Pakistan J. Zool., pp 1-4, 2021. **DOI**: https://dx.doi.org/10.17582/journal.pjz/20190410130418

Short Communication

Expression of Malaria in Swat Valley, Pakistan

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ABSTRACT

The present study was carried out in Swat valley for malarial disease in 2017. Blood samples were collected from all age groups patients visiting hospital labs and District Head Quarter Laboratory who were recommended for diagnosis of malaria. About 1050 patients were examined and malaria was detected through microscopy of thick and thin blood smears and rapid diagnostic test of which 118 (11.24%) were found infected with malaria parasite. Comparative Data was also collected from other labs of District Swat which accumulated to a total of 9255 patients, among which 932 (10.07%) patients were found positive for malaria parasite. Male infected were 558 (59.87%) while 374 (40.13%) were female. The collective data showed majority of the infected patients belonged to age group 1-10 years (41.42%). The least infected were aged above 60 years (0.86%). Month wise prevalence was found highest in the month of July (39.48%) i.e. summer season and lowest in the month of February (2.25%) i.e. winter month. Patients tested for malaria parasite belonged to the following seven (7) Tehsils wherein the rate of positive infected cases in descending order was: Barikot > Kabal > Babuzai > Matta > Khwazakhela > Charbagh > Bahrain. It is concluded from this study that *P. vivax* is the prevalent malaria causing parasite in district Swat. No case of *P. falciparum* was recorded. Furthermore, male are infected more than female, and malaria is common in children and youth of the area.

Article Information
Received 10 April 2019
Revised 30 une 2019
Accepted 26 September 2019
Available online 17 July 2020

Authors' Contribution
HBS collect-ed field data. SS and MEB designed the experiments. MS analysed the data. HG wrote the article.

Key words
Occurrence, Malaria, Plasmodium, Khwazakhela, Swat valley

Malaria is considered as the most deadly parasitic disease worldwide and in developing countries malaria is credited for the high rate of morbidity and mortality (Kim *et al*., 2006). It is generally transmitted by a female *Anopheles* mosquito acting as a vector for the dispersal of protozoan parasite of the genus *Plasmodium* (Goering *et al*., 2013; Shah *et al*., 2018).

Plasmodium is transferred from mosquito to human blood through its bite. This *Plasmodium* can either enter RBCs or hepatocytes via blood stream (Levinson, 2010). Human Red Blood Cells (RBCs) or erythrocytes are the major site of infection of these parasites (Hulden *et al*., 2014). Five species of *Plasmodium* parasite are known to cause malaria namely: *Plasmodium malariae, P. falciparum, P. vivax, P. ovale* and *P. knowlesi*. In Pakistan *P. falciparum and P. vivax* are commonly found (Collins, 2012).

Malaria is now known as the disease of the tropics (Hulden *et al*., 2014). Large masses of stagnant water are the perfect home for the breeding of *Plasmodium* vector *i.e.* mosquitoes (Khattak *et al*., 2013). Mortality rate due to malaria in 2015 has been recorded as 438,000 globally and about 3.5million cases have been reported from Pakistan (WHO, 2013). In spite of an established malaria control program in Pakistan, annual mortality rate of 50,000 per annum due to malaria has been estimated. Pakistan has been continuously subjected to floods in the recent years which boosted the cases of malaria in the country. Increase in the infection rate by the *P. falciparum* has also been seen due to its resistance to Chloroquine and continuous arrival of the Afghan refugees to the country (Khattak *et al*., 2013). Malaria cases are reported throughout the year in District Swat but the number of cases drops in winter and is found more frequent in summer. Among the various *Plasmodium* species *P. vivax* is known to have the most devastating effect and is the most predominant species (Oliveira-Ferreira *et al*., 2010).

Objectives of the present study were to find out symptomatic cases of malarial patient in general population of Swat valley and to carry out a survey in the diagnostic laboratory registered patient for malarial disease in government hospitals and private diagnostic laboratories of District Swat.

