

Epidemiological Study of Malaria  
in Mahendranagar VDC of Sunsari District

A dissertation submitted in Partial fulfillment of the  
requirement for the Master's Degree in Zoology  
with special paper Parasitology

Submitted By  
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Submitted to  
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### RECOMMENDATION

This is to certify that **Mr. Ram Prasad Shahu** has completed his dissertation work entitled "**Epidemiological Study of Malaria in Mahendranagar VDC of Sunsari District**" as a partial fulfillment of the M. Sc. Degree in Zoology with special paper Parasitology under my supervision. This is the candidate's original work which brings out important findings in parasitological research. It is my pleasure to recommend this work for the partial fulfillment of Master of Science in Zoology at Tribhuvan University.

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## Abstract

The present study was conducted to assess the prevalence of malarial parasites among the people inhabiting the Mahendranagar VDC that lies in tropical region in Eastern Nepal. An active detection was made by collecting 250 blood samples from suspected individuals visiting at Health Post and Janasewa Clinic during the study period, by microscopic examination of thick and thin blood smear preparation. Besides this, structured questionnaire was used to assess the socio-economic status and the environmental aspects in relation to the malaria. Out of 250 blood samples collected, 10 samples were positive for the malaria infection. The slide positivity rate was found to be 4% and the causer organism was found to be *Plasmodium vivax* only. Age wise data revealed that of the total infected population the highest age specific slide positivity rate was found in 31-40 years age group (7.69%). Similarly in relation to sex, slide positivity rate was 5.33% in males and 2% in females. The prevalence rate of malaria was found to be higher in poor class people (5.15%) and those living in hut houses (7.54%). The malaria infection was common among Hindu people (4.65%) when compared to Muslim(0%) and Buddhist(0%). Likewise , prevalence of malaria was found to be the highest in illiterate (5.31%). Occupation wise prevalence was found to be highest in jobholders (5%) followed by farmer (4.8%) and labours (4.76%). High rate of infection was recorded during four summer months (April, May, July and September) with the largest number of patients during July (14.4%). In relation to environmental aspects and preventive measures malaria infection was common in hand pump users (4.13%) and in those applying no preventive measures (15%). With respect to migration, 80% (8 cases) of the total infected people were permanently inhabiting the area whereas 20% (2 cases) were migrated from India. Based on the present study; biological, physical and socio-economic measures are recommended to eradicate the malaria in the study area where there still needs to be conducted some integrated programmes for the eradication of malaria.

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## ABBREVIATIONS

ABER	Annual Blood Examination Rate
ACD	Active Case Detection
AP	Andhra Pradesh (India)
ASSPR	Age Specific Slide Positivity Rate
BC	Before Christ
BSE	Blood Slide Examination
DDC	District Development Committee
DDT	Dichloro-diphenyl-trichloroethane
DHO	District Health Office
DHS	Department of Health Service
EDCD	Epidemiology and Disease Control Division
EHP	Environment Health Project
HA	Health Assistant
HMG HMIS	His Majesty Government Health Management Information System
HP	Health Post
ICT	Immuno-chromatographic Test
IFAT	Immunofluorescent Assay Tests
IG	Immune Growth
KAP	Knowledge, Attitudes and Practices
ME	Malaria Eradication
MoH	Ministry of Health
MP	Madhya Pradesh (India)
MSL	Mean Sea Level
NMEP	Nepal Malaria Eradication Programme
NZFHRC	National Zoonoses and Food Hygiene Research
PCD	Passive Case Detection
PCDH	Passive Case Detection Health Institutions
PCDV	Passive Case Detection Volunteers
PCR	Polymerase Chain Reaction
PHC QBC	Primary Health Centers Quantitative Buffy Coat
RMB	Roll Back Malaria
S/'P	Sulfadoxine/Pyrimethamine
SEAR	South East Asia Region
SFR	Slide Falciparum Rate
SHP	Sub-Health Post
SPR	Slide Positivity Rate
SSPR	Sex-specific Slide Positivity Rate
VBDRTC	Vector-Borne Disease Research and Training Centre
VDC	Village Development Committee
VHW	Village Health Worker
WB	West Bengal (India)
WHO	World Health Organization

## INTRODUCTION

Of all the diseases of mankind, malaria is one of the most widespread, best-known and most devastating protozoal disease. It is a serious cause of morbidity and mortality for people living in endemic areas.

Malaria is possibly the most infectious disease of humans, infecting 5-10% of the world's population with 300-600 million clinical cases and more than 2 million deaths annually (Schofield and Grau, 2005). Malaria remains the third leading cause of death attributable to an infectious disease worldwide (Gardiner *et al.*, 2005). In Sub Saharan Africa most infections are due to *P. falciparum*.

It is a disease resulting from infection by minute parasitic protozoa of the genus *Plasmodium* which is spread from human to human through the bite of infected *Anopheline* mosquitoes. There are nearly 120 species of *Plasmodium* not only infecting man but also apes, monkeys, bird and other vertebrate host. (Bruce-Chwatt, 1993).

The four species of *Plasmodium* parasitic to man are *P. vivax*, *P. falciparum*, *P. ovale* and *P. malariae* which results in four kinds of malaria fever.

*P. vivax* (Grassi and Feletti, 1890): Benign simple or tertian malaria

*P. falciparum* (Welch, 1897): Malignant tertian, sub tertian, Aestivo-autumnal malaria

*P. malariae* (Laveran, 1881): Quartan malaria

*P. ovale* (Stephens, 1922): Ovale tertian malaria

Out of four species *P. vivax* and *P. falciparum* account for 5% of infections. Some estimates indicate *P. vivax* may account for 80% of the infections and is widely distributed in tropics, subtropics and temperate zones. (Ichhpujani and Bhatia, 1998). Among these four species, only *P. vivax* and *P. falciparum* are reported infecting the people of Nepal.

Malaria is seen in all countries extending from 40<sup>0</sup> S to 60<sup>0</sup> N of the equator covering a large portion of the tropical and subtropical regions. Highly endemic areas are often seen in the tropical regions, where humidity and temperature are favorable for the

breeding of *Anopheles* mosquitoes and growth of the parasite in the insect vector. (Anonymous, www.pubmed.com).

The life cycle of *Plasmodium* in human is complicated. Malaria parasites show alteration of generation with alteration of hosts. Man is the intermediate host and mosquitoes are the definitive hosts of the parasite.

Malaria is transmitted by the bite of infected female *Anopheles* mosquitoes. In context of Nepal, only four species of *Anopheline* mosquitoes are capable of transmitting human malaria parasites which are *Anopheles minimus*, *A. fluviatilis*, *A. annularis*, *A. maculatus*.

The incubation period varies with the parasite species and more over with the temperature. At the common tropical temperature of 80<sup>0</sup>F (=26.7<sup>0</sup>C) the incubation period has been found for

*P. vivax*: 8-31 days (Depending on vector species)

*P. falciparum*: 7-27 days (Depending on vector species)

*P. malariae*: 28-37 days

*P. ovale*: 8-31 days

The definitive diagnosis of malaria is made by the identification of malarial parasites in peripheral blood film (Park, 2000).

For many years, the standard treatment for acute malaria was chloroquine. However resistant to this drug has been developed so the most reliable alternatives to chloroquine is quinine and sulphadoxine pyrimethamine (combinely) is used.

## **Epidemiology**

The malarial disease burden is increasing in many countries despite the existence of effective preventative strategies and anti-malarial drugs. Malaria remains a major infectious disease and is worsening in some areas, particularly in Sub Saharan African where rapid urbanization has played a major role in malaria epidemiology.

At present, about 100 countries or territories in the world are considered malarious,

almost half of which are in Africa, south of the Sahara. Although this number is considerably less than it was in the mid-1950s (140 countries or territories), more than 2,400 million of the world's population are still at risk.

Malaria is thought to kill between 1.1 and 2.7 million people worldwide each year of which about 1 million are children under the age of 5 years in Africa and south of the Sahara. Malaria is Africa's leading cause of under-five mortality (20%) and constitutes 10% of the continent's overall disease burden. Malaria has been estimated to cost Africa more than US\$ 12 billion every year in lost GDP, even though it could be controlled for a fraction of that sum (RBM, 2003).

Malaria is endemic almost everywhere in Papua New Guinea. Malaria remains a serious health problem in coastal and inland regions comprising 15 provinces. It is endemic up to an altitude of around 1200-1500 meters, where it becomes epidemic. In these areas, transmission is persistently high throughout the year, with *P. falciparum* causing an estimated 75% of infections. Malaria is the third leading cause of hospital admissions and deaths (Anonymous, [www.wpro.who.int](http://www.wpro.who.int)).

It is estimated that 1.2 billion people out of the 1.4 billion people of SEA Region live in malarious areas. In 1995, malaria cases in the region were estimated to be 21.9 million, with almost 32,000 deaths. India accounts around 85% of the total reported cases in the region in the same year. India contributed 83% of total malaria cases in SEA Region (Lal *et al.*, 1997).

Malaria has also been a major public health problem in Bangladesh. Approximately 88% of the 128 million populations are at risk of malaria. Majority of malaria cases are reported from 13 out of the total 64 districts in the country. The cases recorded were 68,594 in 1997; 60,023 in 1998; 63,723 in 1999; 55,599 in 2000 and 55,646 in 2001 (WHO; SEARO, 2002).

Of the country's total population 658,000 in 1998, populations residing in malarious areas of Bhutan were 427,000. The outbreak of 1999 reported 12,237 cases of malaria with 16 deaths but the cases reported in 2001 were 5,962 with 25 deaths (WHO; SEARO, 2002).

The populations of Indonesia were 203 million in 2000. Of the total population, 149.7 millions reside in malarious areas. Malaria is a health problem in forest related areas of the outer islands, particularly in the eastern part of Indonesia. Approximately 1.5 million cases are detected annually (WHO; SEARO, 2002).

Out of 19.3 million population of Sri Lanka, 9.3 million resides in malarious areas. During the year 2001, there has been a very significant reduction in the number of malaria patients recorded as compared to the previous years. From January-August 2001, 50,116 confirmed malaria patients were detected from a total number of 925,893 blood smears examined (Pv - 81.3%; Pf - 18.7%). During the previous year, approximately 200,000 confirmed patients were recorded. Around 50% of patients were from the districts of the North-East Province, which had many serious obstacles to malaria control due to the conflict situation (WHO; SEARO, 2002).

