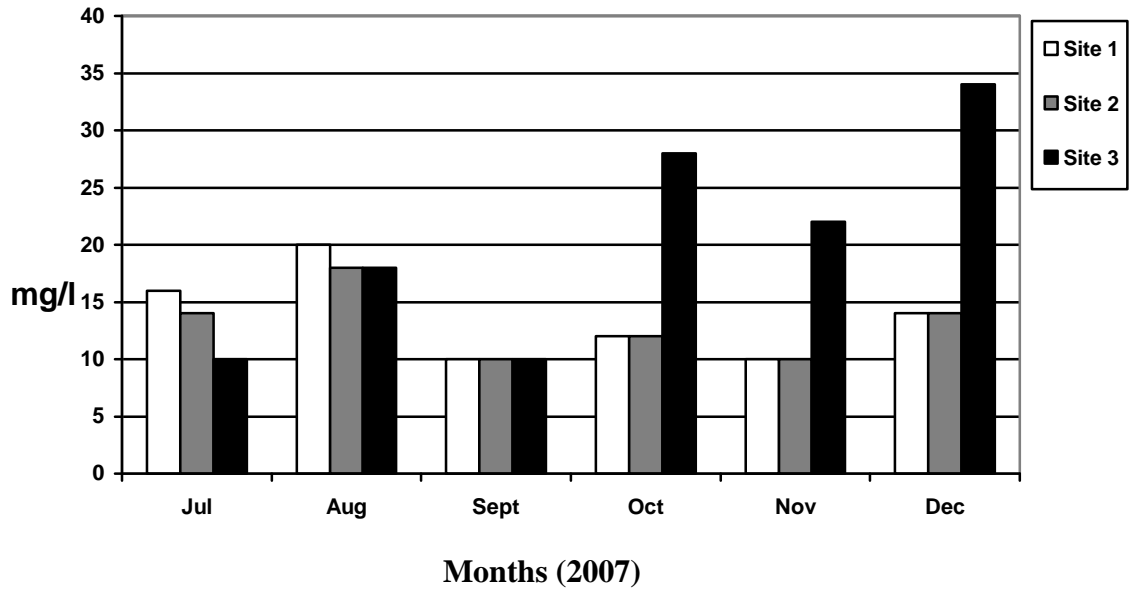


Figure 13: Variation in total hardness of water during the study period of six months

4.1.11. Calcium (Ca) hardness mg/l

Site 1 had the highest calcium (Ca) hardness 6.41 mg/l in the month of August and lowest Ca hardness was 2.40 mg/l in the month of September (Table 24). 4.80 mg/l was highest Ca hardness in the months of July and August and 2.40 mg/l was lowest Ca hardness content in the water during the months of September and November (Table 25). Site 3 had the maximum Ca hardness of 10.42 mg/l in October and minimum Ca hardness content of 3.20 mg/l in July and September (Table 26). The average Ca hardness were recorded in site 1, 2 and 3 were 4.26 ± 0.57 mg/l, 3.6 ± 0.45 mg/l, 6.27 ± 1.28 mg/l respectively (Table 1) and the average Ca hardness of water was highest at site 3 and followed by site 1 and site 2.

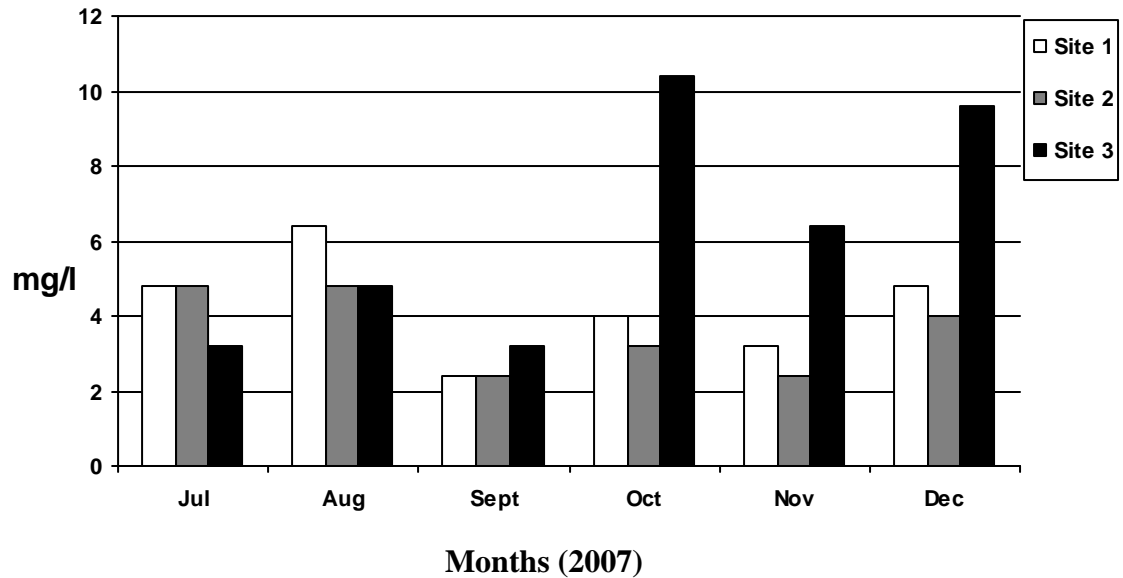


Figure 14: Variation in calcium hardness of water during the study period of six months

4.1.12. Magnesium (Mg) hardness mg/l

Site 1 had the highest magnesium (Mg) hardness was 0.97 mg/l during July, August and September and lowest in October, November and December i.e. 0.48 mg/l (Table 24). Site 2 had 1.46 mg/l as highest Mg hardness in August and 0.48mg/l was lowest in July (Table 25). Site 3 had a maximum Mg hardness of 2.43 mg/l in December and minimum of 0.48 mg/l in July, September and October (Table 26). The average Mg hardness of water in site 1, 2 and 3 were recorded as 0.72 ± 0.10 mg/l, 0.97 ± 0.003 mg/l, 1.13 ± 0.32 mg/l respectively. Thus, Site 3 has highest average Mg hardness of water than site 1 and site 2.

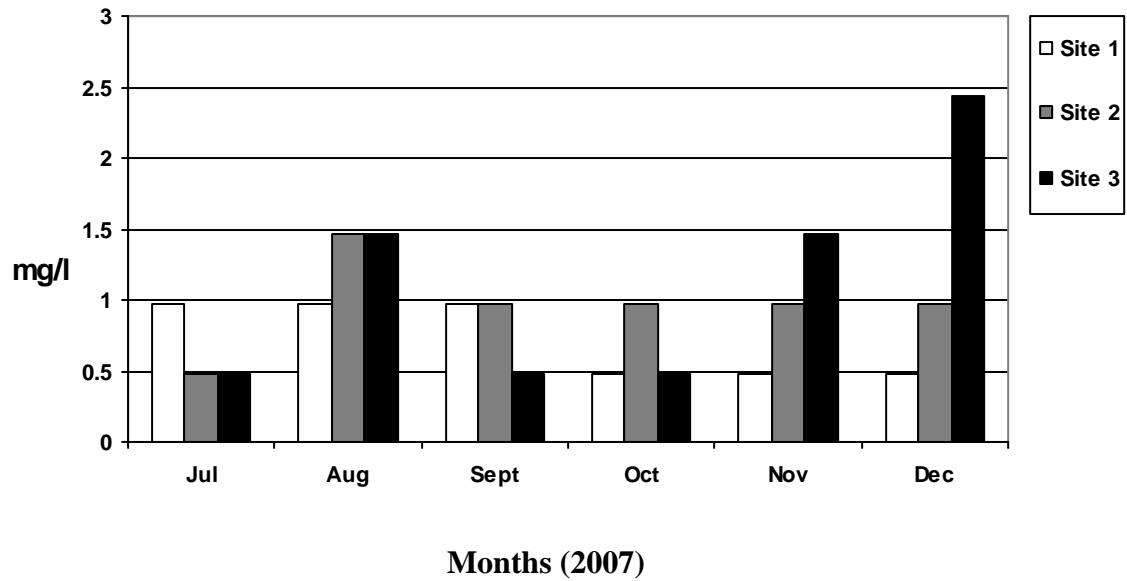


Figure 15: Variation in magnesium hardness of water during the study period of six months

