

## Prevalence of Malaria and Comparison of Peripheral Blood Smear & Rapid Diagnostic Test for Malaria Diagnosis In A Tertiary Care Hospital In Dehradun: Current Scenario

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Date of Submission: 14-01-2020

Date of Acceptance: 30-01-2020

### I. Introduction

Malaria is a mosquito borne infectious disease of humans caused by parasitic protozoan belonging to genus Plasmodium, commonly by *P.falciparum*, *P.vivax*, *P.ovale*, *P.malariae*, *P.knowlesi*. *P. falciparum* & *P.vivax* accounts for most of the cases in India<sup>1</sup>.

Malaria is a serious public health problem in many developing countries. In 2017, an estimated 219 million cases of malaria occurred worldwide with 4,35,000 death occurred from malaria globally. Most of the cases from sub-Saharan Africa and India carried almost 80% of the global malaria burden. The incidence of malaria declined globally between 2010 and 2017 from 72 to 59 cases per 1000 population at risk<sup>2</sup>.

Malaria is a public health problem in India. Majority of the cases are reported from eastern and central part of the country like Odisha, Chhattisgarh, Jharkhand, Madhya Pradesh, Maharashtra, and north east states like Tripura, Meghalaya & Mizoram. In India malaria cases have consistently declined from 2.08 million in 2001 to about 4 lakhs in 2018<sup>3</sup>.

Incidence of malaria occurs throughout the year but are often more common during rainy seasons because of increase in number of breeding sites, relative humidity of 60% and temperature between 20°C and 30°C which favors the spread of malaria<sup>4</sup>.

For efficient treatment and management of malaria, rapid and accurate diagnostic testing is required. In the laboratory, malaria is diagnosed using different techniques, e.g. conventional microscopic diagnosis by staining thin and thick peripheral blood smears (PBS)<sup>5</sup>. Other techniques available are Quantitative Buffy Coat (QBC) method, Fluorescent microscopy, rapid diagnostic tests (RDT) & Molecular methods<sup>6</sup>.

Microscopy is regarded as the 'gold standard' for malaria diagnosis (WHO 1999). However, there are certain limitations of microscopy & to overcome the limitations of microscopy rapid diagnostic test for malaria is used which are fast and easy to perform<sup>7</sup>.

Fluorescent microscopy and nucleic-acid techniques both require skills and equipment which are not universally available<sup>8</sup>.

This study was conducted to ascertain the prevalence of malaria disease burden in patients attending our tertiary care hospital. The objective being the comparative analysis between Peripheral Blood Smear Microscopy (PBS) and Rapid Diagnostic Test (RDT) for the diagnosis of malaria parasite.

### II. Material & method

This was a prospective cross sectional hospital based study performed at a tertiary care hospital over a period of one year from October 2018 to September 2019.

**Inclusion criteria:** Patients of all age group having clinical features suspected of malaria were included in the study.

**Exclusion criteria:** Afebrile healthy individuals were excluded from the study.

**Sample size:** A total 7940 samples were collected from individuals attending Out Patient Department as well as those admitted in our tertiary care hospital.

The ethical clearance was sought from the ethical clearance committee of our institute prior to conducting this study. The patient's demographic and clinical data was recorded in a specifically designed case recording form.

As per the study protocol designed, all the positive samples were tested by both PBS as well as RDT to detect for malaria parasite for the diagnosis of Malaria.

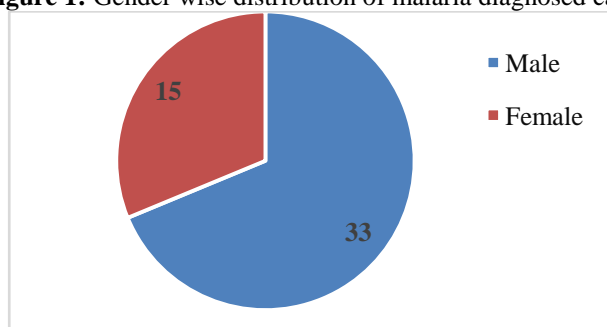
Three ml of whole blood sample was collected in EDTA Vials (BD Vacutainer K2 EDTA 5.4mg) from all the suspected cases. The samples were tested for malarial parasite within 6 hours of the sample collection either by microscopy of PBS by making thick and thin smear stained by Leishman’s stain and/or by RDT using Fast Vue Malaria Pan Kit Rapid Malaria Ag Detection Test for detecting pLDH malaria antigen which is decided by the treating clinician. Sample which came out to be positive by either of the test PBS/RDT were made aliquot and stored at 2- 8°C and were further confirmed by another test either PBS/RDT within 24hours.

**Statistical analysis:** The data obtained was entered in MS excel and it was analyzed using Chi square test for the statistical significance.

### III. Results

Total 7940 samples were tested. Out of the total tests performed, 48/7940 samples came out to be positive for malaria parasite either by PBS &/or RDT. The overall prevalence of malaria in our tertiary care hospital was found to be 0.6%(48/7940). Male preponderance with 69% (33/48) was noted among the diagnosed cases in this study (Fig 1).