Malaria, in its various forms, has been the cause of mortality in Nepal throughout the ages. It has constituted one of the most important causes of economic misfortune engendering poverty and intellectual standards of the nation and hampering prosperity and economic progress in every way (HMG, 2005). During the last four decades there has been fluctuation in the status of malaria situation in Nepal. During the pre-central era, malaria was hyper/mesoendemic but large parts of the country, particularly Southern Nepal was prone to epidemics.

There is no documented record about the prevalence of malaria in Nepal except a few historical descriptions. The first documented epidemiological survey dates back to 1925 by Major Phillips of Indian Military service in Makawanpur valley and Chitwan. The mortality rate in children was estimated at about 43% among Pahadis (hill people) and 17% among Tharus (Tribal of the Terai areas) (Bista *et al.*, 2002).

The decade of the nineties also experienced a periodic malaria outbreak in Central and Far Western Region reporting 29,000 cases in 1991 (Annual report, EDCD-2001).

Sherchand's (1996) study on resurgence of malaria in southern Nepal revealed that ignorance of people's beliefs regarding behavior towards and the disease results in the



development of inappropriate control programs that are not designed according to local conditions and needs. The extensions of urban areas lead to epidemics in the peripheries of the growing cities. Mass migrations of non-immune populations into endemic areas for political reasons further complicate matters. It has been predicted that climate changes might cause some modifications to the present global distribution of malaria close to its present boundaries (Hay *et al.*, 2000).

Up to the 1950s (before the ME activities were undertaken) it was estimated that approximately two million cases of malaria (40% of the total population) occurred annually and ten to fifteen percent among those resulted in death (EDCD, 2001). At present, out of Nepal's total population of 23.2 million approximately 17.3 million (74%) of the people are at risk distributed at 64 districts in 5 development regions (DoH, 2001-2002).

The massive outbreak of malaria in early seventies in Parsa district of Central Region and Nawalparasi, Rupandehi and Kapilvastu district in Western Region resulted because of resistance of the vector *A. annularis* against DDT. Similar outbreaks were reported during eighties in Far Western Region with smaller outbreak in Central Region in 1985 to 1988. In all the years the cases were above 15,000 escalating to as high as 42,231 in 1985. In 1987 and 1988 malaria control program in all districts was integrated into the basic health services as a component of primary health care at district/health post level (Annual report, EDCD-2001).

### **Control Programmes**

Malaria continues to be a major health problem. WHO launched a worldwide malaria eradication programme in 1955. This continued until 1970, after that it was officially declared a failure. In 1998 a second program coined "Roll Back Malaria" was started with the declared objective of halving the global burden of malaria by 2010. Its founding partners the UNDP, UNICEF, the World Bank and WHO agreed to share their expertise and resources in a concerted effort to tackle malaria worldwide with a particular focus on Africa. The main objective of RBM is to reduce the global malaria burden significantly through interventions adapted to local needs and strengthening of the health sector with a high political commitment. Nepal has also adopted RBM

project.

Already, malaria control programme was integrated into the basic health services and included in National Health Policy, 1991. In 1995, WHO calls for the implementation of a reliable reporting system capable of early detecting and monitoring new emerging and re-emerging diseases. Since HMIS (Routine Reporting) reports received late, not all cases that were seen and diagnosed by a health institution were reported and investigated adequately, lack of case definitions, untrained (and unmotivated) health staff and lack of laboratory services; in 1997 an additional surveillance system, EWARS was established. Periodical exacerbation in hilly area has been attributed to introduction of source of infection, fairly developed health infrastructure and remote accessibility. Knowledge, attitudes and practices of people play important role.

There is a need to study human behavior in relation to socio-economic and environmental aspects, which are related to the vector behavior, before implementation of any control activities.

### **Significance of the Study**

The present work is aimed to seek relationship between risk factors and disease burden so as to assist the control strategy. HMG reports on malarial prevalence in Sunsari district and other previous studies conducted in the areas of some ecological belt still indicate that there is still existing considerable malarial prevalence in the eastern tropical part of the country. Sunsari district ranked fifth top district in east region for 2003 year as it alone showed 895 suspected malaria cases in the year (HMG, 2005). In this endeavor, any study revealing the prevalence of malaria in Mahendranagar VDC, which is the largest VDC with greatest population than in any VDCs of Nepal, would be very advantageous.

## **II**

### **OBJECTIVES**

#### **General Objective**

The general objective of the study is to determine the prevalence of malaria in relation to socio-economic and environmental factors in Mahendranagar VDC of Sunsari district.

#### **Specific Objectives**

- ) To determine the prevalence of malaria in Mahendranagar VDC of Sunsari district.
- ) To assess the socio-economic and environmental risk factors of malaria.
- ) To aware the people of Mahendranagar VDC about malaria.

### **III**

## **MATERIALS AND METHODS**

### **Materials**

Microscope, cotton wool, beaker, sterile lancets, slide and slide box, staining troughs, timing clock, cover slips, measuring cylinder, slide drying rack and record form or register

### **Chemicals**

Giemsa's stain, distilled water, methylated spirit, water and methanol

### **Study Area**

The study was carried out in Mahendranagar health post and Janasewa clinic located in Mahendranagar VDC of Sunsari district. Mahendranagar VDC lies in Sunsari district which is bordered with India in the south, Morang district in the east, Dhankuta district in the north and the Koshi River in the west. The VDC is the largest VDC and incorporates largest number of population than in any other VDCs of the country (C.B.S). It is located between Koshi River in the west, Singhiya, Prakashpur and Bharauli VDCs in the east, south and north respectively.

The VDC is characterized by different ethnic groups comprising Brahmin, Chhetri, Tharu, Newar, Tamang, Rai and Damai-Kami-Sharki-Majhi. There is one sub health-post and one laboratory clinic. Agriculture is the main source of economy. The houses are often with adjacent cattle sheds made of bamboos with thatched roof. The annual rainfall ranges from 1400 to 2200 mm, and the relative humidity varies between 60% and 85%. The temperature ranges from 10 - 42<sup>0</sup> C.

### **Study period**

The total study period was of 12 months from October 2004 to September 2005.

### **Study Population**

A total of 250 symptomatic patients having fever for 2-3 days, anemia, headache, splenomegaly and hepatomegaly were included for the study.

### **Sample collection**

Among the patients attending the health post and clinic of Mahendranagar VDC, only those patients were selected for the study who had complains. They were explained about the purpose and procedure of the study. Basic information and specific history were recorded from the suspected patients. The blood was withdrawn from the symptomatic patients by piercing the third finger of the patient's left hand after cleaning with an alcohol swab spirit. The blood was dropped on the slide to prepare the thick and thin smears.

### **Sample processing**

The thick and thin blood smears were prepared on the same slide taking three drops of blood and spreading in an area of 10mm, thick smears was prepared 10mm away from the edge of the slide. A single drop was taken for thin smear. It was uniformly spread bringing the spreader at an angle of 30<sup>0</sup>-45<sup>0</sup> from the horizontal and pushing the spreader steadily down the surface of the slide drawing the blood behind till the smears was formed. The smears were air dried (for 10 minutes). After drying, the thin smear was fixed in methanol. This was done by either dipping the thin smear into methanol for 5 seconds or by dabbing the thin smear with a methanol soaked cotton ball. While fixing the thin smear, all care was taken to avoid exposure of the thick smear to methanol.

The slides were placed in staining rack ensuring that thick films were placed along the end of rack. Thin film was obtained with 3% Giemsa's solution for 30 minutes. The stain was washed under tap water. The slides were kept with the film side downward in the drying rack and observation was done with the help of microscope under 40x and 100x.

### **Microscopic examination**

The blood films were examined microscopically. In the thick film only parasites present are more quickly seen under the microscope as they are concentrated in or smaller area than in thin film. The thin film is used as label to identify the species of the malaria parasite or other morphological characters of the parasite which are not visible in thick film preparation. The malarial parasite can be recognized by observing the shape and colour of stain of chromatin and cytoplasm of the parasite. The

chromatin (part of nucleus) is usually round in shape and stains deep red. Cytoplasm varies in shape from ring shape to a totally irregular shape. It always stains blue but the shade of blue may vary between the malarial species. Staining of slides and preliminary examination were carried out in Mahendranagar sub health post and Janasewa laboratory clinic. Cross checking and final confirmation of slides were done in OM Hospital's Laboratory, Kathmandu. The slide examination was carried for the malaria parasite and cell morphology.

### **Questionnaire survey**

After history taking and microscopic examination all findings were recorded in the printed questionnaire. During questionnaire survey, the questions were asked verbally to each patient. The questions asked to them were about their general information i.e. name, age, address, sex, economic status, education, knowledge about the disease etc.

### **Data collection**

The data collected was based on the primary as well as secondary data. Primary data was collected from questionnaire survey and microscopical findings. Secondary data was taken from the published and unpublished sources.

### **Data analysis**

Data collected were presented and analyzed using appropriate statistical tools.

## IV

### LITERATURE REVIEW

This chapter reviews the fundamental literature on malaria and various researches on it. Malaria continues to be a major cause of mortality and morbidity especially throughout the developing world.

It is a very old disease and prehistoric man is thought to have suffered from malaria. It probably originated in Africa and accompanied human migration to the Mediterranean shores, India and South East Asia. In the past, it used to be common in marshy areas around Rome and the name is derived from the Italian word, mal-aria or "bad air". It was also known as Roman fever.

It was in 1880 that Laveran, a French army surgeon in Algeria, first saw and described malaria parasites in the red blood cells of man. He gave the name *Oscillaria malariae* in 1881 to what is now known to include the three principal *Plasmodia* of man, and was the first to describe crescentic gametocytes. Although Richards, in 1882, confirmed the observations of Laveran, they were not accepted generally until 1885 because of the belief that the disease was of bacillary etiology due to the *Bacillus malariae* of Klebs and Crudeli (1874). In 1891, Grassi and Feletti initiated studies of the avian *Plasmodia* which led to the disclosure of the sporogonic cycle and the transmission of malaria by mosquitoes. In 1895, Ross observed the exflagellation in the stomach of mosquito. In 1898, Bignami, Bastianelli and Grassi discovered the zygote of the parasites of human malaria in *Anopheline* mosquitoes and demonstrated the transfer of infection from man to man by mosquitoes. Between 1948-1955, the pre-erythrocytic cycles of the malarial parasites of man and mammals were demonstrated by Shortt and his co-workers.