4.2. Biological Parameters of water

4.2.1. Benthic Macro-invertebrates reported over the period of six months

Altogether 32 taxa belonging to 9 orders, 29 families and 28 genera of benthic-invertebrates were identified in Panimuhan stream during the present study periods which were as follows:

I) Order: Tricladida

A) Family: Planariidae

Genus: *Dugesia* sp

II) Order: Ephemeroptera

A) Family: Heptageniidae

Genus: *Epeorus* sp

Genus: *Heptagenia* sp

B) Family: Baetidae

Genus: *Baetis* sp

C) Family: Caenidae

Genus: *Caenis* sp

D) Family: Ephemerellidae

Genus: *Serratella* sp

Genus: *Ephemerella* sp

III) Order: Odonata

A) Family: Aeshnidae

Genus: *Cephalaeschna* sp

B) Family: Gomphidae

Genus: *Ophiogomphus* sp

C) Family: Cordulegastridae

Genus: *Anotogaster* sp

D) Family: Chlorolestidae

Genus: *Megalestes* sp

IV) Order: Placoptera

A) Family: Perlidae

Genus: *Neoperla* sp

B) Family: Nemouridae

Genus: *Protonemura* sp

Genus: *Amphinemura* sp

C) Family: Peltoperlidae

Genus: *Peltoperla* sp

V) Order: Neuroptera

A) Family: Corydalidae

Genus: *Corydalis* sp

VI) Order: Coleoptera

A) Family: Psephenidae

Psephenidae larvae

B) Family: Gyrinidae

Gyrinidae larvae

C) Family: Elmidae

Elmidae larvae and adult

VII) Order: Trichoptera

A) Family: Glossosomatidae

Genus: *Glossosoma* sp

B) Family: Hydropsychidae

Genus: *Hydropsyche* sp

C) Family: Stenopsychidae

Genus: *Stenopsyche* sp

D) Family: Hydroptilidae

E) Family: Leptoceridae

Genus: *Nectopsyche* sp

F) Family: Lepidostomatidae

Genus: *Lepidostoma* sp

G) Family: Rhyacophilidae

Genus: *Rhyacophila* sp

H) Family: Philopotamidae

Genus: *Chimarra* sp

VIII) Order: Diptera

A) Family: Simuliidae

Genus: *Simulium* sp

B) Family: Chironomidae

Genus: *Chironomus* sp

C) Family: Tipulidae

Genus: *Antocha* sp

D) Family: Athericidae

Genus: *Atherix* sp

IX) Order: Stylommatophora

A) Family: Physidae

Genus: *Physa* sp

In this present study the number of benthic macro invertebrates taxa recorded in all sites could be ordered as 3>1>2 with the number of 1878 (29.65%), 2108 (33.28%) and 2347 (37.05%). In July the total number of 945 (14.92%) of benthic macro invertebrates was collected. Among them site 3 had a maximum number of benthos (Table 2). During August total number of 1004 (15.85%) including all sites and also site 3 had highest number of fauna. Similarly in September site 3 had greater benthos then followed by site 1 and site 2 with altogether of 1093 with 17.25% of benthos during the investigation period. In the same way October and November had somewhat similar percentage of benthos i.e. 17.59% and 17.67% respectively. And in December site 3 stream had maximum number of species 385 followed by site 1 and site 2 i.e. 373 and 298 with altogether 1056 (16.67%).

Table 2: Total numbers of benthic macro-fauna collected from July -December 2007

Months	Sites			Total	Percentage (%)
	S₁	S₂	S₃		
July	289	297	359	945	14.92
August	342	308	354	1004	15.85
September	367	326	400	1093	17.25
October	370	321	423	1114	17.59
November	367	328	426	1121	17.70
December	373	298	385	1056	16.67
Total	2108	1878	2347	6333	
Percentage (%)	33.28	29.65	37.05		

4.2.2. Monthly fluctuation in mean values of benthic macro invertebrates

In present study period of six month from July to December, nine orders of taxa were identified in the Panimuhan stream. Among them Tricladida was found maximum in September i.e. 73 with a total value of 383. Order Ephemeroptera was dominant group during the study period and were reported highest i.e. 321 during December with a total value of 1812. Odonata was found maximum in July i.e. 66 with a total value of 279. The total value of 915 order Placoptera was third dominant group which was highest i.e. 166 in December. Neuroptera was maximum in October with a total value of 163. Altogether 135 Coleoptera was recorded and was numerous in September i.e. 30. Second dominate group was Tricoptera with a total value of 1621 and found maximum in August i.e.295. The maximum total value of Diptera was 844 and was highest in 191 in December. Stylommatophora were absent in July and August and was highest in November i.e. 50 with a total value of 181 (Table 3).

**Table 3: Monthly fluctuation in mean values of benthic-fauna distribution
(no. /m²)**

Taxa	Months						Total	Mean	%
	Jul	Aug	Sept	Oct	Nov	Dec			
Tricladida	60	65	73	70	58	57	383	63.8	6.0
Ephemeroptera	263	286	315	311	316	321	1812	302	28.6
Odonata	66	59	56	47	33	18	279	46.5	4.4
Placoptera	128	143	151	162	165	166	915	152.5	14.4
Neuroptera	28	27	28	32	31	17	163	27.1	2.5
Coleoptera	15	20	30	27	26	17	135	22.5	2.1
Tricoptera	276	295	281	277	266	226	1621	270.1	25.5
Diptera	109	109	119	140	176	191	844	140.6	13.3
Stylommatophora	-	-	40	48	50	43	181	30.1	2.8
Total							6333	1055.5	

4.2.3. The average percentages distribution of each representative species

According to the above table 3 the order Ephemeroptera were reported as a highest number species with 28.6 % during the study period of six months, which followed by another dominant group Tricoptera with a number species 25.5 % and Placoptera 14.4 % from all the sites. During investigation period group Coleopteran were recorded as lowest in number i.e. 2.1 % followed by Neuroptera 2.5 %, Stylommatophora 2.8 % Odonata 4.4 %, and Tricladida 6.0 %. (Figure 16).

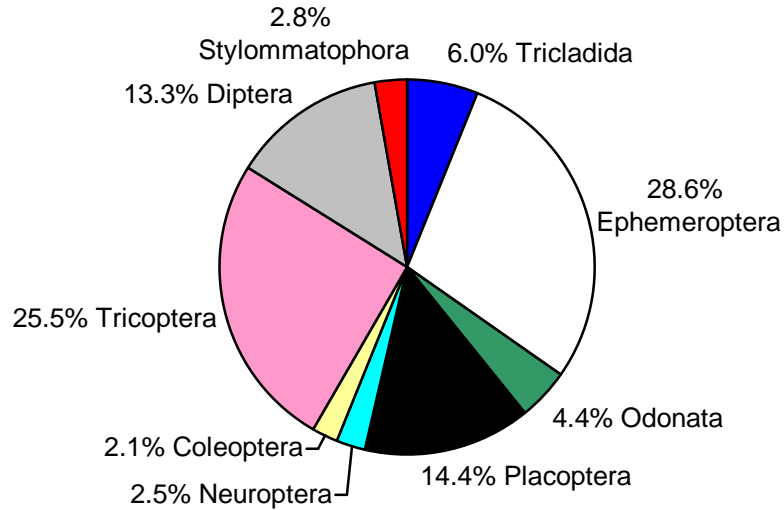


Figure 16: The percentage of benthic macro invertebrates in 2007

4.3. Species diversity of benthic-macro invertebrates

The table-4 shows the value of Shannon-Wiener Diversity Index (\bar{D}) and Evenness Index (e) for all the stations during the study period of six months. The value of (\bar{D}) of site 1 and site 3 i.e. 1.75 showed similar faunal diversity and values of (e) i.e. 0.52, 0.40 and 0.51 at sites 1, 2 and 3 respectively indicates alike evenness throughout the study period (Table 4).