**Figure 1:** Gender wise distribution of malaria diagnosed cases



In the present study maximum cases belonged to >15 year of age group 85% (41/48) (Table no1).

**Table no 1:** Age wise distribution of Malaria diagnosed cases

Age group	Number
< 5	0
5-10	2(4.2%)
10-15	3(6.3%)
>15	43(89.5%)
<b>Total</b>	<b>48</b>

When results of PBS and RDT were analysed it was found that their correlation was significant with a p value of <0.05 (Table no 2)

**Table no 2:** Correlation of PBS with RDT results

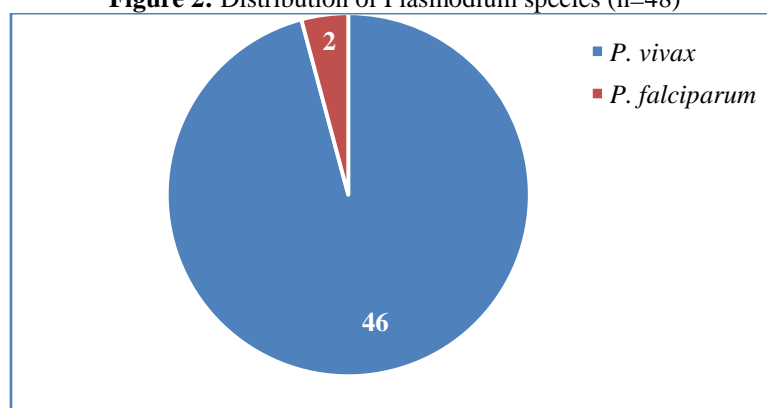
	PBS positive	PBS Negative
<b>RDT positive</b>	38	9
<b>RDT negative</b>	1	7892

Chi: p value < 0.05,significant

Considering PBS as gold standard the Positive Predictive Value of RDT was 80.8% and its Negative Predictive Value was found to be 99.9%. The sensitivity and specificity of RDT in this study was found to be 97.4% & 99.8% respectively.

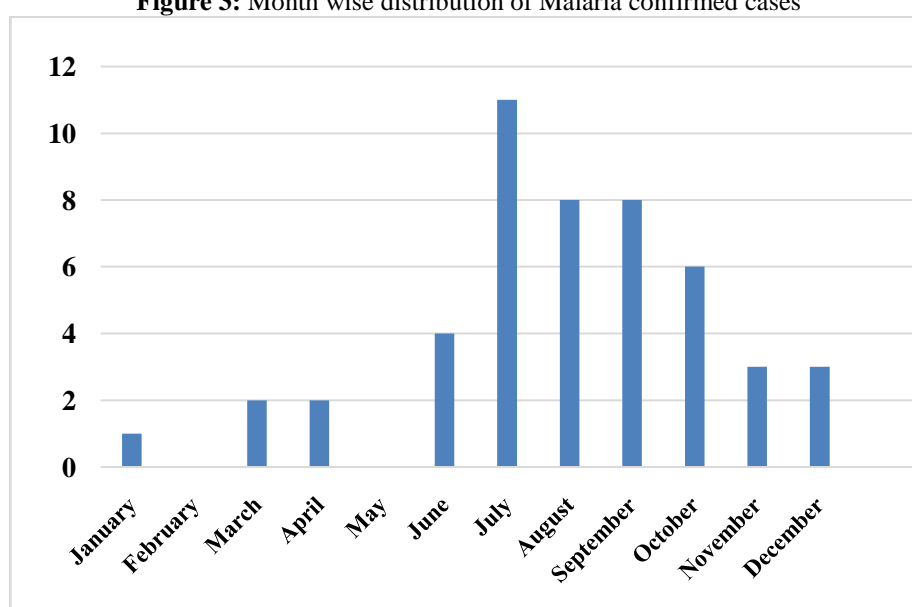
*Plasmodium vivax* was the predominant species isolated in 96%(46/48) cases and in 4%(2/48) cases *Plasmodium falciparum* was demonstrated (Fig 2).

**Figure 2:** Distribution of Plasmodium species (n=48)



Maximum number of cases i.e,77% (37/48) were observed between July to October months of the year. (Fig 3)

**Figure 3:** Month wise distribution of Malaria confirmed cases



#### IV. Discussion

Malaria is a disease of global importance. Malaria presents a diagnostic challenge in most resource poor areas where malaria is endemic. In such areas malaria diagnosis is often made only on the basis of clinical symptoms which is inaccurate<sup>9</sup>. The role of the laboratory diagnosis of malaria is primarily to support and confirm clinical diagnosis<sup>10</sup>. Two most commonly done tests for the diagnosis of malaria are peripheral blood smear (PBS)& rapid card test(RDT). Currently interest is focused on the detection of histidine-rich protein 2 (HRP-2) from *Plasmodium falciparum* and pan lactate dehydrogenase antigen (pLDH)<sup>11</sup>. **World Malaria Day** is celebrated on April 25 every year to promote actions to put an end to malaria.**Directorate of National Vector Borne Disease Control Programme (NVBDCP)** is the central nodal agency for the prevention and control of vector borne diseases including **malaria** and other VBDs (Dengue, Lymphatic Filariasis, Kala-azar, Japanese Encephalitis and Chikungunya) in India.

