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Study on Protective Effects of Malaria Antibody among the Community in Malaria Endemic Areas

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Malaria antibodies have been associated with transmission intensity and antibody responses are not much varied in seasonal condition. Antibody assessment is probable to provide a useful epidemiology tool. This study aimed to detect prevalence of antibody in different risk areas in different seasons. Community-based, cross-sectional descriptive study was conducted at high and moderate risk areas during rainy and dry seasons in 2012-2013. Rapid diagnostic testing was used for examination of antigens and antibodies. Microscopic examinations were done. Among total 414 participants, malaria antigens and antibodies (*P. falciparum/ P. vivax*) were detected in 17.9% (74) and 19.1% (79) of participants, respectively. Participants with older age (35±12.7 years) had more prevalence of antibody than younger ones (31.4±14.4 years). Mean difference was 3.6 years (p=0.040). Antibody prevalence was higher in participants of high risk areas (20.4%) than those of moderate risk areas (17.6%). Variation of antibody prevalence between rainy and dry season was less than that of antigen prevalence. In high risk areas, antibody was 25.5% in rainy season and 10.4% in dry season. Antigen prevalence had much variation with 32.9% in rainy season compared to 5.2% in dry season in high risk area. However, in moderate risk areas, antibody prevalence was 17.7% in rainy season and it remained with 16.7% in dry season. The antigen prevalence was 13.3% in rainy season and it was (0%) not found in dry season in moderate risk areas. Therefore, less seasonal variation of antibody prevalence between rainy and dry season was observed in moderate risk areas. The study concluded that protective effects of malaria antibody were observed in older age and associated with transmission intensity. Therefore, antibody assessment can probably provide useful epidemiology tool as it has less seasonal variation.

Key words: Malaria antibody and antigen, Seasonal variation, Endemic area

INTRODUCTION

Malaria is one of the most severe public health problems worldwide. It is not only a leading cause of death but also a disease in many developing countries, where young children and pregnant women are mostly affected. According to the World Health Organization's World Malaria Report (2009) and the Global Malaria Action Plan; 3.3 billion people (half the world's population) live in areas at risk of malaria transmission in 109 countries worldwide, 35 countries (30 in sub-Saharan Africa and 5 in Asia) account for 98% of global malaria deaths. In 2008, there were estimated 190-311 million clinical malaria episodes, and 708,000-1,003,000 deaths. Malaria is the 5th cause of death from infectious diseases worldwide (after respiratory infections, HIV/AIDS, diarrheal diseases, and tuber-culosis) in low-income countries.[1]

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In Myanmar, according to Myanmar Health Statistics (2010), malaria morbidities were 10.75, 10.00 and 11.28 (per 1,000 populations) in 2008, 2009 and 2010, respectively. Mortalities were 1.84, 1.64 and 1.33 (per 100,000 populations) in 2008, 2009 and 2010, respectively. In 2010, percentage of populations living under malaria high risk, moderate risk, low risk and no risk areas were 22%, 25%, 16% and 37%, respectively.[2]

Malaria infection caused by five species of *Plasmodium* (*P. falciparum, P. vivax, P. ovale, P. malariae* & *P. knowlesi*), produces malaria antibodies in one or two weeks after initial infection.[3] These antibodies persist for three to six months after parasite clearance and remain months or years in semi-immune persons in endemic areas where re-infection is frequent. However, in a naive patient, antibody levels fall rapidly.[4]

Antibody prevalence was different in areas with stable and unstable malaria. In stable malaria area, antibody was more commonly found than unstable malaria area.[5] Accordingly, malaria epidemics could be expected to occur in areas with unstable malaria due to lack of antibody protection in those areas. This antibody protection might also be varied with epidemiological characteristics of the community people such as age, occupation, past history of malaria, migration, high risk group, bed net utilization, past history of treatment, geography of residing areas and existing activities of preventive and control measures in the community. Therefore, by using antibody surveys, antibody prevalence resulting different antibody protection may be analyzed in various epidemiologic conditions of the communities.

