

## EPIDEMIOLOGICAL PATTERNS OF MALARIA INFECTION IN DISTRICT MARDAN, PAKISTAN: A HOSPITAL-BASED STUDY

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### Abstract

Malaria remains one of the most significant vector-borne infectious diseases worldwide and continues to pose a major public health challenge, particularly in tropical and subtropical regions. The disease is caused by Plasmodium parasites and is transmitted to humans through the bite of infected female Anopheles mosquitoes. Despite considerable progress in malaria control programs, the disease remains endemic in many developing countries, including Pakistan, where it contributes substantially to morbidity and places a burden on healthcare services. The present study was conducted to determine the prevalence of malaria among patients visiting the District Hospital Mardan, Khyber Pakhtunkhwa, Pakistan. A total of 420 patient records were collected and analysed to assess the occurrence of malaria infection. The findings revealed that 113 patients (26.9%) were diagnosed with malaria, indicating a considerable disease burden in the study population. The observed prevalence highlights the continued transmission of malaria in the region and emphasizes the need for effective surveillance and control measures. The study underscores the importance of preventive strategies to reduce malaria transmission. These include the use of insecticide-treated mosquito nets, application of mosquito repellents, environmental management to reduce mosquito breeding sites, and increased public awareness regarding malaria prevention. Early diagnosis and prompt treatment remain essential for reducing disease complications and transmission. Antimalarial drugs such as chloroquine and primaquine continue to play an important role in the management of susceptible malaria cases. In conclusion, malaria remains a significant public health concern in District Mardan. Strengthening preventive measures, improving community awareness, and ensuring timely diagnosis and treatment are critical steps towards reducing the prevalence and impact of malaria in the region.

### INTRODUCTION

Parasites are living organisms that live in or on the bodies of other living organisms, taking advantage of them and inflicting harmful effects. Parasites are cosmopolitan, and concerning 70% of them are microscopic, like malaria infection parasites, whereas the remaining can be seen with the eye (Elbishti, et al. 2025). Malaria could be a critical

condition caused by female genus Anopheles mosquitoes (parasites) of the protists genus Plasmodium via a bite that injects into the organism from its spit into the human's cardiovascular system (Solomon, et al. 2016). The disease is sometimes transmitted to individuals through the bites of infected mosquitoes, and

these parasites specifically target the red blood corpuscle (WHO, 2019).

In tropical and subtropical countries, Malaria is known as the main cause of illness in human beings (WHO, 2019). Four different protozoan species are responsible for malaria: *Plasmodium falciparum*, *Plasmodium vivax*, *Plasmodium ovale*, and *Plasmodium malariae*. *Plasmodium vivax* is responsible for the major cause of this infection (Jabeen, *et al.* 2014). The transmission greatly varies from region to region, village to village, and even from patient to patient within a village. The term malarial infection begins from mala aria, which means bad air or marsh fever, because of its association with lowland and marshland. The first evidence was found in mosquitoes, which are approximately 30 million years old. (Kristine Krafts, 2013).

*Plasmodium falciparum* is a unicellular parasite of humans and causes falciparum malaria (formerly called malignant tertian malaria), the most dangerous disease, and is responsible for around 50% malarial cases (Rich, *et al.* 2009). *Plasmodium vivax* is a human parasitic pathogen, and the most common and widely distributed cause of repeated malaria (White NJ 2018). *Plasmodium Ovale* species causes rare ovule malaria (formerly called triennial malaria) with a long gestation period and reverts at three-month intervals (Chessbrough, 2000). *Plasmodium malariae* species causes quartan malaria with fever repeating every 72 hours. It is remarkable in that it can remain in the blood of a patient for years at very low concentrations but does not have a dominant stage in the liver. Sometimes, after half a year of infection, it can relapse (Shankar, *et al.* 2026). Malarial parasites belong to the protozoan genus (phylum Apicomplexa). In humans, malaria is caused by *P. falciparum*, *P. malariae*, *P. ovale*, *P. vivax*, and *P. knowlesi*. *P. falciparum* and *P. vivax* are the most infectious species, causing infection in more than 660,000 individuals among all five known *Plasmodium* species (Van Hellemond, *et al.* 2009). The signs and symptoms of malaria infection typically take 10-28 days to months following infection. Early symptoms may include flu-like symptoms and a blood disorder. Other common

