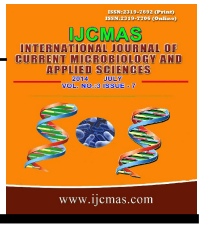
Int.J.Curr.Microbiol.App.Sci (2014) 3(7) 635-643

International Journal of Current Microbiology and Applied Sciences
ISSN: 2319-7706 **Volume 3 Number 7 (2014) pp. 635-643**
http://www.ijcmas.com



Original Research Article

Diagnostic Study on Plasmodium In Blood Bags at Taiz City, Yemen

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Keywords

Malaria; blood bags; stages of Plasmodium

A B S T R A C T

Malaria is one of the most important tropical infectious diseases and the estimated prevalence rates in the world between 300-500 millions while the mortality rate is estimated between one million- two million people every year around the world. It's one of the endemic and widespread diseases greatly in Yemen especially Taiz governorate. This work was based on diagnostic study for the presence of malaria parasite in the blood bags where 100 samples for thick and thin blood films were collected from blood bags in different hospitals (4 Public and 3 privates) Al-Askary, Aljamhori, Althwra, Socity blood friends(AlRawda, AlSafwa and Al-Abbas hospitals, Private hospitals) time was from 5th June-25th November, regardless the age or sex. Thick and thin blood samples were prepared and stained by Giemsa stains then readed under light microscope. Data analyzed by used Statistical Program (SPSS 17) No others diagnostic tests used like ELISA,ICT,PCR , only the thick and thin blood film method was used in the research. It was found that the total malaria cases in the blood bags is about 8.33% (5 positive cases out of 100samples) 3 positive malaria cases out of 16 samples were from Al-Askry hospital (Highest rate)and 2 positive malaria cases out of 16 samples were from Al-Jamhori hospital .The main Plasmodium species was P.falcip1999: arum(100%) .The main Phases were the ring form which was about 5 % and the gametocyte phase was about 3 %. The thick film method was more accurate in diagnosis than thin film method as showed in table (5)

Introduction

Malaria remains one of the most serious global health problem annually there are 300-500 million cases [1], [2]. Malaria kills between 1-2 million people every year most cases of malaria are found in sub-Saharan Africa [3].

Malaria caused by Plasmodium falciparum remains the major life-threatening parasitic infection in the world [4],[5]. The transmission of malaria parasite by blood transfusion is a serious risk, since the diagnosis of malaria in the donor, being

unexpected, is often missed[6]. *P.malariae* is a serious transfused risk (27% of transfused malaria in USA) because of its very long persistent as an asymptomatic infection(25 years or more)[6].Transfused malaria due to *P.falciparum* is less frequent, as the parasite has normally only longevity of less than 1year [6]. Parasites will remain viable in stored blood for over 10 days, particularly since all modern blood preservation contain dextrose [6].

Transfusion associated malaria is a potentially serious complication that continues to pose risks in blood bank setting[7]. There is in need for effective malaria screening of blood donations to improve the current exclusion and on blood donors during blood transfusion .The use of PCR is not, despite its much increased sensitivity, complete guarantee of safe blood, since the absence of parasites in a 20µL sample doesn't exclude the possibility of infection in the remaining 450 of the blood unit [6].

Malaria is one of the most common diseases in Republic of Yemen, malaria cases in the our country are registered all over the year, P. falciparum malaria cases are much more frequent than those of tertian and quartan malaria[8]. Plasmodium falciparum represented >90 %, 20% of these cases resistant to chloroquin [9] In Yemen, about 12 million individuals constituting 60%of the total population live in the malarious area, the estimated number of malaria cases was 3 million in 2001 [10] out of them more than 90% were due to *P.falciparum*, malaria causes 10% deaths of children [11] and 12.5% school absentism[12]. RIII (high level of resistance to anti malaric therapy) was recorded for first time in Yemen and should be taken into consideration in the treatment of infected malaria patients [13]

Materials and Methods

General Information about Yemen and the area of study

The Republic of Yemen is located at the south-southwester edge of Arabian Peninsula. Sana'a is the capital of Yemen. The country has a total area of approximately population 25 million 555.000 Km^2, and subdivided into 20 governorates. Population is mainly dependent on agriculture. Yemen has a predominantly semi-arid to arid climate (tropical to subtropical) with rainy seasons during spring and summer, and with high temperatures prevailing throughout the year in low-altitude zone. Annual rainfall varies from year to year and from place to place [14],[16].