In the last 25 years or so a number of significant advances have been made that have the potential to make a major contribution to the control of this disease.

## **Some recent findings on Malaria in Global context**

Hazra *et al.*, (1998) found that malaria had been changing its clinical profile by studying 60 cases of *P. falciparum* and 165 cases of *P. vivax* clinically along with species identification at Calcutta. The classic paroxysm is evident only in 40 cases of *P. falciparum* and 47.27% of *P. vivax* malaria, but the difference between the two groups is not statistically significant. In 40% of *P. falciparum* and 27.7% of *P. vivax* cases continuous or remittent types of fever has been observed. Absence of classic paroxysms of fever in association with splenomegaly when present, poses a diagnostic difficulty with enteric fever. Association of jaundice in 40% and 9.09% cases with *P. falciparum* and *P. vivax* respectively along with hepatomegaly in 80% and 63.63% in them in conjunction with nausea and vomiting leads to clinical mimicry with infective hepatitis. Splenomegaly which has been described as cardinal feature of malaria was deserved in 40% cases with *P. falciparum* and only in 18.18% cases of *P. vivax* malaria and this is a clear deviation from earlier description.

Gunawardena *et al.*, (1998) studied malaria for 18 months in a population of 1,875 residents in 423 houses in an endemic area in Southern Sri Lanka and found the risk of malaria to be 2.5 fold higher in residents of poorly constructed house than in those living in house of good construction type.

Batra *et al.*, (1999) reported the breeding of *Anophele* mosquitoes in irrigated areas of south Punjab and Pakistan facilitating in increase of *Anophele* mosquitoes.

Praetorius *et al.*, (1999) described the importation of *Anopheles* mosquitoes in luggage or aeroplane. The blood smear of an inhabitant near the Frankfurt airport, admitted with pyrexia of unknown cause revealed *P. falciparum*. She had no history of travelling any malaria infected regions and the source of malaria could not be identified "Baggage malaria" is more likely than the bite of a mosquito expelled from the "planes under carriage." Imported malaria should be the case of fever of unknown case without history of foreign travel as there are no screening procedures; a blood smear preparation was suggested.

Bangali, (2000) mentioned that approximately 88% of the 128 million people in Bangladesh are at malaria risk. *P. falciparum* is the most predominant species and the current situation of the country according to the paper presented Bangali is seen to be declining every year.



Chirebvu *et al.*, (2000) reported the epidemiological and clinical survey and evaluative aspects of severe malaria in Antananarivo. This retrospective study included 48 children less than 15 years old, hospitalized at the paediatric limit Debre of the center Hospitalize Universities de Befelantanana (Antananarivo) for severe malaria as defined by WHO criteria. The hospitalization frequency was 0.87% and higher frequency was 0.87% and higher frequency was noticed for the children less than 5 years old and the sex ratio was 1.4/1. The cerebral complications as seen in many African countries were the most frequent clinical form. The death rate was 14.58% and the proportional mortality was 1.07%, 2.1% of the patients had sequel. The improvement of severe malaria prognosis was not only on better equipment in intensive care wards, but also on improved and early diagnosis and management.

Kitchener *et al.*, (2000) analyzed malaria in the Australian Defense Force during an after participation in the International Force in East Timor (INTERFET). Malaria in the Australian Defence Force members has been for more common in East Timor than in other recent overseas deployments. By 6 months after all 5500 members of the International Force in East Timor had return to Australia, 267 malaria infections had been reported to the Army Malaria Institute. Only 64 of those affected had their first clinical episode during their 4-5 months in East Timor and about two-thirds of these infections were caused by *P. faciparum*. The remaining 212 soldiers developed their first symptoms after retiring to Australia, and all but two infections were caused by *P. vivax*. After treatment, 44 soldiers had relapses of their *vivax* infection; 11 had a second relapse and 2 had a third relapse. The findings raise several issues about prevention and management of malaria in the Australian Defense Force.

Mukhopadhyay *et al.*, (2000) surveyed on the malariogenic situation amounting to premonition in areas of Ajodhy Hills of District of Purulia, West Bengal. Since long time, Purulia District in West Bengal has been endemic for malaria. In 1997 and 1998, the district contributed 12.4% (9932 out of 79811) and 10%\$ (13248 out of 130288) of malaria cases respectively occurring in West Bengal, resulting 9.45% (7 out of 74) and 5.5% (4 out of 72) of deaths respectively in the state. Annual Blood Examination Rate (ABER) of the district was 10.4% in 1997 and 8.5% in 1998.

Zangpo *et al.*, (2000) reported that about more than half the country's population of Bhutan is at risk of malaria. The malaria situation started to worsen from 1990 on

ward with a peak in 1994. The percentage of *P. falciparum* ranged from 31.5% to 51%.

Olliario *et al.*, (2001) proposed that anti-malarial drug resistance is major public health challenge and the principal reason for the erosion of efficacious treatments. Cost and the limited number of anti-malarial drugs in current use impose considerable constraints on malaria control, especially in sub-Saharan Africa. The paper describes multilateral, multidisciplinary research project on artemisinin-based combination therapy, which offers a new and potentially highly effective way to prevent or retard the development of drug resistance.

Simonsen *et al.*, (2001) investigated "One Spot" synthesis and anti-malarial activity of formamidine derivatives of 4 anilinoquinoline. Amodiaquine (AQ) is an anti-malarial which is effective against chloroquine resistant strains of *P. falciparum* but whose clinical use is severely restricted because of associated hepatotoxicity and agranulocytosis, "One Spot" synthesis of formamidines likely to be transformed into AQ derivatives is reported.

Trisirivanich *et al.*, (2001) suggested that anti-malarial drugs clear resistant parasites from partially immune host. Circumstantial evidence in human malaria suggests that elimination of parasites by drug treatment meets higher success rates in individual having some background immunity.

Bismildin *et al.*, (2001) reported about the current malaria situation in the Republic of Kazakhstan. Chuan *et al.* (2001) described the current status of human parasitic infections in Taiwan in the Journal of Microbiology and Immunology. The eradication of the two mosquito-borne parasitic diseases-malaria and lymphatic filariasis, is one of the greatest achievements of the parasitic control campaigns in Taiwan.

Flick *et al.*, (2001) found that infections with *P. falciparum* during pregnancy lead to the accumulation of parasitized RBCs in the placenta. Hamilton, (2001) explored the hypothesis of parasite mediated sexual selection and host-parasite co-evolution maintains variation in male genetic quality and allows for strong inter sexual selection in species with high rates of infection by using molecular techniques to examine variation in the prevalence of *Plasmodium* species.

Orago *et al.*, (2001) analyzed hematological parameters in patients and individual residents of *P. falciparum* malaria in holoendemic area of western Kenya. The analysis of service statistics showed that the annual blood examination rate (ABER) increased in comparison to the previous year whereas the slide positivity rate (SPR) decreased. This indicates that slides were collected from among the inhabitants of endemic areas who probably may not have evident clinical picture of malaria. Previous year's higher SPR may also have been related to the outbreak situation in some of the areas. Decreased API indicates better implementation of malaria control including IRS and CDPT. This is important to make a note of the fact that Pf % over the past few years shows increasing trend indicating the resurgence of falciparum malaria. Therapeutic efficacy of SP combination towards Pf cases needs to be assessed meticulously in this connection. There is a 40% increase in the imported malaria cases in the country. This in the face of increasing trend of Pf % over the years gives thought for cross border monitoring of malaria cases in the country (Malaria profile FY 1996/97 to 2000/2001).

Bell *et al.*, (2001) compared the efficacies of remote symptom - based diagnosis of malaria, rapid diagnostic tests and microscopy in an area of low endemicity in the Philippines. In trial, 1,350 symptomatic patients were tested using *P. falciparum* and *P. vivax* immune chromatographic tests (ICT - tests) and blood films stored and read under local conditions. The result revealed that a history of fever alone was sensitive (95.4%) but poorly specific (16.5%) for predicting parasitemia. ICT tests achieved high sensitivity (97.9%) but many cases indicated as positive by ICT tests were found to be negative by microscopy. Analysis in trial I indicated that ICT tests were detecting low level parasitemias missed by microscopy and that local microscope has poor accuracy.

Kengne *et al.*, (2001) proposed a multiplex PCR-based method derived from random amplified polymorphic DNA (RAPD) markers for the identification of species of the *A. minimus* group in southeast Africa. Effective control of *A. minimus*, an important malaria vector in south East Asia, is based on the accurate identification of species within *A. minimus* complex, which cannot be distinguished using morphological characters.

Schoone *et al.*, (2001) presented a journal on detection and quantification of *P.*

*falciparum* in blood samples using quantitative nucleic acid sequence-based amplification essay (QT-NASBA). Primers and probes were selected on the basis of the sequence of the small subunit rRNA gene. Quantification was achieved by co-amplification of RNA in the sample with one modified in vitro RNA as a competitor in a single ranging from 10 to 10<sup>8</sup> (*P. falciparum*) per ml. could be demonstrated and quantified in whole blood. This is approximately 1,000 times more sensitive than conventional microscopy analysis of thick blood smears. QT-NASBA may be especially useful for the detection of low parasite levels in patients with early stage of malaria and for the monitoring of the efficacy of drug treatment.

Tarimo *et al.*, (2001) suggested malaria diagnosis and treatment under the strategy of the integrated management of childhood illness (IMCI) with a laboratory support from the rapid immuno-chromatographic tests or ICT. To detect and measure the development of the liver stages of malaria parasite in mice infected with sporozoites ranging in number from 25 to more than 164000, a highly sensitive PCR is used. It also detects the parasite loads in the liver of mice due to bite of single anopheles mosquito. Such study evaluates very precisely this efficacy of anti-malarial experimental drug treatments and vaccination regimens in conditions of infections resembling those found in the field (Bruna *et al.*, 2001).