Table 4: Shannon – Wiener Diversity Index for benthic-macro invertebrates

Sites	\bar{D}	e
S ₁	1.75	0.52
S ₂	1.49	0.40
S ₃	1.75	0.51

4.4. Correlation between Physicochemical Parameters and Benthic-macro fauna at sampling Sites

Correlation coefficients of different physicochemical parameters with number of species recorded from different sampling sites were calculated (table 5).

Benthic-macro fauna showed negative correlation with velocity, temperature, pH, free carbon dioxide, total hardness, and calcium hardness. It implies the increase in number of species at lowering the above mention variables.

Table 5: Correlation Coefficient between variables and Benthic-macro fauna

Physicochemical Parameters	Number of Benthic-macro fauna at sampling sites
Velocity m/sec	-0.01
Temperature °C	-0.64
Hydrogen Ion Concentration	-0.009
Total solids (TS) mg/l	+0.51
Total dissolved solids (TDS)mg/l	+0.08
Total suspended solids(TSS)mg/l	+0.41
Dissolved Oxygen (mg/l)	+0.11
Free CO ₂ (mg/l)	-0.03
Total Alkalinity(mg/l)	+0.07
Total Hardness(mg/l)	-0.10
Ca Hardness (mg/l)	-0.1
Mg Hardness(mg/l)	+0.54

Similarly, total solids, total dissolved solids, total suspended solids, dissolved oxygen, total alkalinity, and magnesium hardness showed positive correlation that implies the fact that high concentration of above mention variables in water favored the high diversity of aquatic macro fauna.

4.5. The monthly fluctuation in abundance of benthic macro invertebrates

In the study period of six months order Tricladida has one family (Planariidae) with one genus *Dugesia* sp (Plate: 1) was reported from Panimuhan stream. It was commonly found among all sites. During over period of six months a maximum density of this family was recorded from site 1 i.e. 40 individuals/m² in the month of October (Table 6).

Table 6: Monthly variations in the abundance of Tricladida (no. /m²) at all three sites

Months	Sites			Total
	S ₁	S ₂	S ₃	
July	30	12	18	60
August	34	11	20	65
September	35	15	23	75
October	40	9	21	70
November	31	10	17	58
December	28	12	17	57

Six genera of mayflies *Epeorus* sp. (Plate: 2), *Heptagenia* sp. (Plate: 3), *Baetis* sp. (plate: 6), *Caenis* sp. (Plate: 7), *Serratella* sp. (Plate: 9) and *Ephemerella* sp (Plate: 11) were recorded from 4 families (Heptageniidae, Baetidae, Caenidae and Ephemerellidae) belonging to order Ephemeroptera. During study period of six months a maximum density of 122 individual/m² of Ephemeroptera was reported at site 1 in the month of December (Table 7).

Table 7: Monthly variations in the abundance of Ephemeroptera (no. /m²) at all three sites

Months	Sites			Total
	S ₁	S ₂	S ₃	
July	84	79	100	263
August	106	87	93	286
September	118	96	101	315
October	118	86	107	311
November	114	83	119	316
December	122	87	112	321

A total of four genera of Odonata, *Cephalaescha* sp. (Plate: 12), *Ophiogomphus* sp (Plate: 14) *Anotogaster* sp. (Plate:16) and *Megalestes* sp. (Plate: 18) belonging to 4 families Gomphidae, Aeshnidae, Cordulegastridae and Chorolestidae respectively were recorded from all sites in all months of investigation period except in site 2

during the month of December. The highest of 26 individual/ m² were recorded from site 3 in August in six months (Table 8).

Table 8: Monthly variations in the abundance of Odonata (no. /m²) at all three sites

Months	Sites			Total
	S ₁	S ₂	S ₃	
July	20	21	25	66
August	16	17	26	59
September	17	19	20	56
October	15	14	18	47
November	15	7	11	33
December	14	–	4	18

Altogether four genera of stoneflies *Neoperla* sp. (Plate: 20), *Protonemura* sp. (Plate: 21), *Amphinemura* sp. (Plate: 22), *Peltoperla* sp. (Plate: 24), and belonging to 3 families Nemouridae, Peltoperlidae and Perlidae of an order Placoptera were recorded from all three sampling sites. The maximum density of 65 individual/m² was recorded in the month of December at site 1 with in six months (Table 9).

Table 9: Monthly variations in the abundance of Placoptera (no. /m²) at all three sites

Months	Sites			Total
	S ₁	S ₂	S ₃	
July	42	43	43	128
August	52	51	40	143
September	58	48	45	151
October	61	54	47	162
November	58	56	51	165
December	65	48	53	166

Order Neuroptera compared only one genus *Corydalus* sp (Plate: 25) belongs to family Corydalidae. The maximum number of Neuroptera was found 11 individual/ m² recorded from site 1 in September, November and December also from site 3 in

July but it was absent in December from site 3. The highest number of Neuroptera collected in October i.e.15 individual/ m² during study period (Table 10).

Table 10: Monthly variations in the abundance of Neuroptera (no. /m²) at all three sites

Months	Sites			Total
	S ₁	S ₂	S ₃	
July	8	9	11	28
August	10	7	10	27
September	11	8	9	28
October	15	5	12	32
November	11	7	13	31
December	11	6	-	17

The aquatic beetles of order Coleoptera were recorded regularly from all sites in all months except in the month of July from site 1. In Coleoptera three families; Psephenidae (Plate: 26), Gyrinidae (Plate: 28), Elmidae (Plate: 29) and were identified. From family Elmidae larvae were recorded from site 2 and site 3, and was absent in site 1 but the adult was identified from all sites. In case of Psephenidae, only larvae were recorded from site 3. In the six months period the highest value of Coleoptera was 12 individual/ m² from site 2 in August and September (Table 11).

Table 11: Monthly variations in the abundance of Coleoptera (no. /m²) at all three sites

Months	Sites			Total
	S ₁	S ₂	S ₃	
July	-	10	5	15
August	4	12	4	20
September	8	12	10	30
October	9	10	8	27
November	8	10	8	26
December	7	7	3	17

Among seven genera of order Tricoptera; *Glossoma* sp. (Plate: 31), *Hydropsyche* sp. (Plate: 32), *Stenopsyche* sp. (Plate: 33), *Nectopsyche* sp. (Plate: 35), *Lepidostoma* sp. (Plate: 36), *Rhyacophila* sp (Plate: 37) and *Chimarra* sp (Plate: 38) and belonging to families Glossomatidae, Hydropsychidae, Stenopsychidae, Hydroptilidae (Plate: 34), Leptoceridae, Lepidostomatidae, Rhyacophilidae and Philopotamidae were recorded while specimens belongs to the family Hydroptilidae was unidentified. This Tricoptera was second dominant order of stream. During overall study period maximum density was found at site 1 i.e. 108 individual/ m² in November (Table 12).

Table 12: Monthly variations in the abundance of Tricoptera (no. /m²) at all three sites

Months	Sites			Total
	S ₁	S ₂	S ₃	
July	88	91	97	276
August	105	91	99	295
September	105	86	90	281
October	101	80	96	277
November	108	65	93	266
December	103	38	85	226

Among four genera of aquatic dipteran larvae; *Simulium* sp (Plate: 40), *Chironomus* sp. (Plate: 42) *Antocha* sp. (Plate: 43), *Atherix* sp. (Plate: 44), and belonging to families Tipulidae, Athericidae, Simuliidae and Chironomidae respectively were collected. However, the *Chironomus* sp were absent in site 2. Among all dipteran larvae *Simulium* sp was found highest in site 2 and *Antocha* sp. and *Atherix* sp. were found at site 1 and site 3. The maximum number of 100 individual/ m² of dipteran were collected within the study periods from site 2 in December (Table 13).