symptoms of malaria infection include high fever, vomiting, nausea, headache, shivering, muscle pain, body fatigue, joint pain, anaemia, haemoglobin in the body waste, retinal harm, and convulsions (Almaw, *et al.* 2024). Analysis of malaria is generally proclaimed by the tiny examination of blood films or by antigen-based, fast, demonstrative test (RDT) (Kattenberg, *et al.* 2011). In the RDT, a few zones have the capacity to recognize whether the malaria fever side effects are caused by the *Plasmodium falciparum* or sometimes caused by different types of parasites (Abba, *et al.* 2014). For the identification of the malaria parasites, the microscopic strategy is used mostly, but industrially available RDTs are frequently more accurate than the films at forecasting the presence of malaria fever parasites (Abbas, Tasheen, and Alla, 2012).

The blood of malarial infected person can be tested with a magnifier, the most effective laboratory technique used for confirmation of malaria. Flavouring treatment is well practiced in Pakistan. It is fascinating that in Pakistan, five hundredth of the population is being cured with flavouring medicines by virtually thousands of ancient flavouring physicians Hakims (Khan, *et al.* 2008; Ahmad, *et al.* 2008; Ruler and Hatcher, 2005). Vaccination for infection would be the best, cost-effective, and safe public health practice to cut back the burden of malaria infection. There are varied reasons for the continuation and re-emergence of malaria infection, for instance, economic reasons, decline in management programs and mosquitoes/ parasites to pesticides medication and environmental changes all contribute or play a crucial role in the development and increase of malaria infection diseases. (Beare, *et al.* 2006).

Malaria infection can be transmitted from an infected person to a healthy person through the donation of blood. Alternative factors for transmission include population-connected risk factors like climatic variability, urban agriculture, proximity to dipteran breeding environments, and urbanization (Badu, *et al.* 2013). Prevention is also dispensed either by interrupting transmission in vector management or by giving the patient prophylactic medication. For treatment,

chloroquine-sensitive malaria is treated with intravenous chloroquine. For drug-resistant forms, quinine dihydrochloride mixed with a dose of tetracycline antibiotics should be used. However, tetracycline in pregnant women and children under one year old is not recommended (Almaw, *et al.* 2024)

Sustaining malaria is due to the resistance of the Plasmodium species. Plasmodium falciparum shows resistance to formal anti-malarial drugs like chloroquine (Flores, *et al.* 2013). Out of four species of plasmodium which causes human malaria in humans, three are reported for their resistance to anti-malarial drugs, which are Plasmodium falciparum, Plasmodium vivax, and Plasmodium malariae. Chloroquine was the most successful anti-malarial drug of the 20<sup>th</sup> century and had a wide use also at that time (Abba, *et al.* 2014).

Malaria remains one of the most significant parasitic diseases worldwide presenting about 250 million clinical reports and about one million deaths occur annually worldwide, particularly in poorer tropical and subtropical regions (Kar, 2010). Malaria continues to pose a major public health challenge due to its high morbidity, mortality, drug resistance, and socio-economic impact. Early diagnosis, vector control strategies, effective treatment, and vaccination are essential for minimizing disease risk. The significance of the current study is to improve understanding of parasite biology, transmission dynamics, and emerging resistance patterns, which are critical for developing sustainable control measures. Further research study is needed to develop novel antimalarial drugs, improved vaccines, rapid and accurate diagnostic tools, molecular surveillance of drug-resistant strains, and integrated vector management approaches. Moreover, practicing public health awareness, proper management of the environment, and community-based prevention programs will be critical measures for achieving long-term malaria control and its elimination.

## MATERIAL AND METHODS

### Study Area

Mardan is located in the Khyber Pakhtunkhwa province of Pakistan. In 1937, Mardan was set up as an autonomous region after the name of its base camp town. It is limited on the North by the Buner local and Malakand secret zone, on the East by the Swabi and Buner regions. On the South by Nowshera region, and on the West by Charsadda region and Malakand insured zone. The aggregate region of the area is 1632 square km. The late spring season is amazingly hot. An increase in temperature can be seen from May to June. Indeed, even July, August, and September recorded temperatures were very high.