Materials and methods

The present study was carried out in District Swat which is divided into seven tehsils i.e. Babuzai, Bahrain, Barikot, Charbagh, Khwaza Khela, Kabal and Matta

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0030-9923/2021/0001-0001 $ 9.00/0

(Hamayun *et al*., 2006) for a period of 7 months from January to July, 2017. Blood samples were collected from 9255 patients visiting the hospital, malaria diagnostic labs and District Head Quarter Hospital Laboratory showing symptoms for malaria disease belonging to all age groups in seven tehsils of Swat and several areas. Malaria was detected through microscopy of thick and thin blood smears and rapid diagnostic test.

Both thick and thin blood smears were prepared and Giemsa stained appropriately on slides and then observed through microscopy under 100X with oil immersion for the detection of malaria parasite i.e. *Plasmodium* sp. (Shah *et al*. 2016). Clendennen *et al*. (1995) key was used to identify the type of malaria parasite i.e., *P. vivax and P.falciparum* species among the positive samples.

After collection of blood samples in an anti-coagulant tube, 0.5 µl blood was taken and then transferred into the round sample well of the rapid diagnostic test kit (SD Bioline Malaria kit). Afterwards we add assay diluents only 4 drops as instructed by the manufacturer. The presence of two lines after 15 min of test checking was considered as positive whereas if only a single control line appeared it was considered as a negative result. The appearance of three lines would indicate the presence of mixed infection of *P. vivax* and *P. falciparum* (Zeb *et al*., 2015).

Results and discussion

Table I shows the overall findings of the data collected for at DHQ lab and other labs of District Swat. A total of 9255 patients were examined over a period of seven (7) months (Jan-July) among which 932 patients were found positive for malaria parasite showing a prevalence rate of 10.07%. The malaria parasite found was *P. vivax* and no patients suffering from *P. falciparum* was found. Table I shows maximum number of patients was tested in the month of July (24.02%) and the least number of patients were found in January (0.74%). Out of the total 932 infected patients, month wise prevalence of malaria parasite in the district was highest in the month of July (39.48%) and lowest in the month of February (2.25%). Out of the total 932 infected blood samples 558 of them were male (59.87%) while 374 were female (40.13%).

All the patients examined were divided into various age groups i.e., 1-10, 11-20, 21-30, 31-40, 41-50 and above 60 years, the collective data for District Swat showed majority of the infected patients belonged to age group 1-10 years (41.42%), followed by 11-20 years (29.51%) and the least infected age groups were aged above 60 years (0.86%) as shown in Table II. Patients tested for malaria parasite belonged to the following seven Tehsils *i.e.* Barikot, Babuzai, Kabal, Matta, Charbagh, Khwazakhela and Bahrain of District Swat. Table III shows the tehsil wise result for District Swat. The rate of infection in these tehsils showed maximum infection 584 (62.66%) from Barikot followed by 113 (12.12%) from Kabal and least number of patients 1 (0.10) from Bahrain. Moreover, among the 9255 tested patients there were also out district patients which were tested for malaria parasite. The tested samples were 331 (3.57%) in which 30 (3.21%) were infected.

Table I. Month wise result in Swat Valley.

Months	Total tested n (%)	Positive n (%)	P.V	P.F	Male +ve	Female +ve
Jan	690 (7.45)	22 (2.36)	22	0	13	9
Feb	833 (9.00)	21 (2.25)	21	0	12	9
March	919 (9.92)	31 (3.32)	31	0	19	12
April	1185(12.80)	56 (6.00)	56	0	32	24
May	1761 (19.02)	169 (18.13)	169	0	97	72
June	1642 (17.74)	265 (28.43)	265	0	164	101
July	2225 (24.04)	368 (39.48)	368	0	221	147
Total	9255	932	932	0	558	374
%		10.07	100.00	0.00	59.87	40.13

P.V, *Plasmodium vivax;* P.F, *Plasmodium falciparum.*

Table II. Age wise data set of patients n Swat Valley.

Age Group (Years)	Jan	Feb	March	April	May	June	July	Total	Per-centage
1-10	6	6	9	17	75	115	158	386	41.42
11-20	7	3	12	18	49	75	111	275	29.51
21-30	4	9	7	10	18	34	48	130	13.95
31-40	2	1	2	3	14	24	31	77	8.26
41-50	2	2	1	6	5	11	8	35	3.76
51-60	0	0	0	1	8	4	8	21	2.25
Above 60	1	0	0	1	0	2	4	8	0.86
Total	22	21	31	56	169	265	368	932	

Table III. Tehsil wise data of the patients in Swat Valley.