Table 13: Monthly variations in the abundance of Diptera (no. /m²) at all three sites

Months	Sites			Total
	S ₁	S ₂	S ₃	
July	17	32	60	109
August	15	32	62	109
September	15	42	62	119
October	11	63	66	140
November	22	90	64	176
December	23	100	68	191

Only one genera *Physa* sp. (Plate: 45) of order Stylommatophora belonging to family Physidae was collected from site 3 in September, October, November and December. It was absent in other sampling sites. The maximum number of Stylommatophora 50 individual/ m² was recorded in November within six months of period (Table 14).

Table 14: Monthly variations in the abundance of Stylommatophora (no. /m²) at all three sites

Months	Sites			Total
	S ₁	S ₂	S ₃	
July	-	-	-	-
August	-	-	-	-
September	-	-	40	40
October	-	-	48	48
November	-	-	50	50
December	-	-	43	43

4.6. Analysis of Statistical Tools

The value of arithmetic mean (\bar{X}), variance (S^2) and chi-square (χ^2) of different taxa are presented as follow in the tables 15 to 23. The tabulated value of chi-square (χ^2) at 2 (n-1) degree of freedom under 0.05% (p=0.05) C.L. showed 5.991.

The calculated value of chi-square (χ^2) was compared with tabulated value. The tabulated value of chi-square (χ^2) exceed the calculated value so the result meant there was no significant different on the variance of sample from the mean of order Tricladida (Table 15).

Table 15: Value of Arithmetic mean (\bar{X}), Variance (S^2), Chi-square (χ^2) with significance level (P) for order Tricladida at different sites

Site 1

Months	\bar{X}	S^2	χ^2	P	n
July	10.0	1.0	0.2	<0.05	3
August	11.3	1.3	0.2	<0.05	3
September	11.6	2.3	0.4	<0.05	3
October	13.3	2.3	0.3	<0.05	3
November	10.3	2.3	0.4	<0.05	3
December	9.3	6.3	1.3	<0.05	3

Site 2

Months	\bar{X}	S^2	χ^2	P	n
July	4.0	1.0	0.5	<0.05	3
August	3.6	6.3	3.5	<0.05	3
September	3.0	1.0	0.4	<0.05	3
October	3.0	1.0	0.6	<0.05	3
November	3.3	0.3	0.2	<0.05	3
December	4.0	3.0	1.5	<0.05	3

Site 3

Months	\bar{X}	S^2	χ^2	P	n
July	6.0	4.0	1.3	<0.05	3
August	6.6	4.3	1.3	<0.05	3
September	7.6	4.3	1.1	<0.05	3
October	7.0	3.0	0.8	<0.05	3
November	5.6	0.3	0.1	<0.05	3
December	5.6	4.3	1.5	<0.05	3

The calculated value of chi-square (χ^2) was compared with tabulated value. The tabulated value of chi-square (χ^2) exceed the calculated value so the result meant there was no significant different on the variance of sample from the mean of order Ephemeroptera (Table 16).

Table 16: Value of Arithmetic mean (\bar{X}), Variance (S^2), Chi-square (χ^2) with significance level (P) for order Ephemeroptera at different sites

Site 1

Months	\bar{X}	S^2	χ^2	P	n
July	28.0	1.0	0.07	<0.05	3
August	35.3	8.3	0.4	<0.05	3
September	39.3	0.3	0.01	<0.05	3
October	39.3	2.3	0.1	<0.05	3
November	38.0	7.0	0.3	<0.05	3
December	40.6	0.3	0.01	<0.05	3

Site 2

Months	\bar{X}	S^2	χ^2	P	n
July	26.0	3.0	0.2	<0.05	3
August	29.0	1.0	0.06	<0.05	3
September	32.0	4.0	0.2	<0.05	3
October	28.6	1.3	0.09	<0.05	3
November	27.6	6.3	0.4	<0.05	3
December	29.0	3.0	0.2	<0.05	3

Site 3

Months	\bar{X}	S^2	χ^2	P	n
July	33.3	0.3	0.01	<0.05	3
August	31.0	1.0	0.06	<0.05	3
September	33.6	1.3	0.07	<0.05	3
October	35.6	0.3	0.01	<0.05	3
November	39.6	4.3	0.2	<0.05	3
December	37.3	2.3	0.1	<0.05	3

The calculated value of chi-square (χ^2) was compared with tabulated value. The tabulated value of chi-square (χ^2) exceed the calculated value so the result meant there was no significant different on the variance of sample from the mean of order of Placoptera (Table 17).

Table17: Value of Arithmetic mean (\bar{X}), Variance (S^2), Chi-square (χ^2) with significance level (P) for order Placoptera at different sites

Site 1

Months	\bar{X}	S^2	χ^2	P	n
July	14.0	1.0	0.1	<0.05	3
August	17.3	2.3	0.2	<0.05	3
September	19.3	2.3	0.2	<0.05	3
October	20.3	2.3	0.2	<0.05	3
November	19.3	0.3	0.03	<0.05	3
December	21.6	1.3	0.1	<0.05	3

Site 2

Months	\bar{X}	S^2	χ^2	P	n
July	14.3	0.3	0.04	<0.05	3
August	17.0	1.0	0.1	<0.05	3
September	16.0	1.0	0.1	<0.05	3
October	18.0	4.0	0.4	<0.05	3
November	18.6	0.3	0.03	<0.05	3
December	16.0	1.0	0.1	<0.05	3

Site 3

Months	\bar{X}	S^2	χ^2	P	n
July	14.3	0.3	0.04	<0.05	3
August	13.3	2.3	0.3	<0.05	3
September	15.0	9.0	1.2	<0.05	3
October	15.6	0.3	0.03	<0.05	3
November	17.0	4.0	0.4	<0.05	3
December	17.6	9.3	1.0	<0.05	3

The calculated value of chi-square (χ^2) was compared with tabulated value. The tabulated value of chi-square (χ^2) exceed the calculated value so the result meant there was no significant different on the variance of sample from the mean of order Tricoptera (Table 18).

Table 18: Value of Arithmetic mean (\bar{X}), Variance (S^2), Chi-square (χ^2) with significance level (P) for Tricoptera at different sites

Site 1

Months	\bar{X}	S^2	χ^2	P	n
July	29.3	2.3	0.1	<0.05	3
August	35.0	1.0	0.05	<0.05	3
September	35.0	9.0	0.5	<0.05	3
October	33.6	2.3	0.1	<0.05	3
November	36.0	4.0	0.2	<0.05	3
December	34.3	4.3	0.2	<0.05	3

Site 2

Months	\bar{X}	S^2	χ^2	P	n
July	30.3	2.3	0.1	<0.05	3
August	30.3	6.3	0.4	<0.05	3
September	28.6	1.3	0.09	<0.05	3
October	26.6	4.3	0.3	<0.05	3
November	21.6	2.3	0.2	<0.05	3
December	12.6	6.3	1.0	<0.05	3

Site 3

Months	\bar{X}	S^2	χ^2	P	n
July	32.3	6.3	0.3	<0.05	3
August	33.0	4.0	0.2	<0.05	3
September	30.0	1.0	0.06	<0.05	3
October	32.0	1.0	0.06	<0.05	3
November	31.0	1.0	0.06	<0.05	3
December	28.33	2.3	0.1	<0.05	3

The calculated value of chi-square (χ^2) was compared with tabulated value. The tabulated value of chi-square (χ^2) exceed the calculated value so the result meant there was no significant different on the variance of sample from the mean of order Odonata but in October at site 2 the calculated value exceed the tabulated value so there was significant different on the variance of sample from mean (Table 19).