### Methodology

#### Sample collection

A total of 420 malaria patients were studied, and the samples were collected using a questionnaire for each patient. These samples were collected in the DHQ hospital, Mardan, in a period of 7 months.

#### Questionnaire

A questionnaire was filled out, including questions like patient name, age, area, gender, education level, socioeconomic status, sleeping patterns, treatment, type of medication, awareness of the disease, use of mosquito net, mesh window doors, and Plasmodium species

#### Materials Needed

Different materials were used in the study, including gloves, lancets, glass slides, Giemsa stain, methanol, and staining rod

#### Procedure

About 5mL of blood was collected from the ring finger of each patient with the help of a sterilized needle and prepared a slide.

#### Slide Preparation.

For preparation, 2 drops of blood were put on a clean glass slide; one drop was circled while the other drop was spread along the slide. Left it for 5 minutes to dry in the dryer. The circled drop was

a thick smear, while the spread drop was a thin smear.

**Thick smear**

The thick blood smear was a drop of blood on a glass slide, which was not fixed with methanol. A thick smear was very useful for detecting the presence of parasites because it examined a larger sample of blood. Often, there were few parasites in the blood at the time the test was done.

**Thin smear**

The thin smear was a drop of blood that was spread across a large area of the slide. After drying, the thin smear was fixed in methanol. It helps to discover what species of malaria is causing the infection.

**Staining**

Rapid fields and Giemsa’s staining were used for screening of parasites. Left it for 30 minutes and then washed and dried. The sensitivity of a thick blood film is 5-10 parasites/uL. The optimal PH of the stain was 7.2, and then observed under a microscope.

**Microscopy**

Put a drop of oil emergent on a stained-glass slide and then put it under a microscope for further study.

**RESULTS**

**Overall Incidence of Malaria**

In the current study, a total of 420 blood samples were processed to evaluate malaria. In these samples, 113 samples were tested positive, predicted 26.9% as the overall prevalence, whereas the remaining 307 samples (73.1%) were negative (Table 1).

Table 1. Overall prevalence and diagnostic distribution of malaria within the study population

Total	Positive	Positive %	Negative	Negative%
420	113	26.90%	307	73.0%

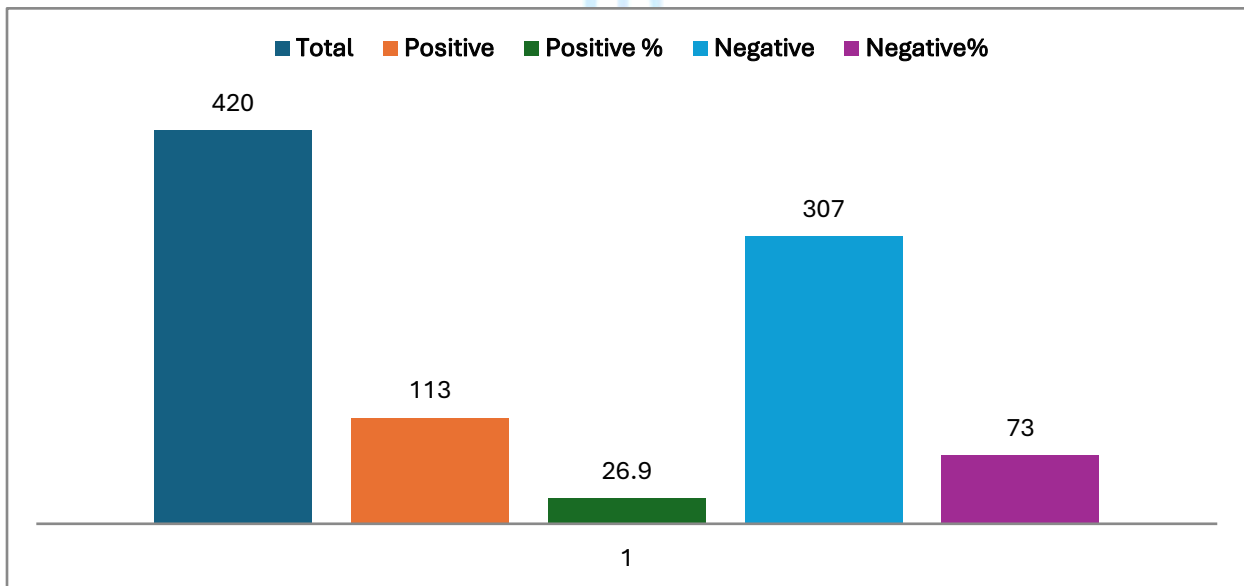


Figure 1. Frequency and percentage distribution of positive and negative test results among the analyzed samples (n = 420). Positive cases: 113 (26.9%); Negative cases: 307 (73.1%).