Name Tehsil	Total tested n (%)	P.V n (%)	P.F	MIX	Total
Barikot	2973 (32.12)	584 (62.66)	0	0	584
Babuzai	1379 (14.90)	104 (11.15)	0	0	104
Kabal	1612 (17.41)	113 (12.12)	0	0	113
Matta	1973 (21.31)	48 (5.15)	0	0	48
Charbagh	368 (3.97)	12 (1.28)	0	0	12
Khwazakhela	511 (5.52)	40 (4.29)	0	0	40
Bahrain	108 (1.16)	1 (0.10)	0	0	1
Other	331 (3.57)	30 (3.21)	0	0	30
Total	9255	932	0	0	932
%		10.07	0.00	0.00	10.07

Rate of positive infected cases in these 7 Tehsils in descending order was: Barikot > Kabal > Babuzai > Matta

> Khwazakhela > Charbagh > Bahrain.

In our study a total of 9255 patients were studied for the period of seven months (Jan-July) among which 932 patients were found positive for malaria parasite in the whole district showing a prevalence rate of 10.07%. Literature shows (6.8%) prevalence of malaria parasite in local population of District Mardan, Khyber Pakhtunkhwa, in the year 2013 (Majid *et al*., 2016). A study conducted at district Dir detected 324 (39.5%) out of 821 patients as positive for malaria. It is worth notable that as Dir is warmer as compared to swat hence showing larger number of patients with malaria parasite (Shah *et al*., 2016). Similarly, another study carried out in District Lower Dir, Khyber Pakhtunkhwa, Pakistan in 2011 reported a collection of 3760 blood samples, diagnosis was done using RTD and microscopy, results showed 12.2% positive cases among which *P. vivax*, *P. falciparum* and mixed species (*P. vivax* and *P. falciparum*) were found in the ratio of 94.3%, 3.9% and 1.7%, respectively (Zeb *et al*., 2015).

Month wise prevalence of malaria parasite in the district was found highest in the months of July (39.48%) i.e. summer season and lowest in the month of February (2.25%) i.e. winter months. The overall results showed a gradual increase from January towards July showing a higher rate of infection in summer months instead of winter months. Similarly, Muhammad and Hussain (2011) also showed maximum prevalence in August (11.66%) i.e. summer months than March (3.98%) *i.e.* winter months which showed the minimum prevalence rate. Malaria parasite showed more prevalence at the beginning of summer and lower in winter in another study (Khan *et al*., 2016). July was also reported to be the month showing maximum prevalence of malaria parasite (Ibrahim *et al*., 2014). *P. vivax* occurrence was reported minimum in January and maximum in June (Ahmad *et al*., 2013). An investigation was carried out in 20 different localities of District Bolan, Baluchistan, Pakistan, to find out the prevalence of malaria in the general population. The study showed *P. vivax* (86.2%) and *P. falciparum* (13.7%), both parasites also showed seasonal variation with *P. vivax* at peak (91.4%) and lowest (71.4%) in January, whereas *P. falciparum* was found at peak (28.5%) in January and lowest (8.5%) in December (Yasinzai and Kakarsulemankhel, 2009). The reason behind the maximum prevalence of *P. vivax* during summer season might be the gradual rise in temperature and humidity hence climatic fluctuation providing the parasite more favorable condition hence bringing a speedy increase in the occurrence of malaria parasite i.e. *P. vivax* in the district.

The present study showed a total of 932 infected blood samples 558 of them were male (59.87%) while 374 were female (40.13%). Male and female percentage of infection shows similarity to the study of Zeb *et al*. (2015) that also showed 58.5% male infection rate to 41.5% female. Similar high susceptibility rate of male 7.10% as compared to female 6.52% was showed during another study (Muhammad and Hussain, 2011). Khan *et al*. (2016) and Shah *et al*. (2016) also discussed male being at more risk to malaria than females. A higher rate of infection (75.9%) was also shown in a study conducted in the District Ziarat and Sanjavi (Yasinzai and Kakarsulemankhel, 2009). Another study of Yasinzai and Kakarsulemankhel (2013) at district Panjgur also describes male to be found at high risk (78%).