Targett *et al.*, (2001) described that artesunate reduces but does not prevent post treatment transmission of *P. falciparum* by *A. gambiae*. Combination therapy that includes artemisinin derivatives cures most falciparum infections. Lowering transmission by reducing gametocyte infectivity would be an additional benefit. To examine the effect of such therapy on transmission, Gambian children with *P. falciparum* malaria were treated with standard regimens of chloroquine or pyrimethamine sulfadoxine alone or in combination with 1 or 3 doses of artesunate. The infectivity of mosquitoes of gametocytes in peripheral blood was determined 4 or 7 days after treatment. Infection of mosquitoes was observed in all treatment groups and was positively associated with gamatocyte density. The probability associated with gametocyte density. The probability of transmission was lowest in those who received pyrimethamine sulfadoxine and 3 doses of alone.

Desai *et al.*, (2002) reported 912 cases of malaria among persons in the United States with a date of onset between January 1, 2002 and December 31. 2002. The infecting

species of *Plasmodium* was identified in 753 (82.6%) of these cases. 910 (99.8%) of the 912 cases were imported. 591 (64.9%) of the 910 cases were in U.S. residents (includes both civilians and military personnel) who acquired the infection outside the United States. Of the 591 cases, 395 (66.8%) were acquired in Africa, 63 (10.7%) in the Americas and 99 (16.8%) in Asia.

Guenther *et al.*, (2002) estimated the frequency of renal dysfunction in falciparum malaria in total of 108 adult patients. It was found that using a sensitive marker 55% of patient have a reduced Glomerular Filtration Rate (GFR).

Cortes *et al.*, (2003) surveyed on malaria in Manitanian. It was found that malaria infection rate was 18.5% (77 of 446), *P. falciparum* caused 61.85% of these, *P. vivax* 35.5% (28 of 77) in Nouakchott. In Kaedi 106 of 416 cases were recorded with *P. falciparum* as the sole pathogenic species.

A study was done by Mueller *et al.*, (2003) in 709 representative children aged 6-31 months in rural Burkina Faso to describe the pattern of malaria morbidity treatment seeking behaviour and mortality. Their findings call for more effective prevention and treatment of malaria as well as for better supervision of existing malaria treatment guidelines in formal health services.

Mulberger *et al.*, (2004) reported a total of 4,801 patients with travel-related malaria within the 16 Trop net Europe network. Within the surveillance period 4,801 cases of imported malaria were reported. *P. falciparum* was leading number followed by *P. vivax*. European travelers and immigrants were the largest patient groups, but their proportion varied among the reporting countries. The main regions of infection in descending order were the Indian subcontinent, Indonesia, South America and Western and Eastern Africa, as a group accounting for more than 60% of the cases. All 16 Trop Net Europe countries reported *P. vivax* malaria. However, the number of cases varied strongly between countries. Germany (24.3%), Spain (15.5%) and the UK (12.0%) reported most cases, whereas reports from Switzerland (1.8%), Poland (1.6%), Finland (1.0%), Ireland (1.0%) and Portugal (0.3%) were scarce.

Baruah *et al.*, (2005) evaluated drug sensitivities of *P. falciparum* in four endemic villages of the Sonitpur District of Assam, involving 218 cases who were tested in

vivo over 35 days. Chloroquine resistance was detected at the RI level in 29 cases (13%) and RII level in 8 cases (4%). No RIII chloroquine resistant cases were detected in the study. RI resistance was observed in the age groups 6-10 years, 11-14 years, and 15 years and above in 16%, 17% and 13%, respectively. RII level resistance was observed in 4% of all those groups combined. All the RI and RII resistant cases responded well to a single dosage of Metakelfin (sulfamethoxypyrazine I.P 1,500 mg and pyrimethamine I.P 75mg).

Bell *et al.*, (2005) studied the influence of several malaria risk factors and volunteer health worker (VHW) accessibility on parasite prevalence and treatment-seeking in a remote area of Mindanao, the Philippines. It was found that parasite prevalence was significantly higher among patients living in villages lacking a resident VHW (adjusted OR=3.88, p=0.02), where proposed delays in consulting VHWs and the official health service, and the use of alternative medicine, were also significantly higher.

Planche *et al.*, (2005) studied on the relevance of malaria pathophysiology to strategies of clinical management. The findings review about the pathophysiology of these complications and the implications for future adjunctive therapy of malaria, including the proposed importance of fluid volume depletion and sequestration of parasitized red cells in severe malaria.

From the survey of Harris *et al.*, (2005) it was found that malaria is a growing problem in the Bolivian Amazon where there has been a four-fold increase between 1991 and 1998, largely owing to forest clearance bringing human and vector into closer association. The principle vector in this region is *Anopheles darlingi* root, the behaviour of which has been little studied in this part of South America. Peak biting occurred between 19:00 and 21:00 hours, when 48% of the total night's biting took place. This early biting habit has implications regarding control of malaria via the use of insecticide-treated bed nets. *Anopheles darlingi* was the most prevalent vector in the study, although *A. albitarsis* s.1. and *A. braziliensis* were also present.

Kimbi *et al.*, (2005) evaluated the prevalence and consequences of malaria infection in two hundred and forty six randomly selected school children aged three to sixteen years in the Muea area and its environment. It was found that prevalence of malaria

was 98% and highest prevalence rate (100%) and geometric mean parasite density (1520 parasites/microl of blood) occurred in the  $\leq 5$  years age group. Prevalence of anaemia was generally low (10.8%) and there were no cases of severe anaemia (PCV  $< 20\%$ ). *P. vivax*-like parasites were detected for the first time in this area. Plasmodium falciparum was the predominant species (93%) followed by *P. malariae* (52%), *P. ovale* (42.7%) and *P. vivax*-like parasites (33.3%).

### **Some recent findings on Malaria in Nepalese context**

From the data available since 1963 the malaria in Nepal reveals periodic upward lifts followed by sharp falls in next two years and then the period of stagnation with slide fluctuation between 2,500 to 4,000 cases lasting for 5-6 years (1963-1971). The cases started rising from 1972 and in 1973 and 1974 the number of cases reached to 14,000 due to the epidemic in the west region. The year 1977 could contain malaria in between 10,000 and 13,000 cases annually.

From 1978 the real deterioration of the situation started when the cases reached to 14,212 in 1978 from 11,615 in 1977. With steady increase every year, the number of cases reached to 16,719 in 1983. There was steep rise to 29,388 cases in 1984 which again escalated to 42,321 in 1985. After 1985, the cases in the country started decreasing. By 1989 the cases again increased to 22,366, which reached to 22,856 in 1990. After 1991 the number of cases gradually decreased and reached to 8,498 in 1998 (HMG, MoH, VBDRTC, 1999).

The country reported 42,321 and 22,333 malaria cases in 1985 and 1989 respectively. *P. falciparum* cases reduced at the same rate as the total malaria cases, but after 1988 the reduction of the former, especially indigenous *P. falciparum* cases was slightly accelerated (Bista *et al.*, 2000)

During 1994 to 1995, 54.67 and 54.77 prevalence rate of malaria cases were detected, out of them 72.02 and 63.13 incidence rates were imported from India. Among the 26 bordering districts of Nepal, Kanchanpur and Kailali of the far western, Bardia of mid-western, Nawalparasi of western, Dhanusa, Mahottari of Central and Morang and Jhapa of the eastern regions are the main contributors of malaria cases. The total malaria cases of 26 bordering districts constituted 64.44% of the total malaria cases in 1997 (Bista *et al.*, 2000).

Sherchand *et al.*, (1998) suggested that the patients with encephalitis and meningitis which are manifestations of central nervous system must be diagnosed with high index of suspicious of malaria. Medical officers are trained to recognize clinical malaria by symptom which is characteristic of vivax malaria. There is a need to maintain medical officers so as to detect as well as to treat severe form of malaria mostly occurring with falciparum malaria since malaria does not always present as characteristic "vivax paroxysm."

Sherchand *et al.*, (1999) examined thick blood films in rural endemic areas of southern Nepal by comparing the dipstick and Parasight-F test. The study revealed that the unpredictable cross-reactivity with *P. vivax* may render it difficult to use in situations where *P. vivax* is the predominant species, even though Parasight-F test is easy to perform in field conditions. This disadvantage could be overcome as there are possibilities of an early diagnosis of *P. falciparum* malaria and preventing the development of severe malaria.

Bista and Banerjee (2000) presented some data of *P. falciparum* resistant to chloroquine. During 1979 and 1990, a total of 178 and 84 *P. falciparum* cases were monitored by in-vitro and in-vivo method respectively. A resistance of 63.2% was recorded to chloroquine by in vitro test. Out of 84 in vitro tests, 32 cases showed a resistance of 38% at S/RI and RJI level. No resistance was found to methoquine and sulfadoxine / pyrimethamine. Therapeutic efficacy monitoring has revealed rate treatment failures among recipients of S/P treatment. The current first time treatment of microscopically diagnosed *P. falciparum* is S/P in Nepal.

Chowdhury (2002) found a new rapid method of staining blood cells and malaria parasites with R.C. stain. The traditional methods for staining blood films with Leishman's/Wright-Giemsa's stain for identification of blood cells and blood parasites are good but they are time consuming, cumbersome and require costly reagents. Moreover, the beginners viz, medical students make errors like understanding, over staining or precipitation of stain leading to difficulty in identification of cells. To alleviate these problems and to help the workers of the malaria-eradicating programme, this rapid and simple method has been devised to stain blood cells and malaria parasite with in 50 seconds.



## V RESULTS

A total of 250 blood samples were collected from suspected cases having some symptom of malaria visiting at health post and Janasewa Clinic at Mahendranagar VDC during October 2004 to September 2005.

### )] **General prevalence of malaria**

Among 250 blood samples, only 10 were found to be infected with malaria i.e. the slide positivity rate was found to be 4%.