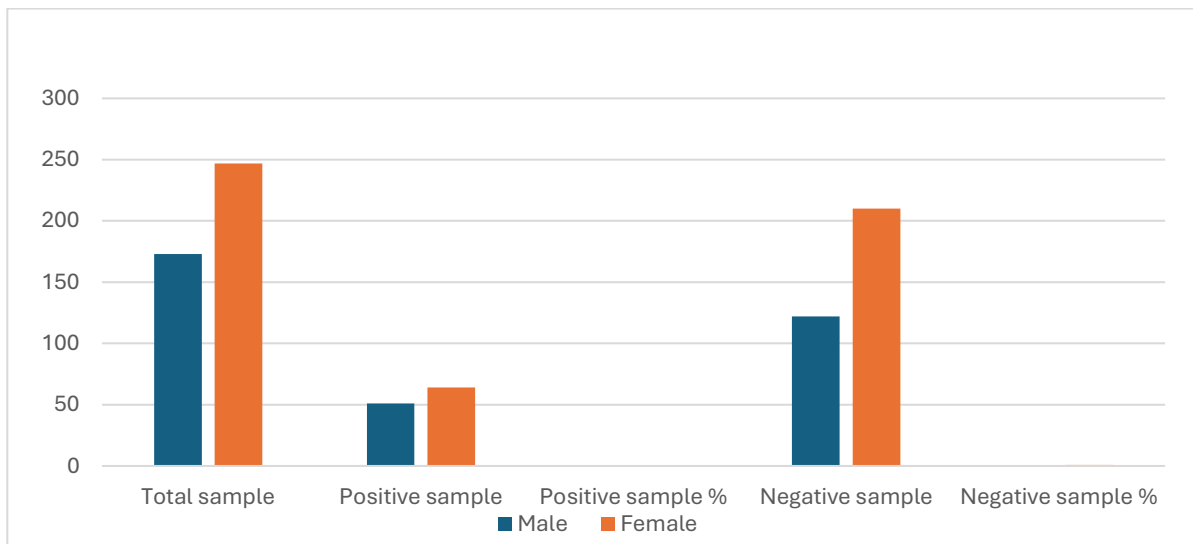
**Gender-wise incidence of Malaria**

In a total of 420 cases, collected from the DHQ hospital, 173 were males, and 247 were females. Malaria positive cases were observed in 51 males

(12.14%) and 64 females (15.2%). The prevalence was recorded as higher among females than males (Table 2).

**Table 2. Distribution of malaria diagnostic outcomes stratified by participant gender.**

Gender	Total sample	Positive sample	Positive sample %	Negative sample	Negative sample %
Male	173	51	12.14%	122	29.04%
Female	247	64	15.20%	183	43.57%



**Figure 2: Gender-based sample distribution and percentage of total study population**

**Age-wise incidence of malaria**

Among all participants, the highest malaria prevalence was recorded in children aged 1-10 with 37 positive cases (8.80%), followed by

individuals aged 11-20 years with 29 positive cases (6.90%). In our study, the lowest prevalence was observed in the age group 41-50 years, where only 7 cases (1.60%) were positive (Table 3).