Study area

Taiz governorate situated about 250Km South Sana'a Capital of Yemen. It lies in the foothills and middle heights, which range from 200-2000m elevation from sea level The climate has many sub- tropical features, the mean annual temperature between 20-30C^0 with little seasonal variation and relative humidity ranging between 40-60%. The annual rainfall is approximately 800-1200mm, and most of this fall in March – May and August and September.The majority of population is working in agriculture which is the primary source of income. The study was in Taiz city Republic of Yemen. We collected blood samples from blood bags, time was from 5th June to 25th November 2011 and the totals of samples were 100 samples. These samples were taken from the following directorates: -

-Directorate of(kahira)
-Directorate(Sala)

The present study done in these directorat s hospitals of Taiz city, Al-askri, Aljomhwri, AL-thawrh,society blood friends, AL-abbas, AL-safwh and AL-rawda hospital. All blood samples taken, regardless the age or sex, the study design is stratify random samples from the previous mentioned hospitals. We used pre-designed questionnaire sheet for each case, which included information a bout locality, time of collection, name of hospitals, methods for ex. Smear, type of malaria parasites and its stages. We used only one diagnostic method for detect malaria parasite in blood bags (microscopic method, thick and thin smear). No other diagnostic methods used in our study. Thick and thin blood smears preparation and their Giemsa staining was according to Cheesbrough-Monica: 2006 [17], Bruce chwat, 1988[15] Analysis of data: By using the statistical program (SPSS 17).

Results and Discussion

A total of 100 blood specimens were collected from blood bags during the period from 5th June to25th November 2011 investigated the malaria parasite in different hospitals in Taiz city as follow Al-askri, AL-jomhwry, AL-thawrh, society blood friends, AL-abbas, AL-safwh, and Al-rawda, regardless the age and sex .We used 2 methods for diagnosis thick and thin films, and we noticed the films under microscope. The result reported that 5 samples out of 100 samples taken from blood bags were positive malaria cases (8.33%).The result reported that in Al-askri hospital three cases from sixteen specimens were malaria positive (18.75%), follow by AL jomhwry hospital, two positive malaria cases (12.5%) from sixteen specimens and the other hospitals there was no cases of malaria infection found in blood bags**.**

Specimens diagnosed by two methods, Blood Smear for thick film and thin film. The result showed that the diagnosis by thick film method was more accurate than the thin film; there were 4 cases positive for malaria out of 100 cases by using thick film method, and only one positive case diagnosed by thin film.

From all 5 positive specimens we found one type of Plasmodium species, Plasmodium falciparum that had main distribution in study sites (100%).The phases of P.falciparum noticed under microscope were three ring forms and two gametocytes and there was no Plasmodium vivax, Plasmodium malariae or Plasmodium ovale in these specimens.

In our study which was about the examined the blood bags samples for malaria parasites, we found that the transfused malaria was a serious problem and ignored by a lot of laboratories and doctors, as an important condition for safety transfused blood. In our study we found that 5 cases were positive for malaria out of 100 samples obtained from blood bags and all the positive transfused malaria was P. falciparum, and the most phases were ring forms 3 cases and gametocytes 2 cases. All the blood samples were examined under microscope. and staining with Giemsa stain, thick &thin film for M P S.