The national strategic Plan (NSP) has been developed by NVBDCP, Ministry of Health and Family Welfare, Government of India with the support of WHO to provide a road map for making India malaria free by 2027<sup>3</sup>.

In our study total 7940 samples were tested for malaria and the prevalence in our hospital was found to be 0.6%(48/7940). other studies done by various workers with respect to the year and place of study is given in Table no 3

**Table no. 3:** Comparison of prevalence in other studies

Worker	Year of study	Place of study	Duration of study	Sample size	Prevalence
JivabhaiH T et al <sup>12</sup>	2014	Gujarat	3years	1,87,000	2.1%
Mehta R et al <sup>13</sup>	2018	Ahmedabad	9 years	3,00,000	21.1%
Gupta P et al <sup>14</sup>	2018	Uttarakhand	3 years	2982	4.4%
Pardal M P S et al <sup>18</sup>	2009	Assam	5 years	16386-18272	2.64-4.88%
<b>Present study</b>	<b>2019</b>	<b>Uttarakhand</b>	<b>1 year</b>	<b>7940</b>	<b>0.6%</b>

Prevalence in other studies are higher than the present study which can be due to the longer duration and larger sample size of study done by other workers. Study done by Gupta P et al<sup>14</sup> (Rishikesh) which shows prevalence higher than our study. It could be due to more greenery in Rishikesh also patient footfall, in their hospital is mainly from near by areas while our hospital is located in the heart of the city and due to precautionary measures for prevention of dengue fogging is routinely carried out which takes care of malaria also. Malaria incidence in north east region is higher as compared to the other regions which could be explained as, due to abundance of hills, inaccessible tribal areas, poor communication and large scale population movement<sup>18</sup>.

Male preponderance with 69% (33/48) was noted in our present study. Similar male preponderance was seen in published research work by Singh S et al<sup>15</sup> (52%), Singh R et al<sup>16</sup> (58.3%), Kumar S et al<sup>11</sup> (63%), Gupta P et al<sup>14</sup> (67.4%). Male are more prone to mosquito bite due to more outdoor activities, while females fully cover their bodies in comparison to males, which protect them from mosquito bite.

Maximum cases belonged to >15 year of age group i.e., 85% (41/48) which is in concurrence with other studies done by Kumar S et al<sup>11</sup>, Gupta P et al<sup>14</sup>. Most common species observed was *Plasmodium vivax* 96% (46/48) followed by *P.falciparum* 4% (2/48). Similar predominance of vivax species was reported by various workers as shown in (Table no 4)

There is a regional endemicity of plasmodium in different area of the country depending upon the ecological factors<sup>18</sup>.

**Table no 4:** Comparison of predominant species in other studies

Worker	Predominant species	2 <sup>nd</sup> Most Common species	Place of study
JivabhaiH T et al <sup>12</sup>	<i>P. vivax</i> (61.41%)	<i>P. falciparum</i> (38.5%)	Gujarat
Kumar S et al <sup>11</sup>	<i>P. vivax</i> (69%)	<i>P. falciparum</i> (31%)	Rajasthan
Gupta P et al <sup>14</sup>	<i>P. vivax</i> (95%)	<i>P. falciparum</i> (5%)	Uttarakhand
Pardal M P S et al <sup>18</sup>	<i>P. falciparum</i> (86-98%)	<i>P. vivax</i> (14-2%)	Assam
<b>Present study</b>	<i>P. vivax</i> (95%)	<i>P. falciparum</i> (5%)	<b>Uttarakhand</b>

Predominant species variation can occur depending upon the climatic, geographical and also demographic condition like age, density, gender, lifestyle of that area. Seasonal variation was observed in the present study, maximum number of malaria positive cases were observed during July to November which is in concurrence with the various other studies done by Kumar S et al<sup>11</sup>, JivabhaiH Tet al<sup>12</sup>. Seasonal variation in malaria can be seen due to the to annual variation in rainfall, humidity & temperature since rainy season promotes the growth and prolong the survival of the malaria parasite by increasing the breeding sites and by providing favorable conditions for the growth of the parasite which can cause strong seasonal patterns of the disease.

## V. Conclusion

In this study it was observed that Dehradun is not a high prevalence area for malaria. There is gradual decrease in number of positive malarial cases which can be probably due to awareness and preventive measures taken and frequent fumigation carried out by municipal authorities.

Since malaria is a life threatening disease which is preventable and curable therefore there is a need for early diagnosis to decrease the mortality and morbidity for which rapid diagnostic methods like RDT should be used which can be further confirmed by PBS.

Although PBS is a gold standard test but it was observed that many cases were missed by it therefore it is recommended that RDT should also be simultaneously performed.

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