**Table 3. Distribution of malaria diagnostic outcomes across different participant age groups**

Age Groups	Total sample	Positive sample	Positive Sample %	Negative sample	Positive Sample %
1-10 year	74	37	8.80%	37	8.80%
11-20 Year	64	29	6.90%	35	8.33%
21-30 year	82	21	5%	61	14.52%
31-40 year	76	16	3.80%	60	14.52%
41-50 year	69	7	1.60%	62	14.76%
51-60 year	55	11	2.60%	44	10.47%

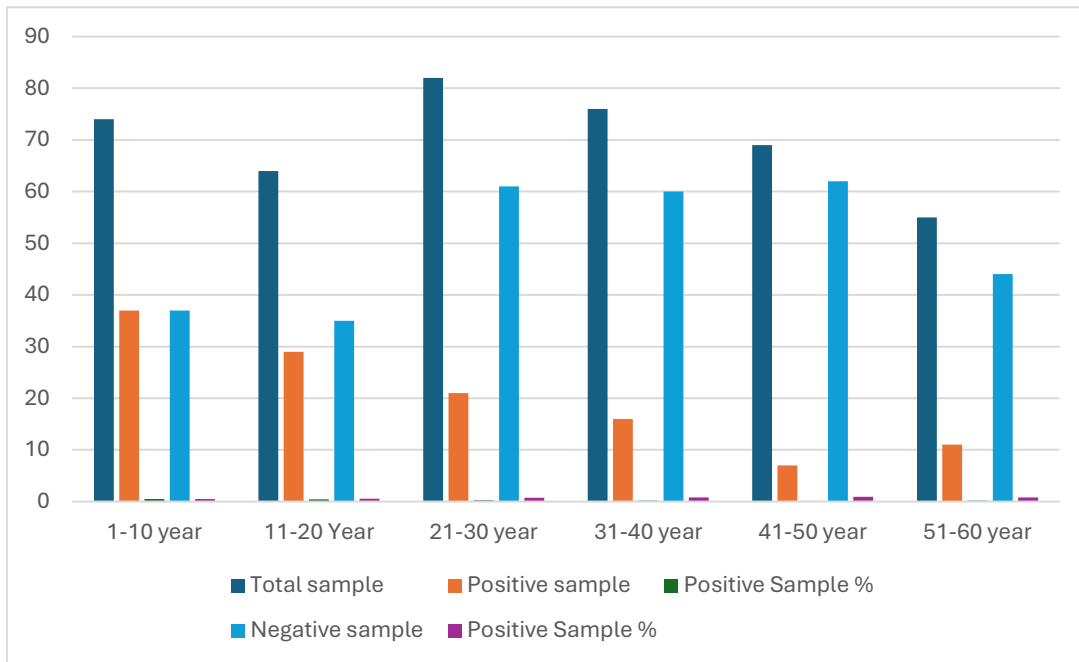


Figure 3: Age-wise sample distribution and percentage of total study population

**Area-wise Incidence of Malaria**

In the current study, a total of 158 individuals were investigated from rural areas, where 52 (12.3%) were malaria-positive, while 72 (17.14%)

positive cases were recorded among 262 urban individuals. Our study represented a higher prevalence in the urban population as compared to the rural population (Table 4).

Table 4. Geographical distribution and area-wise prevalence of malaria within the study population

Area	Total samples	Positive samples	Positive samples %	Negative samples	Negative samples%
Rural	158	52	12.3%	106	25.23%
Urban	262	72	17.14%	190	45.23%