Table19: Value of Arithmetic mean (\bar{X}), Variance (S^2), Chi-square (χ^2) with significance level (P) for order Odonata at different sites

Site 1

Months	\bar{X}	S^2	χ^2	P	n
July	6.6	4.3	1.3	<0.05	3
August	5.3	0.3	0.1	<0.05	3
September	5.6	4.3	1.5	<0.05	3
October	5.0	1.0	0.4	<0.05	3
November	5.0	1.0	0.4	<0.05	3
December	4.6	1.3	0.5	<0.05	3

Site 2

Months	\bar{X}	S^2	χ^2	P	n
July	7.0	1.0	0.2	<0.05	3
August	5.6	4.3	1.5	<0.05	3
September	6.3	2.3	0.7	<0.05	3
October	4.6	16.3	7.1	>0.05	3
November	2.3	4.3	3.7	<0.05	3
December	-	-	-	-	3

Site 3

Months	\bar{X}	S^2	χ^2	P	n
July	8.3	2.3	0.5	<0.05	3
August	8.6	1.3	0.3	<0.05	3
September	6.6	1.3	0.4	<0.05	3
October	6.0	3.0	1.0	<0.05	3
November	3.6	2.3	1.2	<0.05	3
December	1.3	0.3	0.5	<0.05	3

The calculated value of chi-square (χ^2) was compared with tabulated value. The tabulated value of chi-square (χ^2) exceed the calculated value so the result meant there was no significant different on the variance of sample from the mean of order Coleoptera (Table 20).

Table 20: Value of Arithmetic mean (\bar{X}), Variance (S^2), Chi-square (χ^2) with significance level (P) for Coleoptera at different sites

Site 1

Months	\bar{X}	S^2	χ^2	P	n
July	-	-	-	-	3
August	1.3	0.3	0.5	<0.05	3
September	2.6	0.3	0.2	<0.05	3
October	3.0	1.0	0.6	<0.05	3
November	2.6	1.3	1.0	<0.05	3
December	2.3	2.3	2.0	<0.05	3

Site 2

Months	\bar{X}	S^2	χ^2	P	n
July	3.3	0.3	0.2	<0.05	3
August	4.0	7.0	3.5	<0.05	3
September	4.0	1.0	0.5	<0.05	3
October	3.3	5.3	3.2	<0.05	3
November	3.3	0.3	0.2	<0.05	3
December	2.3	6.3	5.5	<0.05	3

Site 3

Months	\bar{X}	S^2	χ^2	P	n
July	1.6	2.3	2.9	<0.05	3
August	1.3	0.3	0.5	<0.05	3
September	3.3	2.3	1.4	<0.05	3
October	2.6	1.3	1.0	<0.05	3
November	2.6	5.3	4.1	<0.05	3
December	1.0	1.0	2.0	<0.05	3

The calculated value of chi-square (χ^2) was compared with tabulated value. The tabulated value of chi-square (χ^2) exceed the calculated value so the result meant there was no significant different on the variance of sample from the mean of order Diptera (Table 21).

Table 21: Value of Arithmetic mean (\bar{X}), Variance (S^2), Chi-square (χ^2) with significance level (P) for order Diptera at different sites

Site 1

Months	\bar{X}	S^2	χ^2	P	n
July	5.6	0.3	0.1	<0.05	3
August	5.0	7.0	2.8	<0.05	3
September	5.0	4.0	1.6	<0.05	3
October	3.6	10.3	5.7	<0.05	3
November	7.3	0.3	0.08	<0.05	3
December	7.6	4.3	1.0	<0.05	3

Site 2

Months	\bar{X}	S^2	χ^2	P	n
July	10.6	4.3	0.8	<0.05	3
August	10.6	1.3	0.2	<0.05	3
September	14.0	4.0	0.5	<0.05	3
October	21.0	1.0	0.09	<0.05	3
November	30.0	7.0	0.4	<0.05	3
December	33.3	12.3	0.7	<0.05	3

Site 3

Months	\bar{X}	S^2	χ^2	P	n
July	20.0	1.0	0.1	<0.05	3
August	20.6	2.3	0.2	<0.05	3
September	20.6	2.3	0.2	<0.05	3
October	22.0	1.0	0.09	<0.05	3
November	21.3	2.3	0.2	<0.05	3
December	22.6	0.3	0.02	<0.05	3

The calculated value of chi-square (χ^2) was compared with tabulated value. The tabulated value of chi-square (χ^2) exceed the calculated value so the result meant there was no significant different on the variance of sample from the mean of order Neuroptera (Table 22).

Table 22: Value of Arithmetic mean (\bar{X}), Variance (S^2), Chi-square (χ^2) with significance level (P) for order Neuroptera at different sites

Site 1

Months	\bar{X}	S^2	χ^2	P	n
July	2.6	0.3	0.2	<0.05	3
August	3.3	5.3	3.2	<0.05	3
September	3.6	4.3	2.4	<0.05	3
October	5.0	3.0	1.2	<0.05	3
November	3.6	4.3	2.4	<0.05	3
December	3.6	8.3	4.6	<0.05	3

Site 2

Months	\bar{X}	S^2	χ^2	P	n
July	3.0	1.0	0.6	<0.05	3
August	2.3	0.3	0.2	<0.05	3
September	2.6	0.3	0.2	<0.05	3
October	1.6	0.3	0.3	<0.05	3
November	2.3	0.3	0.2	<0.05	3
December	2.0	1.0	1.0	<0.05	3

Site 3

Months	\bar{X}	S^2	χ^2	P	n
July	3.6	6.3	3.5	<0.05	3
August	3.3	2.3	1.4	<0.05	3
September	6.0	1.0	0.3	<0.05	3
October	4.0	1.0	0.5	<0.05	3
November	4.3	2.3	1.0	<0.05	3
December	-	-	-	-	-

The calculated value of chi-square (χ^2) was compared with tabulated value. The tabulated value of chi-square (χ^2) exceed the calculated value so the result meant there was no significant different on the variance of sample from the mean of order Stylommatophora (Table 23).

Table 23: Value of Arithmetic mean (\bar{X}), Variance (S^2), Chi-square (χ^2) with significance level (P) for order Stylommatophora

Site 3

Months	\bar{X}	S^2	χ^2	P	n
July	-	-	-	-	-
August	-	-	-	-	-
September	13.3	2.3	0.3	<0.05	3
October	16.0	3.0	0.3	<0.05	3
November	16.6	6.3	0.7	<0.05	3
December	14.3	4.3	0.6	<0.05	3

5. DISCUSSION

Generally the quality of water is defined by physical, chemical and biological components. Similarly water quality also judge by present of benthic fauna (Sheng, 1990). Forbe (1913) who described the biological observation are found to be more dependable in certain ways than chemical determination since they show cumulative effects of the present and past condition.

Velocity of water varies greatly in different parts of the same stream and is determined by the steepness of the surface gradient (Odum, 1971) which controls the deposition of organic matter at the substrate of the stream. The water velocity of Panimuhan stream was highest i.e. 1.0 m/sec in site 2 in July and August which was at the altitude of 1610m and also less number of substratum to block the water current. Roy (2006) also found similar data i.e. 1.4 m/sec as maximum current of water in June around the headwater streams of Sundarijal of Shivapuri National Park. But Basaula (2000) observed highest velocity of Baghdwar water i.e. 0.32 m/sec in August and Chakrabarti (2001) also recorded highest velocity of water during the month of July i.e. 0.52m/sec of Bagmati River.