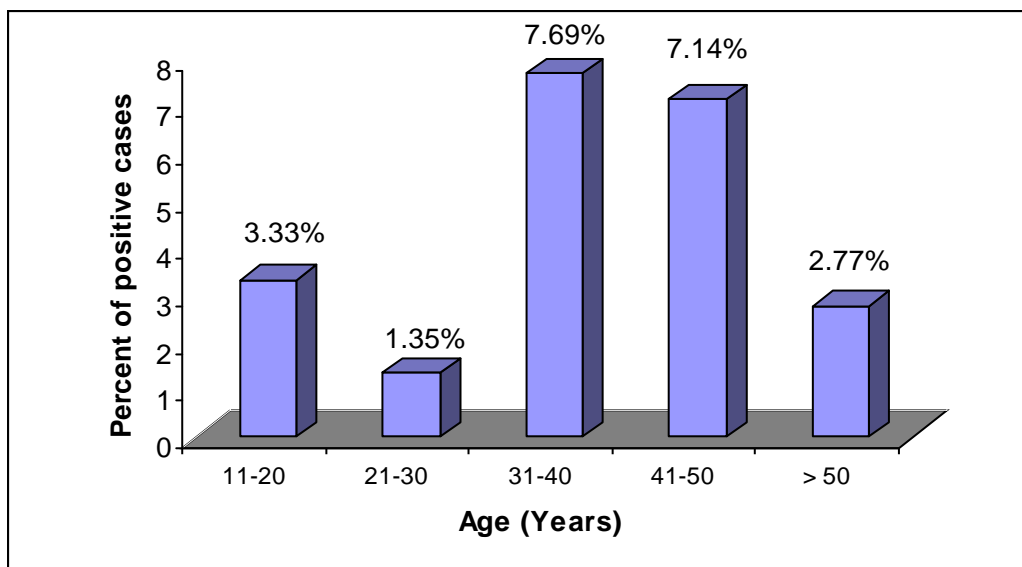
### )] **Agewise prevalence of malaria**

The result of the study indicates that the maximum number of positive cases was from the age group 31-40 years and minimum from 21-30 years and above 50 years. Of the total infected population, highest age specific slide positivity rate was found to be in age group 31-40 years (7.69%) and lowest in 21-30 years age group (1.35%).

**It was found that there was no significant relation between the age and the disease ( $t^2=4.113, P>0.05$ ).**

**Table 1: Agewise prevalence of malaria**

Age (years)	Number of total observed cases (n=250)	Malaria	
		Positive cases (n = 10)	Percent (%)
11-20	60	2	3.33
21-30	74	1	1.35
31-40	52	4	7.69
41-50	28	2	7.14
> 50	36	1	2.77



**Figure 1: Age-wise prevalence of malaria**

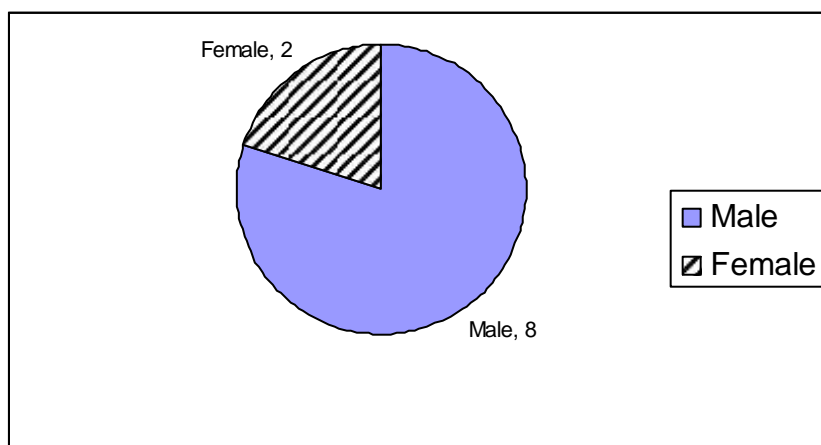
### ) Sex wise prevalence of malaria

In relation to sex, out of 150 male suspected cases, 8 were positive for malarial infection and out of 100 female suspected cases, 2 were positive for it i.e. slide positivity rate was 5.33% and 2% respectively.

**Malaria infection was found to be insignificantly related with sex ( $t^2=1.72$ ,  $P>0.05$ ).**

**Table 2: Sexwise prevalence of malaria**

Sex	No. of total observed cases (n=250)	Malaria	
		Positive cases (n=10)	Percent (%)
Male	150	8	5.33
Female	100	2	2.00



**Figure 2: Sexwise prevalence of malaria**

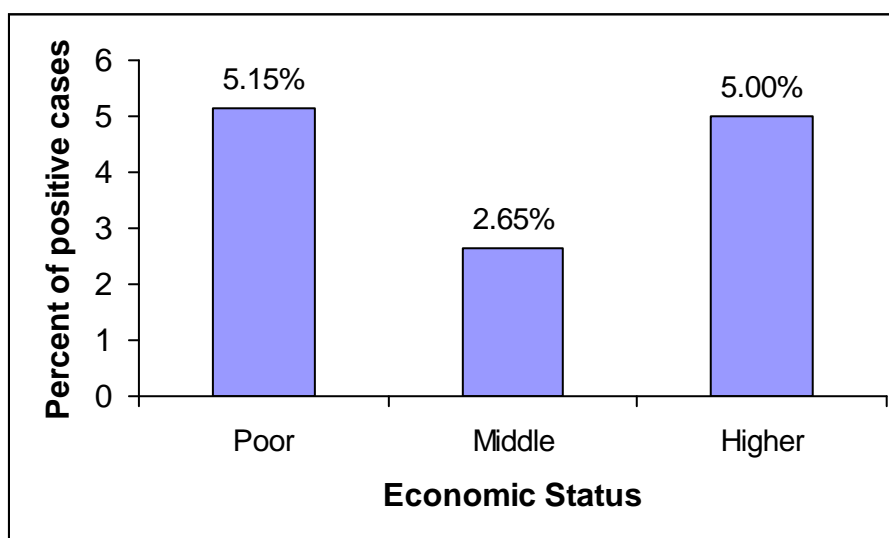
### J) Prevalence of malaria on the basis of socio-economic aspect

Out of 250 suspected cases, the majority of cases were of middle class income group (113 cases) but the higher malaria infection was from poor class i.e. 5 out of 97 cases (5.15%).

The presence of malaria was found to be insignificantly dependent on the socio-economic aspect ( $t^2=0.969$ ,  $P>0.05$ ).

**Table 3: Prevalence of malaria on the basis of economic status**

Economic Status	No. of total observed cases (n=250)	Malaria	
		Positive cases (n=10)	Percent (%)
Poor	97	5	5.15
Middle	113	3	2.65
Higher	40	2	5.00



**Figure 3: Prevalence of malaria on the basis of economic status**

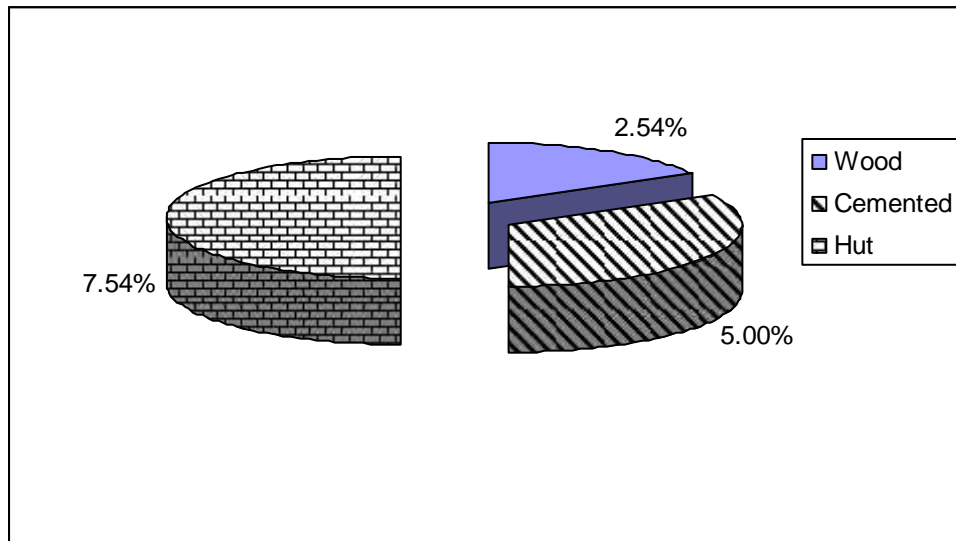
### Prevalence of malaria on the basis of type of houses

Similarly, in case of housing condition 157 houses were made of wood, 40 houses were cemented, 53 houses were of hut and the slide positivity rate was found to be the highest in the population living in hut houses 7.54% (4 cases).

It was found that the malaria infection and the type of houses was not significantly related ( $t^2=2.69$ ,  $P>0.05$ ).

**Table 4: Type of houses and infection of malaria**

Type of houses	No. of total observed cases (n=250)	Malaria	
		Positive cases (n=10)	Percent (%)
Wood	157	4	2.54
Cemented	40	2	5.00
Hut	53	4	7.54



**Figure 4: Type of houses and infection of malaria**

### ) Prevalence of malaria in relation to religion

Religionwise distribution of the population during the study showed Hindu to be 215, Muslim 27 and Buddhist 8. Among 215 Hindu population, 10 (4.65%) patients were found to have malaria infection while none of Muslims and Buddhists were positive for it.

**There was insignificant relation between malaria infection and religion ( $t^2=1.70$ ,  $P>0.05$ ).**

**Table 5: Prevalence of malaria in relation to religion**

Religion	No. of total observed cases (n=250)	Malaria	
		Positive cases (n=10)	Percent (%)
Hindu	215	10	4.65
Muslim	27	0	0.00
Buddhist	8	0	0.00

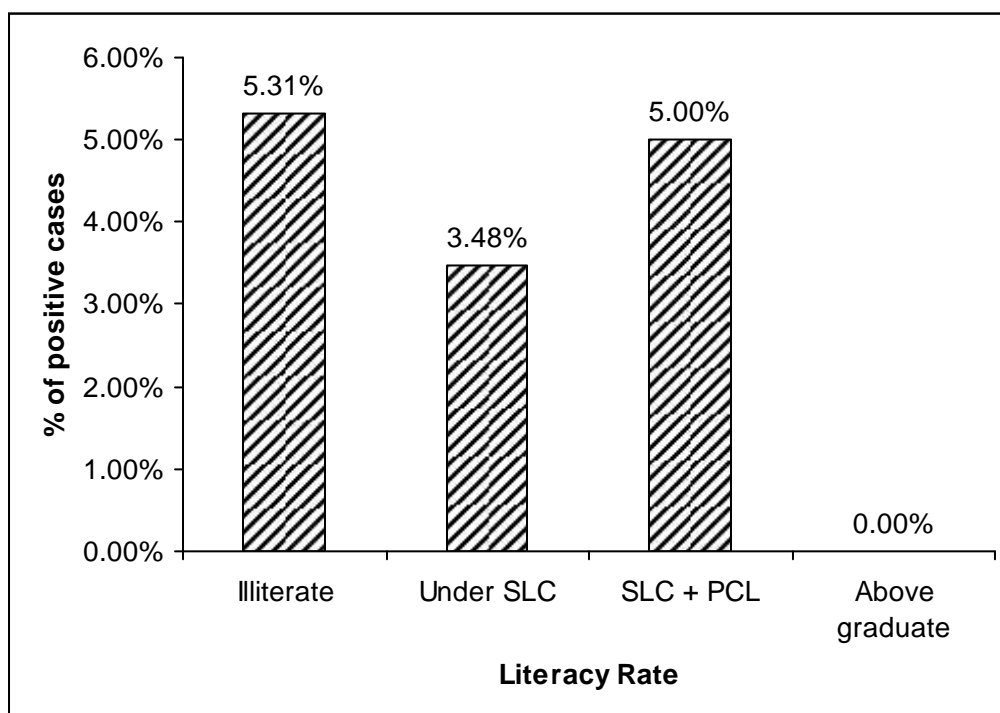
## J Literacy rate and malaria infection

Among 250 study population, majority of cases were from illiterate. The maximum number of positive cases was found in illiterate 5 (5.31%), followed by under SLC 3 cases (3.48%) and SLC + PCL 2 cases (5%).