Malaria antibody responses are long lasting in human blood and those are not much varied in seasonal conditions. Therefore, cumulative responses can be detected in the malaria areas. In addition, antibody detection may facilitate to get accurate findings for malaria situation of the area more easily than antigen detection and microscopic examination for malaria parasites as these examinations cannot detect the infection for long duration, in low parasite density and in areas with low transmission. Moreover, microscopic examination results rarely reveal valid results and which may not reflect situation of malaria in the area. Thus, antibody detection may reveal not only endemicity situation of the area but also reveal the antibody protection among the community.[6]

Malaria antibody assessment by ELISA is probable to provide a useful epidemiology tool which can detect malaria endemicity in transmission areas.[7, 8] Malaria antibody to circumsporozoite antigen has been associated with malaria transmission intensity[9] and also related with repeated exposure to infection in studies done at Brazil and Siri Lanka.[10, 11]

General objective of the study was to detect the protective effects of malaria antibody among the community in malaria endemic areas in selected townships of Mandalay region. This study detected the levels of antibodies acquired against *Plasmodium falciparum* (P.f) and *Plasmodium vivax* (P.v) in residents living in selected townships of Mandalay Region. The study provided the serological data showing antibody protection in different epidemiological information. In addition, malaria situations of different areas were explained by antibody survey. Thus, this valuable information may be useful to apply in the stratification of malaria endemicity of the areas for prevention and control activities.

MATERIALS AND METHODS

A community-based, cross-sectional descriptive study was conducted during wet season, June to November in 2012 and dry season, February to March in 2013 at two villages (Chaung Gyi and Milann) of Thabeikkyin Township and two villages (Gaelong and Banpon) of Pyin Oo Lwin Township in Mandalay Region. In those villages, Milann, and Banpon were high risk transmission

areas (Stratum 1a; according to stratification done by VBDC) because those are mountainous and located in the forest. These areas also have migrant population in development project sites which are gold mine and dam construction.

The other two villages were clarified as moderate risk transmission areas (Stratum 1b) because those are 1-2 miles away from the edge of forest and able to access to nearest health center within 1-3 hours by means of travel available.

Sample populations

A total of 414 participants were recruited in the study. Among these, 188 were from the villages of moderate risk and the others 226 were enrolled from the villages of high transmission areas.

Data collection and data management

Pre-tested pro-forma was used to collect the data concerning epidemiological characteristics of the residents in study townships. Total 30 microliters of blood samples were collected to do (a) microscopic examinations for malaria parasites (b) rapid diagnostic tests for malaria antigens (c) rapid diagnostic tests for malaria antibody. Binocular microscope was used for microscopic examinations of malaria parasites in field surveys.

Parasite density was calculated as multiplication of parasite count by 8000 per number of WBC counted. Rapid diagnostic test (RDT) kits (SD Standard Diagnostics, INC) were used for examination of antigens and antibodies of P.f and P.v. Malaria Antigen P.f/P.v Test is one step, rapid, qualitative and differential test for the detection of HRP-II (Histidine-rich protein II) specific to P.f and pLDH (*Plasmodium* lactate dehydrogenase) specific to P.v. Diagnosis of antibody is the immunochromatographic (rapid) test for the detection of all isotypes of antibodies (IgG, IgM, IgA) specific to P.f/ P.v antigen, Merozoite Surface Protein (MSP). Sensitivity (95% Confidence Interval (CI)) of the RDT for malaria antigen P.f and P.v are 99.7% (98.5-100%) and 95.5% (90-98.1%). Specificity (95% CI) for both antigen is 99.5% (97.2-99.9%). Sensitivity of the RDT for malaria antibody P.f and P.v are 87% and 86%. Specificity of both antibodies is 99.5%. The data relating epidemiological characteristics and serological results were collected with proforma during surveys. The recorded data were compiled, coded, entered into SPSS 20.0 software and analyzed.

Ethical consideration

Ethical approval was obtained from Department of Medical Research (Pyin Oo Lwin Branch) Ethics Committee to collect the blood samples and epidemiological statements for interview questionnaires from the participants.

RESULTS

A total of 414 participants were included in the study. Among them, 307(74.2%) were studied in rainy season and 107(25.8%) in dry season. Males were 279(67.4%) and females were 135(32.6%). Mean age (in year) of participants was 32.1±14.1. The youngest was 2 and eldest was 75 years. Age grouping were made as: young for below 10 years, teen age for 10 to 19, working group for 20 to 49 and old age for 50 and above. According to age group, 29(7%), 40(9.7%), 304(73.4%) and 41(9.9%) were distributed in below 10, 10-19, 20-49 and 50 and above years of age group, respectively.

Forty percent (40.1%) of them were farmers. The occupants such as fishermen, wood cutters, gold mine workers, laborers and charcoal makers were observed in 13.3%, 12.6%, 9.7%, 6.5% and 6.3% of the participants, respectively. More than half of respondents i.e. 232(56%) were not bed net users absolutely. Nearly half of the respondents, 188(45.4%) were from moderate risk areas and 226(54.6%) participants were from high risk areas. Malaria antigen for either P.f or P.v was found in 74(17.9%) and antibodies were observed in 79(19.1%) of total participants.