In comparison with other studies did in different countries, there was study in Sudan by (Button EL, 1993) [18] said that the prevalence of infected donors with malaria was 6.5% and the recognized species was P. falciparum (98.1%) follow by P. vivax (1.9%). Also there was other study in Pakistan did by (Dover AS, 1971) said that a total of 1585 blood donors were screened for the presence of malaria parasite by using microscope, 9 cases were positive

(0.577%). & the predominant species was P. vivax and P. falciparum respectively, and the common phases were ring forms and gametocytes. This study agreed with our study in the types of phases. In Saudi Arabia , there was study by(Arafa AS.1992) [20] said that 12 cases of transfused malaria was obtained in one Centre in AL Riyadh ,the species was P. falciparum ,and 2 cases of post-operative transfusion malaria follow cardiac surgery were reported from AL Madina city (K.S.A) In U S A, noticed that P.malariae was a serious transfused risk (27%) of transfused malaria in U S A (Guerrero IC, 1993) [21]. (Robert slinger et al, 2001) [22]. said that 3cases of transfusion-transmitted malaria in Canada were noticed, all of them were P.falciparum, in 1997, 1995 & 1994. In Brazil, diagnosed one case who lived in Sao Paulo, his thick film confirmed P.malariae, he was asymptomatic donor (Scuracchiop,et al, 2011) [23].

Sample from 595 blood donors were collected in 7 hemotherapy centre in Northern Brazil located in areas at risk for malaria transmission, and analyzed were performed by real-time PCR with TaqMan probes on 7500 Real-time PCR system ,to genotype the mitoch.DNA region specific to P. vivax .The analysis identified 8 Individuals in the sample (1.34%) infected with P.vivax at the time of blood donation(Sergio Batista-dos-Santos et al 2012)[24],but in our study we only used microscope for diagnosis and we found only one Plasmodium species which was P.falciparum .

Fig.1 Malaria: geographical distributions in world (Yemen)