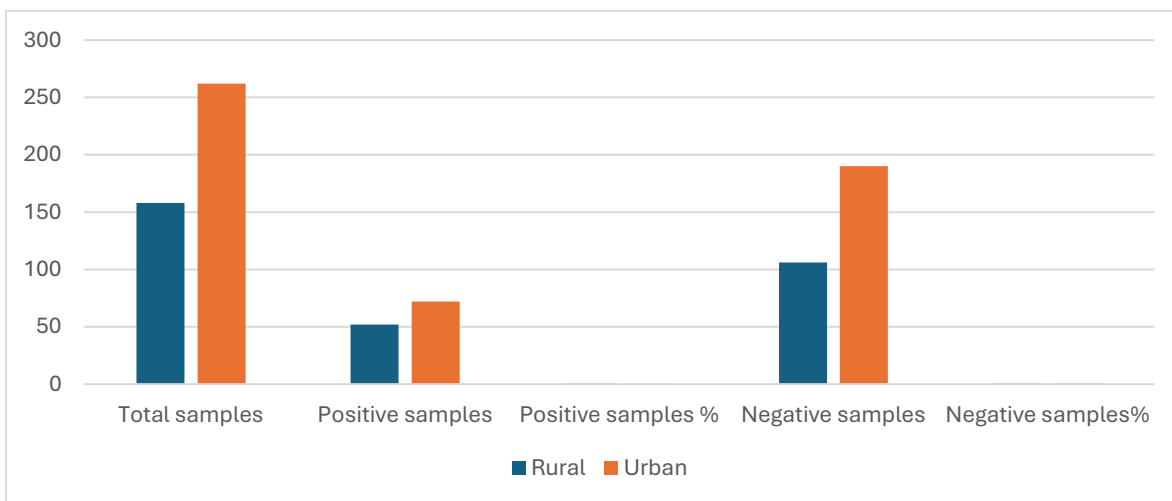


Figure 4: Area-Wise sample distribution and percentage of total study population

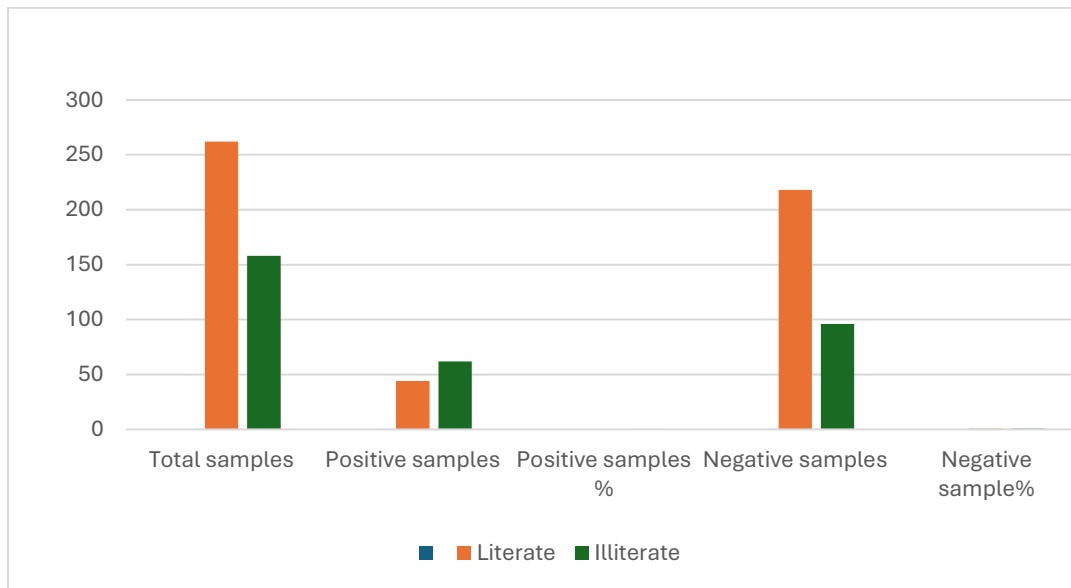
**Education-wise Incidence of Malaria**

On the basis of education level, malaria prevalence was observed to be higher among illiterate

individuals, where 62 cases (14.76%) were tested positive as compared to literate individuals, where 44 cases (10.47%) were malaria positive (Table 5).

**Table 5. Distribution of malaria diagnostic outcomes stratified by participant educational level.**

Education level	Total samples	Positive samples	Positive samples %	Negative samples	Negative sample%
Literate	262	44	10.47%	218	51.90%
Illiterate	158	62	14.76%	96	22.85%



**Figure 5: Distribution of malaria positive cases based on participant education level**

**Economic Status and Malaria Incidence**

According to the economic status, the highest prevalence of malaria was recorded among the population of the lower socioeconomic level,

presenting 51 positive cases (12.14%), followed by the middle level (5.0%) and upper level (3.80%) groups (Table 6).