Table 1. Antibody and antigen prevalence by malaria risk area (n=414)

		High risk area (n=226)				Moderate risk area (n=188)				Total (%)
		Ab(+) (%)	Ag(+) (%)	Ab+Ag (%)	Tested	Ab (+) (%)	Ag (+) (%)	Ab+Ag (%)	Tested	
Age	<10	2(13.3)	2(13.3)	1(6.7)	15	0(0)	1(7.1)	0(0)	14	29(7)
(Year)	10-19	3(13)	6(26.1)	0(0)	23	1(5.9)	1(5.9)	0(0)	17	40(9.7)
	20-49	20(12.1)	23(13.9)	16(9.7)	165	26(18.7)	15(10.8)	1(0.7)	139	304(73.4)
	50 +	2(8.7)	3(13)	2(8.7)	23	4(22.1)	2(11.1)	1(5.6)	18	41(9.9)
Sex	Male	17(11.3)	20(13.3)	15(10)	150	19(14.7)	15(11.6)	2(1.6)	129	279(67.4)
	Female	10(13.2)	14(18.4)	4(5.3)	76	12(20.3)	4(6.8)	0(0)	59	135(32.6)
Season	Rainy	19(12.8)	30(20.1)	19(12.8)	149	26(16.5)	19(12)	2(1.3)	158	307(74.2)
	Dry	8(10.4)	4(5.2)	0(0)	77	5(16.7)	0(0)	0(0)	30	107(25.8)
Total (%)		27(11.9)	34(15)	19(8.4)	226	31(16.5)	19(16.1)	2(1.7)	188	414

Ab(+)=Antibody positive, Ag(+)=Antigen positive, Ab+Ag=Both antibody and antigen positive

Antibody prevalence among age group in high risk areas showed as 13.3%, 13%, 12.1% and 8.7% in below 10, 10-19, 20-49 and 50 and above years of age group, respectively. In moderate risk areas, antibody positive were found in 0.0%, 5.9%, 18.7% and 22.1% in below 10, 10-19, 20-49 and 50 and above years of age group, respectively (Table 1). Participants with older age (35±12.7 years) had more prevalence of antibody than younger aged participants (31.4±14.4 years). Mean difference was 3.6 years (p=0.040).

Among total 414 participants, by RDT testing, antibody positive for P.f, P.v and mixed cases were found in 51(12.3%), 19(4.6%) and 9(2.2%) participants, respectively. Antigen positive for P.f, P.v and mixed were found in 38(9.1%), 31(7.5%) and 5(1.2%) participants, respectively. Those are shown in Table 2 and Table 3.

On microscopic examination, all antigen positive cases i.e. 74(17.9%) were microscopically detected with parasitaemia. On the other hand, all antigen negative cases were not detected with parasitaemia on microscopic examination in this study.

Antibody prevalence by endemic areas and by seasons

In high endemic area, 27 had antibody positive and 19 had mixed with both antibody and antigen among 226 tested population. In moderate endemic area, 31 had antibody and 2 had mixed with antigen and antibody. Therefore, antibody prevalence was higher in high risk areas with 20.4% (27+19) of 226 tested participants residing in those areas and it was lower in moderate risk areas with 17.6% (31+2) of 188 tested participants residing in respective areas. Similarly, antigen prevalence was higher in high risk areas with 23.5% (53 of 226 tested populations) and it was lower in moderate area with 11.2% (21 of 188 tested populations).

Table 2. Antibody and antigen prevalence by season in high risk area (n=226)

Rainy(n=149)						Dry(n=77)					
AB(+) (%)			Ag(+) (%)			AB(+) (%)			Ag(+) (%)		
P.f	P.v	Mix	P.f	P.v	Mix	P.f	P.v	Mix	P.f	P.v	Mix
20 (13.4)	14 (9.4)	4 (2.7)	23 (15.4)	21 (14.1)	5 (3.4)	5 (6.5)	3 (3.9)	0 (0)	2 (2.6)	2 (2.6)	0 (0)

Ab(+)=Antibody positive, Ag(+)=Antigen positive, P.f=*P. falciparum*, P.v=*P. vivax*

Table 3. Antibody and antigen prevalence by season in moderate risk area (n=188)