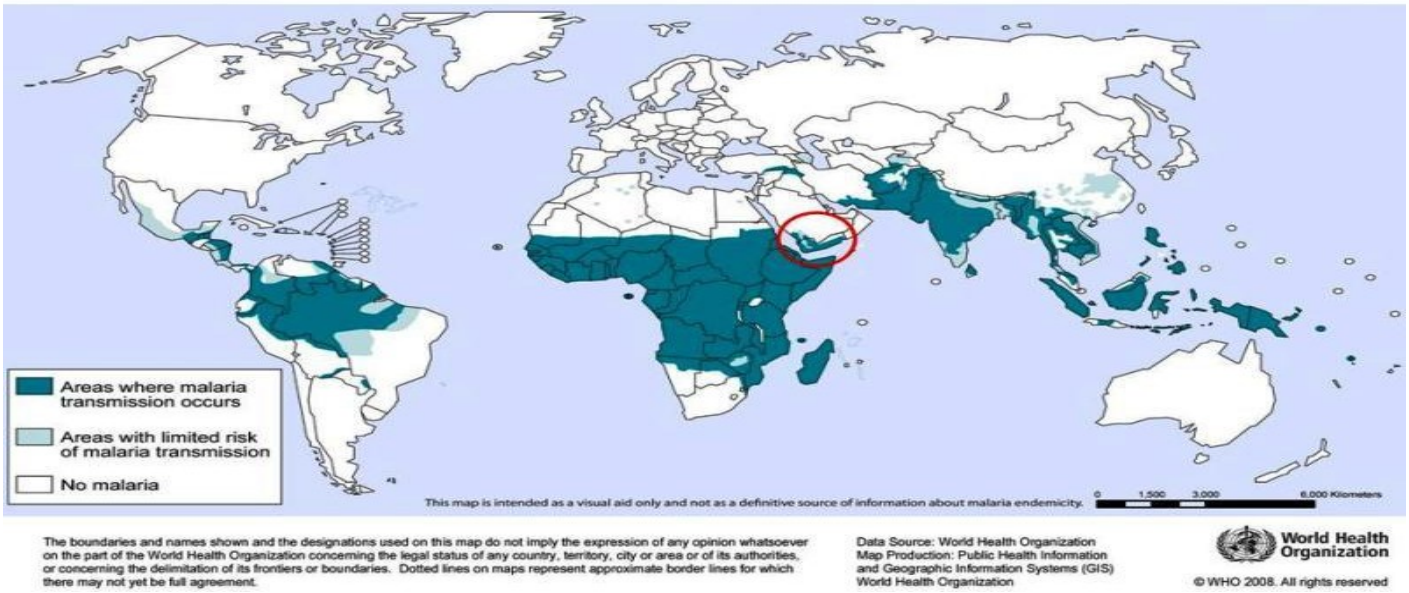


Table.1 Shows number of infected malaria cases during 1998 till 2006/Yemen(11)
Materials and Methods

Species transmitted	Cases estimated	Cases confirmed	Total cases reported	Years
P. falciparum >P.vivax	3.000.000	*NA	NA	1998
P. falciparum >P.vivax	3.000.000	NA	278.640	1999
P. falciparum >P.vivax	3.000.000	NA	1.394.495	2000
P. falciparum >P.vivax	3.000.000	NA	NA	2001
P. falciparum >P.vivax	3.000.000	68.122	172.482	2002
P. falciparum >P.vivax	3.000.000	50.404	162.164	2003
P. falciparum >P.vivax	3.000.000	48.756	158.561	2004
P. falciparum >P.vivax	900.000	44.150	200.560	2005
P. falciparum >P.vivax	900.000	55.000	217.270	2006

Table.2 No. of positive and negative malaria cases obtained from blood Bags

Total examined samples for malaria	Total positive malaria samples	%	Total negative malaria samples	%
100	5	5%	95	95%

Table.3 Types of hospitals and the No. of samples collected

	Total cases collected	Positive cases
Private hospitals	42	Zero
Public hospitals	58	5
Total	100	5

Table.4 Showing results of blood samples test in the following hospitals

NO	Name of hospitals	Number of slides	Diagnosis by Blood film			
			Positive	Ratio%	Negative	Ratio%
1	Al-askri	16	3	18.75	13	81.25
2	AL-jomhwr	16	2	12.5	14	87.5
3	AL-thawrh	16	-	-	16	100
4	AL-safwa (private)	16	-	-	16	100
5	AL-abbas. (private)	16	-	-	16	100
6	Al-rawda (private)	10	-	-	10	100
7	Society blood friends	10	-	-	10	100
Total	7-hospitals	100	5		95	