**Table 6. Prevalence and distribution of positive and negative malaria cases across different economic classes.**

Economic status	Total sample	Positive sample	Positive sample%	Negative sample	Negative sample %
Upper class	68	16	3.80%	52	12.38%
Middle class	88	21	5%	67	15.97%
Lower class	264	51	12.14%	213	50.71%

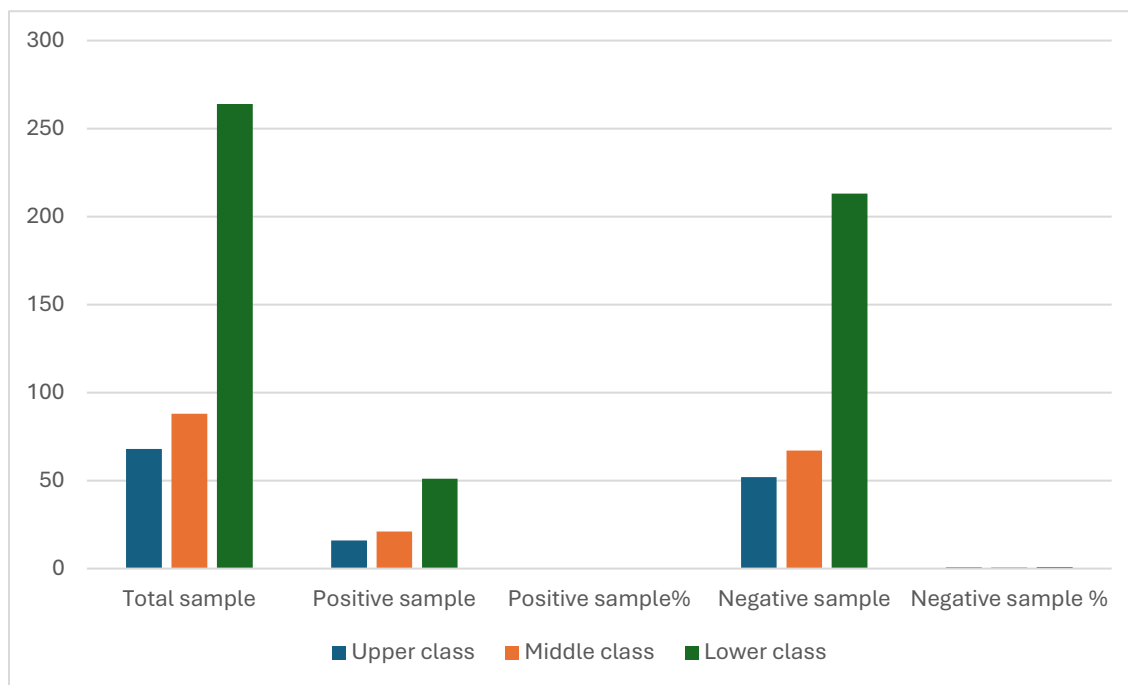


Figure 6: Malaria prevalence and sample distribution stratified by economic status

**Awareness about Malaria and Disease Incidence**

In a total of 420 studied individuals, the aware participants regarding malaria showed a substantially lower prevalence (3.09%) as

compared to those who were unaware of the disease (23.80%). The current findings suggest a strong association between awareness and reduced malaria occurrence (Table 7).

Table 7. Distribution of malaria positive and negative cases by participant awareness level.

Awareness	Total samples	Positive samples	Positive samples%	Negative samples	Negative samples%
Awareness	200	13	3.09%	187	44.52%
Unawareness	120	100	23.80%	20	4.76%