Temperature is basically important for its effects on other properties by speeding up chemical reaction. Temperature is inversely proportional to dissolved oxygen (Holdren and Armstrong, 1980). It is intimately related to latitude, altitude, season etc (Hynes, 1970). During the investigation period, temperature of water was ranging from 9.9 °C in December to 21 °C in August also Basaula (2000) recorded the similar water temperature ranging from 11 °C in March to 21 °C in August however Chakrabarti(2001) and Roy(2006) observed 12°C in February and 22.5°C in June, 10.3°C to 11.8 °C around Sundarijal respectively.

pH indicates the acidity or alkalinity of an aqueous solution and represents its hydrogen ion concentration (Mahida, 1981). Natural water usually has pH values in between 5.0 to 8.5. pH content of the surface water might be related to the photosynthetic activities of phytoplankton. Yadav (1989) observed the temperature, carbon dioxide and oxygen were responsible for the seasonal variation in the pH

values. The present study showed that pH range from 6.4 to 7.5 which can consider to be good for biological activities similarly Roy (2006) also found similar pH value nearly neutral i.e. 7.1. Where Basaula (2000) found average pH value of 8.3 ± 0.62 , which might be due to microbial activities and photosynthetic activity of phytoplankton. But Chakrabarti (2001) observed high pH value of Bagmati River range from 7.3 to 9.4 due to organic pollution of river water.

Suspended solids may have direct selective effects on the invertebrates. The filter-feeders and those such as *Hydropsche* sp and *Simulium* sp which strain food particles including micro-organism from the water are most seriously affected by having their feeding mechanism choked (Hawkes, 1979). Total dissolved solids (TDS) level less than 600 mg/l is considered as good one (WHO, 1993). During the present study TDS was lowest i.e. 400 mg/l in the month of December at site 1 and 600mg/l at site 3.

Oxygen is the most important element for water quality control. Solubility of oxygen in water varies greatly with temperature. Other factors that govern the dissolved oxygen content of natural water are: turbulence at the water surface, the extent of the surface area exposed to the atmosphere, atmospheric pressure and the percentage of the oxygen in the surrounding air. During summer month when temperature is high, solubility of oxygen is low (Mahida, 1981). Concentration of oxygen below 5 mg/l might be adversely affect the functioning and survival of biological communities (WHO, 1971). In present observation, the oxygen level of water was higher than the permitted level. Oxygen content of water was highest in winter season i.e. 9.32 mg/l and lowest during the summer season when temperature was high i.e. 6.89 mg/l. Similarly Roy (2006) and Basaula (2000) also reported oxygen level above the permitted level i.e. 8.58 mg/l to 8.94 mg/l of Sundarijal 6.1 mg/l to 8.6 mg/l of near Baghdwar respectively, but Chakrabarti (2001) found oxygen level below the permitted level in all selected sites of Bagmati River i.e. between 0 mg/l to 2.91 mg/l. A low concentration of dissolved oxygen level indicates organic pollution of river or stream (Verma and Agrawal, 1986).

Free carbon dioxide is very important materials for phytoplankton during the photosynthesis process. It is highly soluble in water. The amount of dissolved carbon dioxide is referred to as free carbon dioxide which much more carbon dioxide is

present in form of bicarbonates and carbonates and is called fixed or bound carbon dioxide (Verma and Agrawal, 1986). The highest concentration of free carbon dioxide was 13.2 mg/l in July and lowest value was 4.4 mg/l during winter and Chakrabarti (2001) also recorded similar very high concentration of free carbon dioxide 26.98 mg/l in February and lowest was 8.43 mg/l in July but Basaula (2000) and Roy (2006) found all highest value of free carbon dioxide in the month of August i.e. 4.7 mg/l and 6.4 mg/l respectively. The value of the free carbon dioxide increased with the decrease in the value of pH and dissolved oxygen (Sharma, 2000).

The alkalinity is caused due to the presence of dissolved carbonates, bicarbonates or hydroxide of calcium, magnesium, sodium, potassium and ammonium in the solution. Julay *et al.* (1934) suggested the presence of carbonate alkalinity is due to aquatic plants mainly phytoplankton which removes half bond carbon dioxide from the bicarbonates leaving a certain amount of carbonates in the water which gives it an alkaline reaction. Usually the total alkalinity shows the inverse relation with carbon dioxide content in water (Kushlan and Hunt, 1979). But the present studied did not show the similar pattern probably alkalinity keep the pH of water in neutral by neutralizing the acidity. In the study highest and lowest value of total alkalinity were observed between 70 mg/l to 30 mg/l. Also Chakrabarti in 2001 found total alkalinity range between 606 mg/l to 106.6mg/l. But Basaula (2000) and Roy (2006) observed total alkalinity between 31 mg/l to 47 mg/l between 13.2mg/l to 18.9mg/l respectively.

Another parameter like hardness of water is due to presence of dissolved calcium and magnesium salts. According to WHO guidelines, when water contain salt of 500 mg/l is said be suitable. The water having hardness 15 mg/l or above may be good for growth of aquatic plants and animals (Swingle, 1967). During investigation period, the hardness of water was found between 34 mg/l to 10 mg/l and similarly Basaula (2000) also observed the hardness of between 19 mg/l to 30 mg/l near Baghdwar, but Chakrabarti (2001) reported hardness of water range from 63 mg/l to 122 mg/l and similarly Roy (2006) observed hardness range from 12 mg/l to 14 mg/l. The observed correlation of hardness and number of aquatic insects showed the negative relation (-0.10) which said to be decrease in hardness concentration with increase in diversity but this present study didn't show the same relation. Many groups of insects like

Simuliidae (Grenier, 1949) and Ephemeroptera (Illies, 1952) were found quite indifferent to hardness of water. In the investigation period calcium hardness was found between 2.40 mg/l to 10.42 mg/l and similarly magnesium hardness range from 0.48 mg/l to 2.43 mg/l but Chakrabarti (2001) recorded magnesium hardness range from 22.8 mg/l to 68.4 mg/l of Bagmati River.

In the present study, benthic-macro invertebrates were explored in Panimuhan stream. Altogether 32 taxa belonging to 9 orders, 29 families and 28 genera and were identified. Among them 1 (6.0%) species of Tricladida belonging to 1 family and 1 genus, 6 (28.6%) species of Ephemeroptera were identified belonging to 4 families and 6 genera, similarly Odonata comprised of 4 (4.4%) species belonging to 4 families with 4 genera, 4 (14.4%) species of Placoptera belonging to 3 families and 4 genera, 1 (2.5%) species of Neuroptera belonging to 1 family and 1 genus on other hand Coleoptera had 3 (2.1%) species with families and 3 genera, Tricoptera had 8 (25.5%) species belonging to 8 families and 8 genera, 4 (13.3%) species of Diptera belonging to 3 families and 4 genera, and 1 (2.8%) species of Stylommatophora had 1 family belonging to 1 genus were identified during the investigation period of six months.

During the present study, site 1 and site 3 had similar high species diversity of ($\bar{D} = 1.75$) while site 2 had slight lower species diversity ($\bar{D} = 1.49$) because of absence of suitable substrate and very speed current of water i.e. 1.0 m/sec in comparison with site 1 and 3 where above correlation also shows negative relation with current of water. Winterbourn (1978) and Linklater (1995) explained that leaf litter act not only just as substrate but as food sources too. And also site 2 had minimum food availability than site 1 and 3. But Basaula (2000) observed higher diversity of ($\bar{D} = 1.16$) and less diversity of ($\bar{D} = 1.14$). Also the value of evenness (e) were 0.52, 0.40 and 0.51 at sites 1, 2 and 3 respectively which depicts more or less similar evenness in all sites. Beside that benthic-macro fauna showed negative correlation with velocity, temperature, pH, free carbon dioxide, total hardness and calcium hardness. It implies the increase in number of species at lowering variables. Similarly, total solids, total dissolved solids, total suspended solids, dissolved oxygen, total alkalinity and magnesium hardness showed positive correlation that implies the fact

that high concentration variables in water favored the high diversity of aquatic macro fauna. Roy (2006) also reported negative correlation with temperature, pH and free carbon dioxide while positive correlation with dissolved oxygen.