Age groups (1-30) i.e. children, teenagers and youth were found most susceptible to malaria in this study of seven months in district Swat. Similar study showed individuals between ages 21-30 years at higher risk of infection to both species (Shah *et al*., 2016). Another study of prevalence of malaria parasite in local population of District Mardan, Khyber Pakhtunkhwa, Pakistan in 2013 showed individuals having age between 0-20 years showed high rate of infection (47.4%) (Majid *et al*., 2016). A study in 2001 carried out in District Buner showed 6.86% cases positive for malaria parasite. Individuals having age 1-10 years were found more susceptible (11.58%) (Muhammad and Hussain, 2011). A survey was carried out in the religious schools of District Bannu in 2002, to assess the infection of malaria disease, Age wise difference was noted showing results of high to intermediate to low prevalence rate in the following order: (5.52%) in 5-9 years, (3.37%) in 10-14 years and (2.2%) in 15-19 years (Shah *et al*., 2012). A survey in district Panjgur, Pakistan during July 2006 to June 2008 wherein a total of 6119 cases were studied among which 2346 (38.3%) were reported as positive, 81.2% (n=334) and 80% (n=860) was the rate of infection of the malaria parasite in age groups 1-10 and 11-20, respectively (Yasinzai and Kakarsulemankhel, 2013).

Among the seven tehsils, tehsil Barikot, Kabal and Babuzai are found to be the major infected areas, furthermore, cold areas like Bahrain show less than 1% infection. Study by Ali *et al*. (2013) indicated cases recorded from urban and peri-urban areas were 496 and 1400, respectively which shows peri-urban areas as having higher incidence of malaria as compared to urban areas. *P. vivax* was found to be the dominant specie. A survey was carried out in the religious schools of District Bannu in 2002, which showed that rural areas had more prevalence (4.56%) than urban ones (2.38%) (Shah *et al*., 2012).

In the current study *P. vivax* was found as the prevalent malaria causing parasite in District Swat and no case of *P. falciparum* was recorded. A survey of district Panjgur (considered as among the hottest areas of the Baluchistan province), Pakistan, was conducted from clinics and hospitals which suggested a higher rate of infection by malaria mainly through parasite of *P. vivax*

and also minor but serious threat can also be posed by *P. falciparum* especially cerebral malaria (Yasinzai and Kakarsulemankhel, 2013). Similarly, District Mardan, shows 92.56% positive cases of *P. vivax*, hence found dominant species in comparison to *P. falciparum* (7.44%) (Majid *et al*., 2016). A one year study conducted from October 2013 to September 2014 in district Dir Lower showed presence of *P. vivax* (30.1%) and *P. falciparum* (9.4%) and no cases of mixed species infection were found (Shah *et al*., 2016). A study in 2001 conducted at District Buner recorded 6.86% cases as positive for malaria parasite. No mixed infections were observed and *P. vivax* and *P. falciparum* was found in the range of 5.78% to 1.08%, respectively (Muhammad and Hussain, 2011). Similar findings were reported in a survey carried out in the religious schools of District Bannu in 2002, and found only infection caused by *P. vivax* parasite (Shah *et al*., 2012). Similarly, an investigation carried out in 20 different localities of district Bolan, Baluchistan, also showed majority of the patients were infected by *P. vivax* (86.2%) and a few from *P. falciparum* (13.7%) (Yasinzai and Kakarsulemankhel, 2009). A study in 2009-2011 in the public and private labs of Karachi showed 78.6% of cases of *P. vivax* and also a few cases of *P. falciparum* (Jamal *et al*., 2014). A survey conducted in 2006-2008 in District Lasbella of Pakistan found *P. vivax* was in 860 (65.5%) patients and 451 (34.4%) patients were found with *P. falciparum*. Hence, *P. vivax* was concluded as the dominant species (Yasinzai and Kakarsulemankhel, 2012).

Acknowledgment

The authors are grateful to DHQ lab and other hospital labs Swat for providing lab facilities for sample collection for the present study.

Statement of conflicts of interest

The authors declare no conflicts of interest.

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