The malaria infection and the literacy rate was found to be insignificantly related ( $t^2=1.869, P>0.05$ ).

**Table 6: Literacy rate and malaria infection**

Literacy rate	No. of total observed cases (n=250)	Malaria	
		Positive cases (n=10)	Percent (%)
Illiterate	94	5	5.31
Under SLC	86	3	3.48
SLC + PCL	40	2	5.00
Above graduate	30	0	0.00



**Figure 5: Literacy rate and malaria infection**

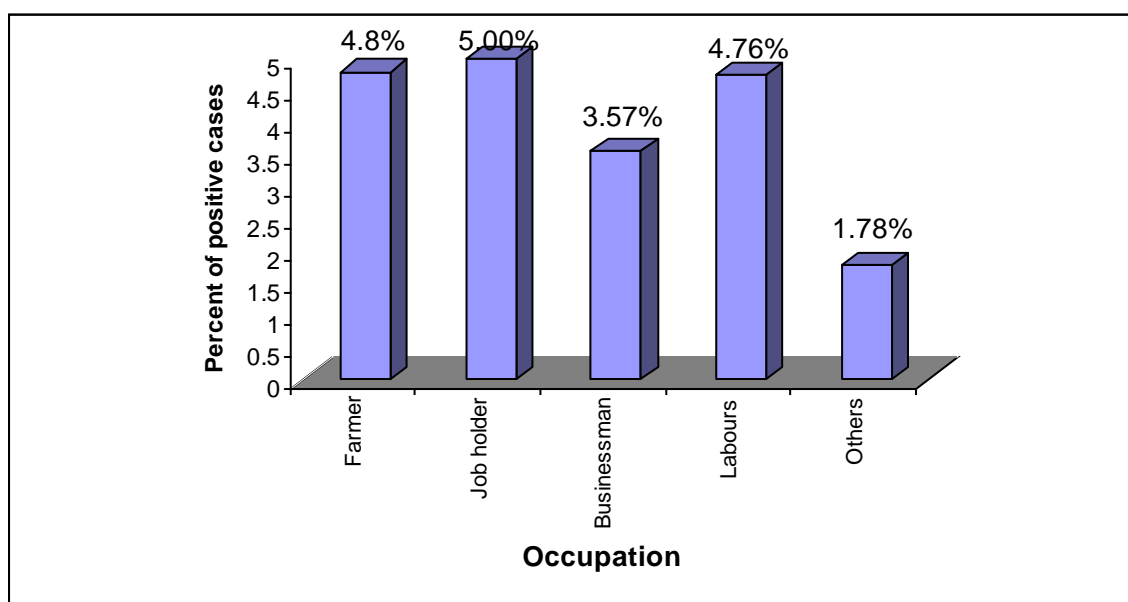
## J Prevalence of malaria on the basis of occupation

During the study, various occupational group individuals were included. Majority of them were farmers and a few of them were jobholders and businessmen. The higher number of malaria infection was observed among farmers 5 (4.8%), job holders 1 (5%), businessmen 1 (3.57%), labours 2 (4.76%) and other 1 (1.78%).

**There was no relation between disease and the occupation ( $t^2=1.0029$ ,  $P>0.05$ ).**

**Table 7: Prevalence of malaria on the basis of occupation**

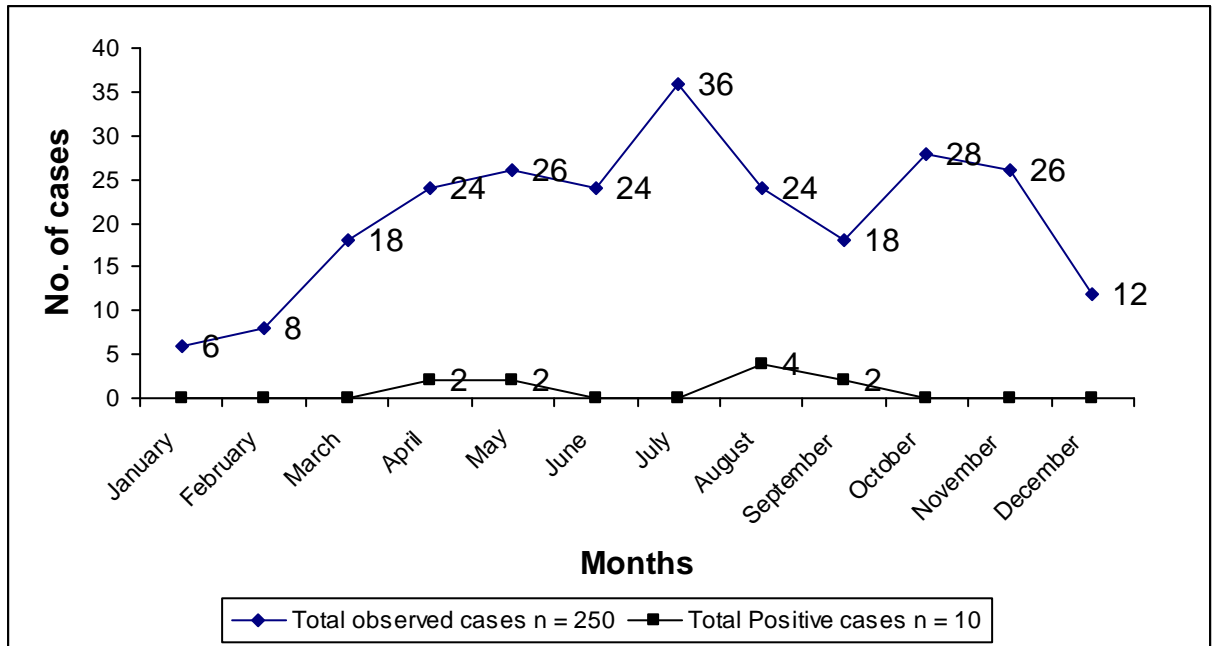
Occupation	No. of total observed cases (n=250)	Malaria	
		Positive cases (n=10)	Percent(%)
Farmer	104	5	4.80
Job holder	20	1	5.00
Businessman	28	1	3.57
Labours	42	2	4.76
Others	56	1	1.78



**Figure 6: Prevalence of malaria on the basis of occupation**

## ) Month wise prevalence of malaria

From the study, the result revealed that the highest malarial positive cases were recorded during four months (summer season of the year). Largest number i.e 36 suspected cases were recorded in July (14.4%). Of the total malarial cases, 40% of cases were recorded in August followed by two cases (20%) prevalence for each month (April, May and September) of summer season.



**Figure 7: Month wise prevalence of malaria**

## ) Malaria infection in relation to environmental aspect

### Drinking water source and malaria infection

From the study it reveals that out of 250 individuals, 242 used hand-pumps for drinking water source and 8 used well water. All the malaria infection was from hand-pump users with 4.13% slide positivity rate.

**It was found that the infection with malaria and water source was insignificantly related ( $t^2=0.3442$ ,  $P>0.05$ ).**

**Table 8: Drinking water source and malaria infection**

Source of water	No. of total observed cases (n=250)	Malaria	
		Positive cases (n=10)	Percent (%)
Hand Pump	242	10	4.13
Well	8	0	0.00

### **Cattle shed and malaria infection**

Among 250 population, 226 (90.4%) were rearing cattle and out of these 176 had cattle shed nearby their houses at a distance of less than 20m. Thirty (30) people had cattle shed little farther from their houses at a distance of greater than 20m. The malaria infection was found high in families where cattle shed was at less than 20m from their houses i.e. 4.54% and 2 were from the houses where the cattle shed was at a distance greater than 20m i.e. 6.66%.

**The malaria infection and the distance of cattle shed was found to be insignificantly related ( $t^2=2.52$ ,  $P>0.05$ ).**

**Table 9: Distance of cattle shed from house and malaria infection**

Distance of cattle shed from the house	No. of total observed cases (n=250)	Malaria	
		Positive cases (n=10)	Percent (%)
Less than 20m	176	8	4.54
Greater than 20m	30	2	6.66
No	44	0	0.00

### **) Prevalence of malaria in relation to the use of different preventive measures adopted by respondent**

In relation to different preventive measures majority of households (170) used bed net, followed by maintenance of hygiene, by covering the breeding places of mosquitoes and by spraying insecticides (50), and use of mosquito repellent (10). Twenty (20) individuals did not use any preventive measures. The highest percent of positive cases were from the people using no preventive measures i.e. 15% and the lowest percent from those using mosquito repellent i.e. 0.00%.