Malla *et al.* (1978) reported 61 species of aquatic insects of different orders, families and genus from various water bodies of Kathmandu Valley. In descending order dominant numbers of species order are arranged as Ephemeroptera (28.6%), Tricoptera (25.5%), Placoptera (14.4%), Diptera (13.3%), Tricladida (6%), Odonata (4.4%), Stylommatophora (2.8%), Neuroptera (2.5) and Coleoptera (2.1%). Khadka (1983) recorded the dominant benthos in descending order of Tricoptera, Placoptera, Ephemeroptera and other fauna and similarly Shakya (1992) arranged the dominant order as Oligochaeta, Diptera, Ephemeroptera, Tricoptera, Annelida, Coleoptera, Sharma (1988) found Diptera as dominant group and followed by Oligochaeta and Ephemeroptera in Bagmati River and Roy (2006) confirm the Tricoptera as dominant order and followed by Ephemeroptera, Diptera, Placoptera, Odonata and so on. The higher abundance of Ephemeroptera and Tricoptera demonstrated the unpolluted water condition (Sharma, 2000; Yadav *et al.*, 1984).

The Tricladida was recorded highest at site 1 and site 3 because there were present of large number substratums like cobbles and gravel where they were found attached on them. Hynes (1970) stated that the larger the stones, the more complex the substratum, and more diverse was the macro-invertebrates. And similarly lowest number of species recorded from site 2 due to absence of suitable substratum for them where site 2 was composed of mostly sand and small pebbles. Ephemeroptera was dominant group of species during study period. The maximum diversity of this species was recorded from site 1 and more or less similar numbers of species were also recorded from site 3 may due to similar pattern of microhabitat. Among them *Baetis* sp and *Caenis* sp were recorded from almost all sites probably due to sufficient amount of dissolved oxygen. Ephemeropteran were observed highest in November and December. Similarly Sharma (1988) also recorded Ephemeropteran as highest order among other in December.

4 genera of Odonata were recorded but at site 2 in December no Odonata were recorded. In July there was maximum number of Odonata among them

Cephalaeschna sp was highest in all sites. Placoptera were also highest at site 1 and similar pattern were seen in other two sites too because of high dissolved oxygen level during the winter month as Mahida (1981) explained that during summer month when temperature is high, solubility of oxygen is low among them *Neoperla* sp was common at all sites. Order Neuroptera comprised one family Corydalidae and one genus *Corydalis* sp. It was found in all sites except at site 3 during December and *Corydalis* sp have a maximum number in October. In order Coleoptera three families were recorded. Gyrinidae larvae were found at site 1 and site 3 and a Psephenidae larva was found only at site 3. Elmidae adults were found in all sites but its larvae were recorded from site 1 and 3. Brown (1987) state that Elmids are found in shallow and fast moving water where they cling to substrate and feed by scraping hard surface for detritus and algae. Similarly Braccia and Voshell Jr. (2006) state that Elmids beetle like *Oulimnius* are found in space between clean substrate because the depth and different size of the substrate provide the continuum of current velocity and refuge from predators and repository for detritus food that would otherwise washed downstream.

The most diversified group and second dominant group found during present study was Tricoptera altogether 8 species were collected. In July, August and September stream had high velocity maximum the number of Tricopteran and lowest in December when the water current was minimum. *Glossosoma* sp had only the case made of small pebbles attached together on upper side of the stone and also Scott (1958); Kovalak (1976) and Frutiger (2002) explained that clinger species like *Glossosoma* sp adapted to exist in clean substrate with exposed surface and rarely reported from underside of the substrate. *Rhyacophila* sp and *Chimarra* sp were presence only at site 1 and site 3 among all Tricopteran, *Nectosyche* sp, *Lepidostoma* sp and *Hydropsyche* sp were maximum at all sites. Illies (1957), Miller and Paetz (1959) Petr (1970) and Philipson (1954) reported that greater velocity of water favor the maximum number of Tricoptera.

The Dipteran was found almost all in sites in present investigation. The Simuliidae was dominating genera than other taxa at site 2 where the current of water was maximum than site 1 and site 3 and pupae of *Simulium* sp were also recorded in similar pattern. Ogbeibu and Oribhabor (2002) studied in Ikpoba River of Southern

Nigeria observed that high water current lower the species. But in present study it doesn't show the similar pattern probably due to direct exposed to sunlight than other sites and low water temperature. Similar maximum number of Chironomidae was recorded from site 3 because as mention above site 3 was slightly polluted and it is pollution tolerant species (WHO, 1976). Stylommatophora had one family Physidae and one genus *Physa* sp was observed attached on the stone only at site 3 from September to December probably due to low water current. Statzner (1981) also explained that water velocity restrict the distribution of certain taxa because during their movement they have to pass through fast waters.

In study period of six months in Panimuhan stream showed abundant of order Ephemeroptera, vast diversity of Tricoptera and Placoptera indicate that stream water of Panimuhan is unpolluted because mayflies and caddis flies do not occur in polluted water also WHO (1976) mentioned that they are pollution intolerant aquatic insects where as *Chironomus* sp are pollution tolerant species which occur mostly at site 3 which indicates that site 3 is slightly polluted because water flow through human settlement. The minimum population density of Ephemeroptera and Placoptera in summer was related to their flight period which occurs in June to August. Overall the physico-chemical parameters and abundant of benthic macro invertebrates shows ecological condition of Panimuhan stream is unpolluted.

6. CONCLUSIONS

The present study emphasis on physicochemical parameters and benthic macro-invertebrates of Panimuhan stream of Shivapuri National Park which lies on the northern hillside of Kathmandu Valley. The study was conducted from July to December for the period of six months. The study sites were divided into three regions on the basis of altitude.

The physicochemical components of stream water i.e. velocity, temperature, hydrogen ion concentration, total solids, total dissolved solids, total suspended solids, dissolved oxygen, free carbon dioxide, total alkalinity, total hardness (Calcium and Magnesium) were analyzed in Natural Product Research Laboratory of NAST. Based on the result obtained, it can be concluded that the water current was highest at site 2 i.e. 1.0 m/sec and 21 °C was highest temperature at site 3 in August. Similarly pH was maximum i.e. 7.5 at site 2. 1900 mg/l was maximum TS at site 2. Similarly, the maximum TDS was 1500 mg/l at site 2 and 3 in July. The highest concentration of dissolved oxygen of 9.32 mg/l confirms at site 2 in November. On other hand highest free carbon dioxide was highest at site 1 i.e. 13.2 mg/l in July. The maximum total alkalinity of present study period was 70 mg/l at site 2 and 3 whereas the highest concentration of total hardness was 34 mg/l at site 3 in December. And 10.42 mg/l was maximum calcium hardness of site 3 and maximum concentration of magnesium hardness was 2.46 mg/l at site 3 in present study sites.

During the study period altogether 32 taxa belonging to 9 orders, 29 families and 28 genera of benthic-macro fauna were identified. Among them order Ephemeroptera was the most dominant group with 28.6 % of the total individual followed by Tricoptera and Placoptera with 25.5 % and 14.4% of the total individual respectively. Similarly site 2 had a minimum diversity of the species with only 29.6 % ($\bar{D} = 1.49$) of the total followed by site 1 with 33.2 % ($\bar{D} = 1.75$) and site 3 had a maximum diversity of 37.05 % ($\bar{D} = 1.75$). Benthic macro invertebrates showed positive correlation with total solids, total dissolved solids, total suspended solids, dissolved oxygen, total alkalinity and magnesium hardness and invertebrates showed negative correlation with velocity, temperature, pH, free carbon dioxide, total hardness and calcium hardness which favor that lower the values lesser the species diversity of the stream. Therefore, physicochemical components and abundance and diversity of benthic macro fauna show water of Panimuhan stream was unpolluted.