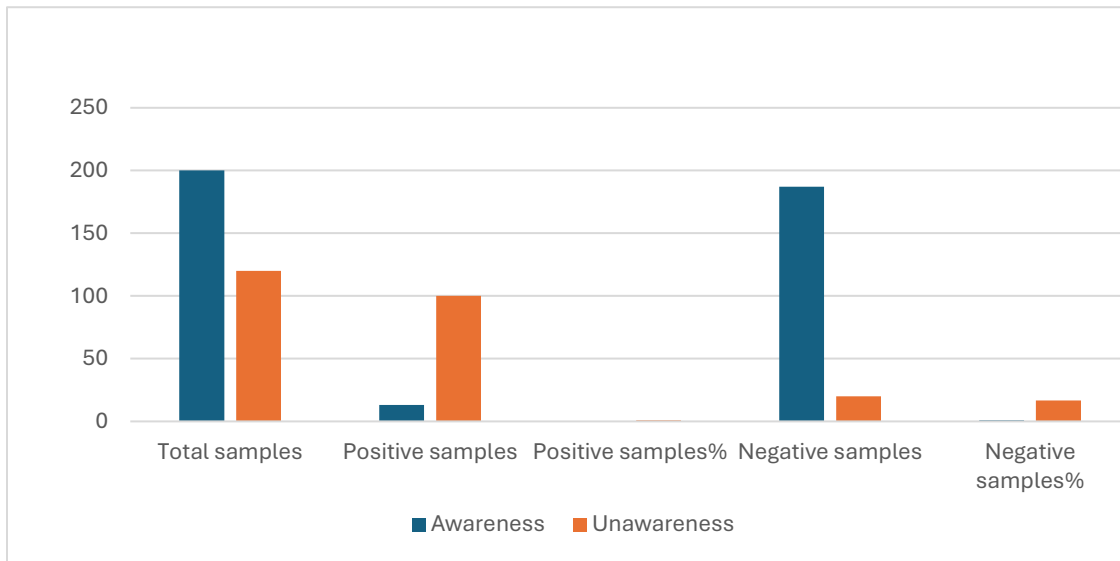


Figure 7: Malaria prevalence and sample distribution based on participant disease awareness

Taken together, these epidemiological parameters underscore that malaria risk in the study cohort is profoundly shaped by underlying social, economic, and educational disparities, which collectively act as primary drivers of vector exposure and transmission persistence.

### DISCUSSION

Malaria is the disease caused by an intracellular parasite of a female anopheles mosquito known as Plasmodium. (Almaw, *et al.* 2024). In the present study, the overall incidence of Malaria recorded 26.90%, while Rehman, *et al.* (2017) recorded 13.99% prevalence of malaria in Shangla, a region of Khyber Pakhtunkhwa (KP). However, Yasinzai and Khan, (2008) reported 34.2% prevalence of malaria in central Balochistan. The lower prevalence of plasmodium species recorded during the present study may be due to the difference in resistance. Similarly, Lathamani and Kotigadd, (2013) reported 43.42% incidence of malaria in India. The reason may be due to the different and climatic conditions.

The current study showed that malaria affects both the male and female where male having 12.14% while female having 15.2% as depicted in the Rehman, *et al.* (2017) where the male was affected 65.24% and the female were affected 34.76%. The difference may be due to the lack of health

facilities, poor sanitation and marshy areas may also be considered as the contributing factors for high prevalence rate. However, Qader, *et al.* (2016) showed 80% of prevalence in male and 68% in female from FATA region in KP.

In current study, the distributions according to age from 1-10 is 8.80%, 11-20 is 6.90%, 21-30 is 5% and 31-40 is 3.80% while Iqbal Yasinzai and Khan, (2008) who's data based on age 1-10 is 16.1%, 11-20 is 23.6% and 21- and above is 21% in central Balochistan. The reason for low overall prevalence rate in the present study might be due to the data collection, carried out during dry weather conditions. On the other hand, Qader *et al.* (2016) recorded prevalence rate under the age 10-20 is 60%, 20-40 is 74% and 40-60 is 81% where the difference might be due to the lack of education and health facilities. However, Rahman, *et al.* (2012) who present incidence in 5-9 years have 5.52%, 10-14 have 3.37%, 15-19 years have 2.2%. that is because most of the people are uneducated, very little awareness about use of mosquito net and mesh widow/doors. In present study, it was reported that the incidence of malaria patients was 12.3% from rural and 17.14% from urban areas as compared to Rahman, *et al.*, (2012) whose showed that the frequency of malaria infection in Banu was 4.56% in rural while 2.38% in urban areas.

Malaria increasingly poses a major public health challenges due to its high morbidity, mortality, drug resistance, and socio-economic impact. Early diagnosis, vector control strategies, effective treatment, and vaccination are essential for minimizing disease risk. The current study signifies to improve the parasite biology, transmission dynamics, and emerging resistance patterns, which are critical for developing sustainable control measures. Further research study is needed to develop novel antimalarial drugs, improved vaccines, rapid and accurate diagnostic tools, molecular surveillance of drug-resistant strains, and integrated vector management approaches. Additionally, following public health awareness, proper management of the environment, and community-based prevention programs will be critical measures for achieving long-term malaria control and its elimination.

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