**Table 10: Prevalence of malaria in relation to the use of different preventive measures adopted by respondent**

Preventive measures	No. of total observed cases (n=250)	Malaria	
		Positive cases n=10	Percent(%)
Bed-net	170	5	2.94
Hygiene	50	2	4.00
Mosquito repellent	10	0	0.00
No measures	20	3	15.00

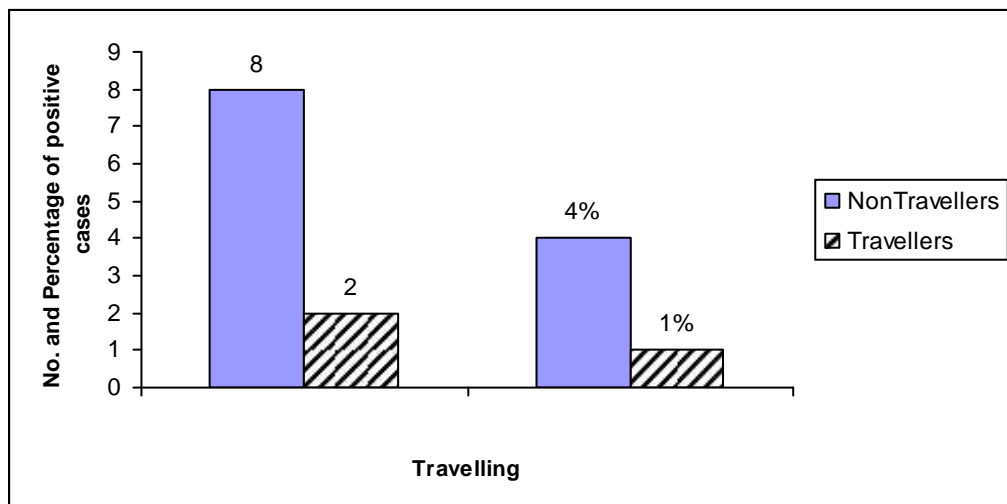
### ) Malaria infection and travelling

From the study the result revealed that, out of 10 malarial patients, 8 (80%) of the total infected people were permanently inhabiting the area whereas 2 (20%) were found migrated from Assam, India, where their relatives were.

**The presence of malaria infection is independent of the travelling ( $t^2=0$ ,  $P>0.05$ ).**

**Table 11: Malaria infection and travelling**

Travelling	No. of total observed cases (n=250)	Malaria	
		Positive cases (n=10)	Percent (%)
NonTravellers	200	8	4
Travellers	50	2	1



**Figure 8: Travelling and malaria infection**



Plate 1: Interview with the patient



late 2: Pricking the finger for blood collection



P

Plate 3: Withdrawing the blood

Plate 4: Preparing the blood film for malarial parasite



Plate 5: Making blood smear

Plate 6: Microscopic blood examination



Plate 7: Trophozoite of *Plasmodium* sp.



Plate 8: Gametocyte of *Plasmodium* sp.



## VI

### DISCUSSION AND CONCLUSION

Out of 250 blood-sample collection and examination, 10 were found positive for malaria. This represents 4% slide positivity rate of the total number of samples. Prevalence rate was found to be 0.235 per thousand in Mahendranagar VDC. Higher number of asexual trophozoites was identified as compared to gametocytes. Prevalence rate (0.23 per thousand) of malaria in Mahendranagar VDC is higher than the mean prevalence rate (0.025 per thousand), reported by HMG in overall Sunsari district for last six years (1999 to 2004) (MoH, 2005) and much less than the Sivakoti's estimation (3.94) in Bhutanese Refugee Camp, Jhapa (2003). HMG (2005) in 2003 report also estimated 4% SPR which is similar to the SPR result of this study.

Prevalence of malarial infection in this study was found higher than Nepal Government's estimation. This is because of the blood samples were collected only from the suspected patients, not randomly. Despite this, present study shows less prevalence of malaria in comparison to other previous studies conducted in other similar areas / districts and VDCs of the region.

Whereas, Joshi (2004) reported 66 patients (21.56%) suffered from malaria out of 306 total samples in Kanchanpur district. This also shows higher prevalence than in the study area. Likewise, Sivakoti (2003) determined 5.1 and 4.03 prevalence rate during two years (1999 and 2000) in Bhutanese Refugee Camp, Jhapa which is about 50 km away from the study area and 10 km near to India border.

In the present survey, a total of 10 positive cases were detected out of 250 slides. Of these positive cases, only the *P. vivax* contributed to the malaria in the area. There was no any case by *P. falciparum* in study area, but it was reported by Sivakoti (2003) in Bhutanese Refugee Camp. Ghimire (2002) states that *P. vivax* has the widest distribution, extending throughout the tropics, subtropics and temperate zones. *P. falciparum* is generally confined to the tropics. Sherchand (2002) also mentioned that *P. vivax* is the predominant species in most of the malarious areas of Nepal by a factor of 10:1 ratio between *P. vivax* and *P. falciparum*. *P. vivax* is more common

than *P. falciparum* as a cause of malaria in parts of the tropics outside Africa (Joshi, 2004). Similarly, a study carried out in three wards of Kavre district in 2000 showed that percentage of *P. vivax* was higher (69.2%) than *P. falciparum* (30.7%) (Karkee, 2001). Likewise, no *P.falciparum* cases were reported from Kavre in the years 1996, 1997 and 1998 (Karkee, 2001).

The total number of *P. vivax* cases was higher (78.1%) than *P. falciparum* cases (20.1%) during the last three years period (1999-2001) in Morang district. The mixed cases were reported only in 1999, which accounted to about 3.4% of the total infection. The number of malaria cases reported from 1999 to 2001 was 145, 98 and 44 respectively in that place. (DoHS,2002).

The prevalence rate of *P. falciparum* was higher than *P. vivax* in the Bhutanese Refugee Camp from 1996-1998. The mixed infection during the same period was 0.35%. In contrast, the prevalence of *P. vivax* was higher than *P. falciparum* among the people of Jhapa district from 1996-1998. The mixed infection during the same period was higher (1.17%) than that of refugees. The analysis of the species wise positivity of malaria among the refugees of Sanischare Camp during three years (1998-2000) revealed that the total number of *P. falciparum* was higher than *P. vivax* in 1998 and 1999. But in 2000, total number of *P. vivax* was more than *P. falciparum*. The total number of positive cases was maximum in 1999 (91 positive cases), followed by 2000 (75 positive cases) and was the least in 1998 (61 positive cases). The mixed infection was reported in 1999 which contributed to 2.19%. (Sivakoti, 2003).

In this study, majority of observed cases was in 21-30 years age group (n = 74) followed by 11-20 years (n = 60), 31-40 years (n = 52), > 50 years (n = 36) and 41-50 years (n = 28). Majority of the infected population was from 31-40 years and minimum positive cases from 21-30 years and above 50 years.

Likewise, Sivakoti (2003) reported that the age group 30-40yrs contributed to the maximum number of cases (39.9%) and least infection rate (3.03%) was observed in age group 50-60yrs. The same is in this study too. The maximum number of positive cases contributed by age group 30-40 yrs might be due to their free movement in malaria endemic areas of Assam and Bengal in economic pursuit.

The least infection was found in the age group 21-30 and above 50 years as very few people belonging to this age group move out of the village either to meet their relatives in India or to earn money in order to fulfill their basic needs. The people above 50 years of age might visit the local places within Nepal but definitely they do not travel several malaria endemic places of India and thus have no role in importing malaria to the VDC. Moreover, the people of this age group might keep themselves away from the mosquito bite by using mosquito net regularly, and wearing proper clothes ensuring that the body is properly covered.

The sexwise analysis of malaria positive cases during the study period showed that 80% of the cases have occurred in males and 20% of the cases have occurred in females. This result is moreover similar to the result of Sivakoti (2003) in Bhutanese Refugee Camp where he reported 85% in males. This can be explained due to their (male) mobility to malarial areas in economic pursuit and practice of norms and social behavior.

The result indicated that the malaria infection was more in poor people and hut houses. This is due to the reason that the general people coming to clinic and health post were mostly of low income group and they do not pay much attention towards their surrounding and environment.

Similarly, malaria infection was found only among Hindu people (4.65%). It can be explained on the basis that most of the population inhabiting Mahendranagar VDC follow Hinduism.

From the survey done among the people of Mahendranagar VDC the result revealed that, awareness of malaria was the highest in literate. It was found that maximum number of positive cases were from illiterate and under SLC level. The awareness towards the malaria prevention seems to be directly associated with the literacy rate. Hence, it can be concluded that education plays a key role in building up of positive social attitude towards common infections, mode of disease transmission in society and its adverse impacts in society due to malaria.

The result reveals that the majority of malaria infection was in farmer 5 cases and labourer 2 cases which can be explained on the basis that the large number of

population is dependent on agriculture and with low socio-economic status.

Regarding month-wise distribution of malaria, blood slide examination showed a peak in July (36) and the least in January (6) and February (8) for the VDC as a whole. The highest percentage of blood slide examination in July was firstly due to heavy monsoon of July which fills the drain, ditches, ponds, puddles with water and the stagnant water in these places which provides suitable breeding ground for mosquitoes. HMG's 2005 report also showed the peak distribution of malaria in June, followed by July and August.

The seasonwise distribution of malaria positive cases showed a peak in Summer season (40%) followed by autumn season (22.7%). The distribution of malaria positive cases in spring and winter were 19.66% and 17.6% respectively. The maximum number of positive cases during summer season might be due to prevalence of favorable conditions in this season for the mosquitoes to survive and breed.

The use of bed-nets in malaria endemic areas is one of the basic preventive tools as described by WHO recommendation. In this survey which was carried out in all the wards of Mahendranagar VDC in Sunsari district revealed that 170 out of 250 people were found to be using bed-net regularly but also 5 cases out of 10 positive cases was found in it. The reason was that the marshy land and paddy field provides suitable breeding places for the mosquitoes. There were 20 people who did not use any preventive measures and accounts for 15% of malarial infection.

Despite the internal migration, migration was not found considerable in the area. This is because out of 10 malarial patients, 8(80%) of the total infected people were permanently inhabiting the area where as 2(20%) were found migrated from Assam, India, where their relatives were. In this sense, travelling abroad, mainly in tropical belt of India, is responsible in disseminating the malaria.

The comparison of malaria case between Sanischare Camp (Sivakoti, 2003), and Morang and Saptari districts (HMG/MoH, 2005) was done due to similar geographical and ecological condition. So, the secondary data of three years (1999-2001) was collected from among the general population of the Morang district and



analyzed. The prevalence rate decreased with subsequent years. It has been reduced from 0.17 in 1999 to 0.05 in 2001. The positive slides have been decreased from 145 in 1999 to 44 in 2001. The blood slide collection and annual blood slide examination were the highest in 2000. The slide positivity rate has almost decreased three and half times from 1999 to 2001. The people of Morang district have realized the necessity to give blood for diagnosis and some of them even knew parasites are seen in the blood if they have malaria.