Fig.2 Result of blood samples test in different hospitals in Taiz city

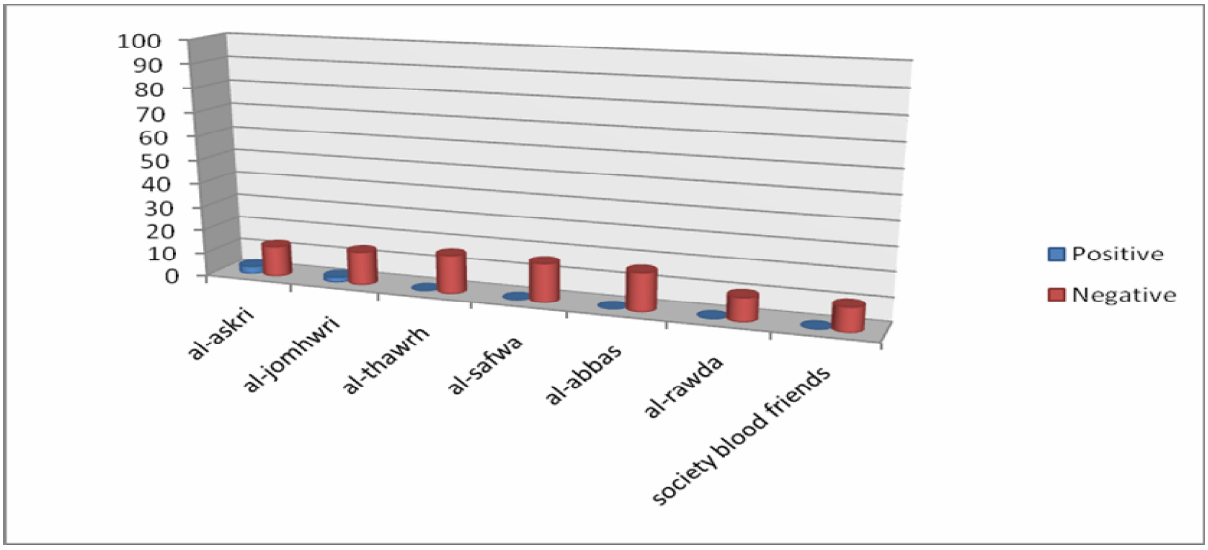


Table.5 Showing the results of thick and thin film in diagnosis of blood samples obtained from blood bags

Result	NO. of Blood samples	Diagnosis by thick film	Diagnosis by thin film
Positive Samples	5	4	1
Negative Samples	95	96	99
Total	100	100	100

Table.6 It showed the distribution of the Plasmodium species and type of phases in the 5 positive cases

NO	Types of plasmodium specie	Diagnosis by blood film		
		No of cases total	%	Type of phases
1	Plasmodium falciparum	5	3%	ring forms
			2%	Gametocytes
2	Plasmodium vivax	0		0
3	Plasmodium malariae	0	0	0
4	Plasmodium ovale	0	0	0

Fig.3 Plasmodium falciparum 100

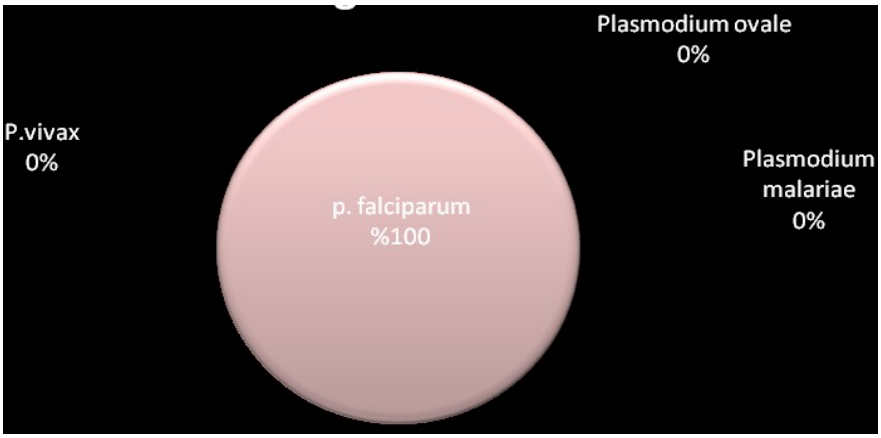
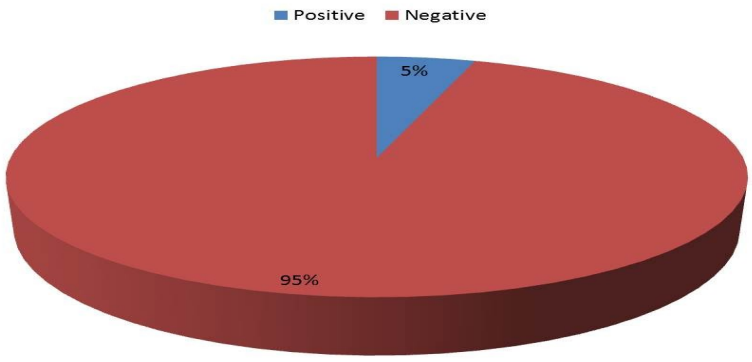


Fig.4 Show Positive and negative malaria blood samples that obtained from blood bags



The present study found Plasmodium species inside some samples of blood that collected from blood bags, which indicated the neglecting the malaria tests before collecting blood from donors and before transfusion to the healthy persons.

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