Rainy(n=158)						Dry(n=30)					
AB(+) (%)			Ag(+) (%)			AB(+) (%)			Ag(+) (%)		
P.f	P.v	Mix	P.f	P.v	Mix	P.f	P.v	Mix	P.f	P.v	Mix
22 (13.9)	1 (0.6)	5 (3.2)	13 (8.2)	8 (5.1)	0 (0)	4 (13.3)	1 (3.3)	0 (0)	0 (0)	0 (0)	0 (0)

Ab(+)=Antibody positive, Ag(+)=Antigen positive, P.f=*P. falciparum*, P.v=*P. vivax*

Antibody prevalence was 25.5% (38 of 149 tested participants) in rainy season and it was only 10.4% (8 of 77 tested participants) in dry season in high risk area. However, in moderate risk areas, antibody prevalence was 17.7% (28 of 158 tested participants) in rainy season and it remained with 16.7% (5 of 30 tested participants) in dry season. There was large difference of antigen prevalence between rainy and dry season in both areas. The antigen prevalence was 32.9% (49 of 149 tested populations) in rainy season and it was only 5.2% (4 of 77 tested populations) in dry season of high risk area. Similarly in moderate risk area,

the antigen prevalence was 13.3% (21 of 158 tested population) in rainy season and it was (0%) not found in dry season. According to prevalence of antibody and antigen for P.f and P.v by seasonal distribution in both moderate and high risk areas, more percentage of antibody and antigen were found in rainy season than dry season. However, antibody prevalence variation between rainy and dry season was less than antigen prevalence variation between rainy and dry season. Those are described in Table 2 and Table 3.

DISCUSSION

Studies on malaria antibody showing in seasonal and endemic area difference are very few in Myanmar. This study provides prevalence of malaria antibody not only in dry and wet season but also in different endemic areas.

Seroprevalence of malaria antibody among females was lower with 6.2% (14/226) in high risk regions. It was higher among males with 14.2% (32/226) in high risk areas. However, it was lower in males in dry season with 2.1% than females with 9%. Antibody prevalence was higher among older aged participants (35±12.7 years) than younger aged participants (31.4±14.4 years). Mean difference was 3.6 years (p=0.040). Study done in Gambia revealed that seroprevalence was increased with age and lower in males 17.5% than females 21.3%.[12]

In this study, malaria antibody assessment showed difference in antibody prevalence between high and moderate risk areas. Antibody prevalence was higher in high risk areas (20.3%) than moderate risk areas (17.6%). Antibody prevalence was also associated with antigen positive rate. The antigen was higher in high risk areas with comprising 23.5% (53 of 226 tested populations) than moderate risk areas which comprised 11.2% (21 of 188 tested populations). Therefore, this study revealed that malaria antibody assessment could estimate malaria endemicity in malarious areas. A biochemical study done in Tanintharyi Region reported that malaria antibody assessment was a useful tool for rapid detection of malaria transmission intensity. It revealed that malarial antibodies reflect cumulative exposure and is less influenced by seasonality due to the longer duration of specific antibody responses. In the study, malaria antibody was found in 121(27.07%), 32(5.77%) and 23(5.08%) for *Plasmodium falciparum* and in 62(13.87%), 20(3.60%) and 21(4.63%) for *P. vivax* malaria in high, moderate and low transmission microstratified areas, respectively. Malaria antigen for *P. falciparum* was discovered in 39 (11.47%), 4 (0.72%) and 1(0.23%) and for *P. vivax* 11(3.24%), 1(0.18%) and 4(0.93%) in high, moderate and low transmission areas, respectively.[13]

This study also revealed different antibody prevalence between rainy and dry season in both areas. According to prevalence of antibody and antigen for P.f and P.v by seasonal distribution in both moderate and high risk areas, more percentage of antibody and antigen were found in rainy season than dry season. However, antibody prevalence variation between rainy and dry season was less than that of antigen. Antibody level remains in sera for longer period after parasite clear. Therefore, antibody level showed less seasonal variation. A study done in Mandalay also revealed as malaria antibody responses are long lasting in human blood and those are not much varied in seasonal conditions. Therefore, cumulative responses can be detected in the malaria areas.[6]

Conclusion

According to the results of this study, it was concluded that malaria antibody was more prevalent in elder age group than younger age group. Antibody was higher in high risk areas than moderate risk areas. Another point to be considered was that malaria antibody prevalence in studied area had less seasonal variation. Antibody prevalence variation between rainy and dry season was less than that of antigen.

Therefore, it was also concluded that transmission endemicity could be estimated by antibody assessment in the malaria endemic areas.

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