7. RECOMMENDATIONS

Among three study sites, sites 1 and 2 were inside the national park so there were no necessary to take precaution but site 3 was outside the national park which flow through the human settlement so according to present study the following measures are necessary to check the rate of the stream water pollution:

1. During the study period at study site 3 there were lots of solid wastes like tin, can, broken glasses, plastic bottles, papers, jute and tires were thrown which should be discouraged and people should aware of negative impact in natural resources.
2. During investigation period large number of chironomid larvae was identified which indicate the water is polluted therefore water should be treated before utilizing for household used.
3. People should be discouraged to use water for washing, swimming and other activities.
4. The locals should be aware about the detrimental effects of pollution and the implications of the pollution control programs.
5. There was limited time for such investigation, so for better result further research should be undertaken.

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9. APPENDICES 1

The monthly fluctuation on physicochemical components of all the three study sites are mention on tabular form.

Table 24: Monthly fluctuation on physicochemical parameters of site 1 during the study period of six months

S.N.	Parameters	Jul.	Aug.	Sept.	Oct.	Nov.	Dec.	Average	S.E.
1.	Velocity m/sec	0.66	0.57	0.54	0.52	0.45	0.35	0.515	0.036
2.	Temperature °C	15.2	18	17.5	15.5	12	9.9	14.68	1.29
3.	pH	7.2	7.0	6.9	6.5	6.4	7.0	6.83	0.011
4.	TS (mg/l)	1500	1400	1200	1200	1100	500	1150	143.73
5.	TDS(mg/l)	1200	1200	1000	1100	1000	400	983.33	122.72
6.	TSS (mg/l)	300	200	200	100	100	100	166.66	74.82
7.	Dissolved Oxygen (mg/l)	7.29	6.89	6.89	7.29	8.10	7.70	7.36	0.193
8.	Free CO ₂ (mg/l)	13.2	8.8	8.8	8.8	4.4	4.4	8.06	1.36
9.	Total Alkalinity (mg/l)	50	40	50	50	30	40	43.33	3.34
10.	Total Hardness (mg/l)	16	20	10	12	10	14	13.66	1.58
11.	Ca Hardness (mg/l)	4.80	6.41	2.40	4.0	3.20	4.80	4.26	0.57
12.	Mg Hardness (mg/l)	0.97	0.97	0.97	0.48	0.48	0.48	0.72	0.10

Table 25: Monthly fluctuations on physicochemical parameters of site 2 during the study period of six months

S.N.	Parameters	Jul.	Aug.	Sept.	Oct.	Nov.	Dec.	Average	S.E.
1.	Velocity m/sec	1.0	1.0	0.78	0.71	0.65	0.62	0.79	0.07
2.	Temperature °C	17.9	20.5	18.3	16	13	10.9	16.1	1.46
3.	Hydrogen Ion Concentration	7.5	7.4	7.1	6.8	6.7	7.2	7.11	0.13
4.	TS (mg/l)	1900	1600	1300	1300	1200	1000	1383.33	130.67
5.	TDS(mg/l)	1500	1300	1100	1100	900	900	1133.33	95.82
6.	TSS (mg/l)	400	300	200	200	300	100	250	42.98
7.	Dissolved Oxygen (mg/l)	7.29	6.89	6.89	7.29	9.32	8.91	7.76	0.43
8.	Free CO ₂ (mg/l)	8.8	8.8	8.8	4.4	4.4	4.4	6.6	0.98
9.	Total Alkalinity (mg/l)	70	60	50	50	40	60	55	4.29
10.	Total Hardness (mg/l)	14	18	10	12	10	14	13	1.23
11.	Ca Hardness (mg/l)	4.80	4.80	2.40	3.20	2.40	4.0	3.6	0.45
12.	Mg Hardness (mg/l)	0.48	1.46	0.97	0.97	0.97	0.97	0.97	0.003

Table 26: Monthly fluctuations on physicochemical parameters of site 3 during the study period of six months

S.N.	Parameters	Jul.	Aug.	Sept.	Oct.	Nov.	Dec.	Average	S.E.
1.	Velocity m/sec	0.66	0.50	0.47	0.42	0.39	0.22	0.44	0.05
2.	Temperature °C	19.1	21	20	18	15.5	14.5	18.01	1.04
3.	Hydrogen Ion Concentration	7.4	7.3	7.2	7.1	6.6	7.2	7.13	0.11
4.	TS (mg/l)	1700	1500	1400	1400	1000	900	1316.66	125.43
5.	TDS(mg/l)	1500	1100	1200	1100	600	600	1016.66	145.28
6.	TSS (mg/l)	200	400	200	300	400	300	300	36.65
7.	Dissolved Oxygen (mg/l)	7.29	6.89	6.89	7.29	8.10	7.70	7.36	0.19
8.	Free CO ₂ (mg/l)	4.4	4.4	8.8	8.8	4.4	4.4	5.86	0.93
9.	Total Alkalinity (mg/l)	50	40	30	70	40	50	46.66	5.57
10.	Total Hardness (mg/l)	10	18	10	28	22	34	20.33	3.94
11.	Ca Hardness (mg/l)	3.20	4.80	3.20	10.42	6.41	9.61	6.27	1.28
12.	Mg Hardness (mg/l)	0.48	1.46	0.48	0.48	1.46	2.43	1.13	0.32

APPENDICES- 2

Some of the photographs of collected benthic macro fauna and study sites of Panimuhan Stream.

Order 1: Tricladida



Plate 1: *Dugesia* sp

Order 2: Ephemeroptera



Plate 2: *Epeorus* sp



Plate 3: *Heptagenia* sp



Plate 4: Anal filament of *Heptagenia* sp



Plate 5: Abdominal gills of *Heptagenia* sp



Plate 6: *Baetis* sp



Plate 7: *Caenis* sp



Plate 8: Caudal filament of *Caenis* sp



Plate 9: *Serratella* sp



Plate 10: Anal filament of *Serratella* sp

Plate 11: Ephemerella sp

Order 3: Odonata

Plate 12: Cephalaeschna
sp

Plate 13: Prementum of
Cephalaeschna sp

Plate 14: Ophiogomphus sp

Plate 15: Prementum of
Ophiogomphus sp

Plate 16: Anotogaster sp

Plate 17: Palp dentation
of Anotogaster sp

Plate 18: Megalestes sp

Plate 19: Anal gills of
Megalestes sp

Order 4: Placoptera

Plate 20: Neoperla sp

Plate 21: Protonemura sp

Plate 22: Anal filament
showing swollen segment

Plate 23: Amphinemoura sp

Plate 24: Peltoperla sp

Order 5: Neuroptera

Plate 25: Corydalus sp

Order 6: Coleoptera

Plate 26: Psephenidae Larva
(Dorsal View)

Plate 27: Psephenidae Larva
(Ventral View)

Plate 28: Gyrinidae Larva

Plate 29: Elmidae(Adult)

Plate 30: Elmidae (larva)

Order 7: Tricoptera

Plate 31: Case of
Glossosoma sp

Plate 32: Hydropsyche sp

Plate 33: Stenopsyche sp

Plate 34: Hydroptilidae sp

Plate 35: Nectopsyche sp

Plate 36: Lepidostoma sp

Plate 37: Case of
Lepidostoma sp

Plate 38: Rhyacophila sp

Plate 39: Chimarra sp

Order 8: Diptera

Plate 40: Simulium sp

Plate 41: Pupae of Simulium
sp

Plate 42: Chironomus sp

Plate 43: Antocha sp

Plate 44: Atherix sp

Order 9: Stylommatophora

Plate 45: Physa sp

Plate 46: Site 1

Plate 48: Site 3

Plate 50: Solid wastes at site 3

Plate 52: Solid wastes near site 3

Plate 54: Trapping Dissolved oxygen

Plate 47: Site 2

Plate 49: Water Reservoir of
Panimuhan

Plate 51: Children swimming
At site 3

Plate 53: Sorting benthic macro
Fauna

Plate 55: Experiment in Lab