The low prevalence rate of malaria among general population in Mahendranagar VDC might be due to i) inadequate reporting of the malaria, ii) increased awareness towards malaria and iii) greater distance from India border. Some people inhabiting the VDC visit different malaria endemic places of Assam and Bengal States of India, either to meet their relatives or to look after their cultivation. They acquire the fever and return to the village to seek treatment. This strongly provides evidence that the imported malaria cases do prevail in the VDC. On the other hand, HMG's estimation showed the prevalence of imported malaria cases among people of Morang district might be low. This may be so because the local people do not visit the Assam and Bengal, states of India, as much as the refugees visited and import the malaria. Moreover, the low prevalence rate of malaria in local population of Jhapa than in population of refugees (estimated by Sivakoti, 2003) might be due to better housing condition and environmental sanitation of the general population in the study area than that of other comparable areas.

## VII

### RECOMMENDATIONS

The findings based on the present study showed that the malaria is major public health problem in tropical region including Mahendranagar VDC of Sunsari district where prevalence of malaria was found to be 4%. So there needs some measures to be undertaken in order to eradicate the malaria. The following recommendations have been made on the basis of the results for effective control of malaria in the area.

1. People awareness programmes should be conducted regularly in relation to prevention and control of mosquito and mosquito borne diseases.
2. Insecticide impregnated bed-nets should be distributed to the local poor people.
3. Malaria screening laboratories should be established in border areas.
4. Administration of prophylatic drugs should be compulsory for every individual before they visit to any endemic areas (eg. Assam and Bengal states of India).
5. Sub Health Post and other clinic staffs must be encouraged with many facilities so that imported cases could be detected and treated actively.
6. Orientation training should be carried out in accomplishing the continuation of presenting the Health Post / Sub Health Post / VDC wise strata and receptivity report to Ministry of Health and Population/Epidemiology and Disease Division which has been discontinued in the recent years either due to ignorance or unavailability of data.
7. Health staffs should be motivated to record details about patients on the appropriate forms, to take blood before giving anti-malarial drugs and to give feedback to the slides taken.
8. Environmental sanitation should be properly managed. For example, the stagnant water should be drained out.
9. Effective insecticide spraying should be continued specially during summer season.

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# Annex – 1

## QUESTIONNAIRE

Sheet No..... House No. (If any)

Date:..../..../200  
D / M/Year

### Demographic Information:

Full Name : Age: Sex: M/F

Religion: Marital Status: Ward no.:  
Occupation: Farmer ( ) Job Holder ( ) Labour ( ) Businessman ( ) Others ( )  
Educational Status: Illiterate ( ) Literate ( ) School ( ) University ( )  
Family Size: Total: ( ) Male ( ) Female ( )

### Economic Status:

Income Generating family members: Fixed ( ) Not Fixed ( )  
Average Income of Family per year (In NRs.): <50,000 ( ), 50,000-75000 ( ),  
75,000-100000 ( ), >1,00,000 ( )

### Environmental Factors:

Type of House: Cemented ( ) Wooden ( ) Hut ( )  
Water Resources: Tap Water ( ) Pond ( ) Well ( ) Hand Pump ( ) Others ( )  
Do you have any domestic animal? Yes ( ) No ( )

If, yes, Types and number of animals:

Type of domestic animal	Number	Remarks

Where do you keep these animals? Nearby house ( )  
Far from house ( ) Sharing the same room ( )  
Condition around households: Pond ( ) Stagnant water ( ) Farm field ( )  
Bushes ( ) Others ( )  
Do you use mosquito net? Yes ( ) No ( )  
Have you gone out of your village recently? Yes ( ) No ( )  
Did any member of your family have malaria attack recently? Yes ( ) No ( )  
When ( ) Where ( )

### Knowledge about the disease

Do you know about the disease malaria? Yes ( ) No ( )  
If yes, what are the clinical histories? High fever ( ) Chilling ( )  
Splenomegaly ( ) Others ( )  
How does this disease transfer? Vector ( ), Contact ( ), Sex ( ), Air ( ), Water ( )  
Do you know about female Anopheles mosquito? Yes ( ), No ( )  
When does mosquito bite? Morning ( ), Evening ( ), Daytime ( ), Night ( )



Do you think the malaria transmission can be prevented? Yes ( ) No ( )  
 What are the preventive measures of malaria? Spraying insecticides ( ), medicine ( )  
 keeping surrounding clean ( ) using bed-net ( ), unknown ( )  
 What do you suppose to do if you get the disease? Consult doctor ( ), Ayurvedic ( )  
 ), Homeopathic ( ), Dhama ( ),  
 Nothing ( )  
 Have you ever heard/seen people died from malaria? Yes ( ), No ( )

**Attitude and Practices towards Malaria**

Have you ever attacked by malaria? Yes ( ), No ( )  
 Has other member of your family are attacked by malaria? Yes ( ), No ( )  
 Where did they get treatment? Health Post ( ), Clinic ( ), Hospital ( ), Others ( )  
 Is that patient completely treated? Yes ( ), No ( ),  
 If no what happened?  
 Where do you sleep? Outdoor ( ), Indoor ( ), Both ( )  
 Do you use any repellent of malaria? Yes ( ) No ( )  
 If yes then what? Cream lotion ( ), bed-net ( ), electrical vaporizer ( )  
 spraying/mosquito coil ( ), traditional method ( )  
 Had any body/organization launched for insecticide spraying? Yes ( ), No ( )  
 If yes then by whom when / which insecticide had been used? DDT ( ), BHC ( )  
 Malathion ( ), Others ( ) Don't know ( )  
 What is the effect of insecticide? Malaria decreased ( ), Increased ( )

Suggestion for the prevention of malaria: .....  
 ...  
 .....  
 .....  
 .....

Blood samlpe slide number: Code no. if any:

*Please don't fill below during questionnarie:*

Questionnarie Sheet No:	Microscopic examination result
Slide No:	Positive ( ) Negative ( )
Remarks / Pathogen:	
Result / Comment:	

Supervisor's comment / suggestion: .....  
 .....  
 ...

## ANNEX - 2

### Signs and symptoms (Clinical features)

**Signs:** Physical examination usually demonstrates an increased temperature, tachycardia, and warm flushed skin. The spleen is often palpable in initial infection, but this is more likely in subsequent attacks. It is usually soft and may be tender. The liver is often enlarged and may be tender; jaundice is not unusual. Orthostatic hypotension often occurs during initial infections. Mental confusion and cyanosis are sometimes encountered.

**Symptoms:** Patients present with a variety of symptoms depending on the stage of infection and the infecting species. Fever is virtually always present, and fever plus any other symptom might be malaria if exposure occurs. Common complaints include mild to moderate malaise, fatigue, muscle aches, back pain, headache, dizziness, loss of appetite, nausea, vomiting, abdominal pain, and diarrhoea. Dry cough and shortness of breath have been reported in some patients. Gastrointestinal complaints can be considerable, suggesting a diagnosis of gastroenteritis. Young children and semi-immune individuals may complain of fever and headache as their only symptoms.

### Malaria Clinical Findings

Signs and Symptoms	Percent with Finding
Fever and Chills	96
Headache	79
Muscle Pain	60
Palpable Liver	33
Palpable Spleen	28
Nausea and Vomiting	23
Abdominal Cramps/Diarrhoea	6

Source-Anonymous, [www.pubmed.com](http://www.pubmed.com)

## Laboratory Findings

Abnormal laboratory findings reflect the severity of hemolysis.

**Blood:** A normocytic, normochromic anemia with leukopenia and thrombocytopenia is sometimes present on initial screening, but is almost always present following medication therapy with the resultant clearing of parasitemia. Massive *P. falciparum* infections causes acute decrease in hemoglobin, hematocrit, and an increase in reticulocyte count.

**Kidneys:** Trace to moderate protein, urobilinogen, and conjugated bilirubin may be found on urinalysis. In severe *P. falciparum* infections, massive hemolysis combined with circulating immune complexes produces acute renal insufficiency or failure ("blackwater fever") with laboratory findings of hemoglobinuria, proteinuria, and an elevated serum creatinine.

## Malaria Laboratory Findings

Finding	Normal Range	Percent with Abnormal Findings
Reticulocytosis	3 – 18%	42
Thrombocytopenia	12K - 150K	36
Bilirubin Increased	1 – 1.8	33
VDRL Positive	(-)	28 (+)
Anemia	5.8 - 12 (Hgb)	28
Leukopenia	3,000-4,700	26
Alk. Phos. Increased	11-27	17
SGOT Increased	40-108	10

Source-Anonymous, [www.pubmed.com](http://www.pubmed.com)

## Pathophysiology

Clinical symptoms and signs of malaria occur shortly before or at the time of red blood cells lyses. Fever is caused by the release of merozoites, malarial pigment, parasite proteins and cellular debris. Chills or rigor, followed by high fever occur in a cyclical pattern in infections due to *P. vivax*, *P. ovale*, and *P. malariae*, but not *P. falciparum*, which is more likely to show continuous fever with intermittent temperature spikes. The malaria paroxysm is the defining clinical feature of the disease. That being said, it is often not present. Fever caused by malaria can have any pattern, and *P. falciparum* infections often present with a constant

fever. The classic paroxysm typically has three stages; and is preceded in some patients by an initial period of nonspecific symptoms. Those symptoms include fatigue, muscle aches, loss of appetite, headache, and a slight fever of 2-3 day's duration. A paroxysm begins with the "cold" or "shilling" stage lasting 15 minutes to several hours during which the patient feels cold and has shaking chills. The second "hot" stage lasts several hours and coincides higher. There is minimal sweating and the patient is at risk of febrile seizures or hyperthermia, brain damage. Clinical signs and symptoms include tachycardia, hypotension, cough, headache, backache, nausea, abdominal pain, vomiting, diarrhoea and altered consciousness. Within 2-6 hours, the patient enters the third "sweating" stage of the paroxysm with generalized sweating, resolution of fever, and marked exhaustion, usually giving way to sleep. Paroxysms occur in regular intervals, but take several days to emerge. As previously stated, the classic paroxysm described above is generally not how *P. falciparum* infections present. *P. falciparum* malaria is more severe and qualitatively different from the other *Plasmodia* that infect human and is the only type that causes micro vascular disease.