

PARASITOLOGY RESEARCH DIVISION

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Parasitology Research Division has been linked with National Malaria Control Programme to fulfill the research needs to solve the priority disease problem of National Health Plan. It is one of the research divisions principally conducting malaria research projects under WHO Collaborating Centre for research and training of malaria. With strong collaboration and support of WHO, therapeutic efficacy studies on currently used artemisinin combination and *in vitro* antimalarial effects of currently used drugs and reputed traditional medicines are carried out. At molecular aspect, it is the only place in Myanmar capable of falciparum malaria parasites genotyping to confirm drug resistance and also capable of doing K13 gene analysis by sequencing in collaboration with University of Maryland. Parasitology Research Division is now taking responsibility to detect asymptomatic malaria by ultrasensitive quantitative PCR method to explore the asymptomatic malaria burden in malaria pre-elimination phase. Real time reporting to NMCP focusing on therapeutic efficacy status of currently using drugs and updated molecular information on drug resistant malaria has been made.

RESEARCH PROJECTS

1. COMMUNICABLE DISEASES

1.1 MALARIA

1.1.1 Drug Resistant Malaria

1.1.1.1 New surveillance tools for malaria elimination in Myanmar (2015-2016) (US qPCR method)

For this project, sample collection surveys were conducted during 2016 from proposed 3 study sites namely Shwe Kyin, Madaya, and Buthidaung. From each study site, 600 samples of whole blood and filter paper samples were collected via finger prick of apparently healthy persons. DMR took responsibility to do molecular analysis of all samples

collected from different study sites by different collaborating partners (DMR, MMA, CPI, Burnet, and PSI).

All samples were properly registered and stored at (-80°C). Ultra sensitive quantitative PCR (US qPCR) method (Level of Detection = 16 parasite/ml) was applied to detect asymptomatic parasitemia. Percent of infected samples with any malaria detected by USqPCR, among 3 study sites was 1.28%. RDT positivity of 3 study sites was 0.33%. The results of the samples collected in 2015 in the same study sites showed total malaria RDT positivity rate of 0.5% (3 out of 600) and malaria positivity rate by USqPCR as 8.3% (50 out of 600). Compared to 2015 research findings, malaria prevalence of these study sites was found to be decreased (0.5% vs 0.3% in RDT, and 8.3% vs 1.3% by USqPCR).

Malaria positivity at DMR 3 study sites detected by RDT and USqPCR (2016)

Study site	Positivity by RDT				Positivity by US qPCR			
	PF	PV	Mixed PF+PV	Total infected	PF	PV	Mixed PF+PV	Total infected
Shwe Kyin (n=599)	1 (0.1%)	2 (0.3%)	0	3 (0.5%)	1 (0.2%)	5 (0.8%)	0	6 (1%)
Madaya (n=600)	0	1 (0.02%)	0	1 (0.02%)	0 (0%)	7 (1.2%)	1 (0.2%)	8 (1.3%)
Buthidaung (n=600)	0	2 (0.3%)	0	2 (0.3%)	2 (0.3%)	7 (1.2%)	0 (0%)	9 (1.5%)
Combined (n=1799)	1 (0.1%)	5 (0.3%)	0	6 (0.3%)	3 (0.2%)	19 (1.1%)	1 (0.1%)	23 (1.3%)

1.1.1.2. Monitoring of drug resistant malaria by therapeutic efficacy trial and molecular tool (Day 3 parasite positivity of ACT (artemether-lumefantrine combination) in uncomplicated falciparum malaria cases in Homalin township, Sagaing Region)

A study to determine day 3 parasite positivity of uncomplicated *Plasmodium falciparum* infected cases after artemether-lumefantrine (AL) combination treatment was conducted in Homalin, Sagaing region during transmission season of 2015-2016. A total of 1627 clinically suspected malaria cases were screened for malaria parasite by RDT (rapid diagnostic test) and confirmed by microscopy. Malaria parasite positivity rate by RDT was 2.9% (48/1627). After confirmation by microscopy, malaria positivity rate was 2.3% (37/1627). Among infected cases, *Plasmodium falciparum* was 89.2% (33/37), *Plasmodium vivax* was 5.4% (2/37) and mixed infection rate was 5.4% (2/37). All falciparum infected 33 cases were enrolled in the study. These enrolled participants were treated with AL treatment and followed them up to Day 3. There were no case of parasite positive on Day 3, therefore effectiveness of artemether-lumefantrine on uncomplicated falciparum malaria in Homalin was 100% on day 3.

1.1.1.3 Molecular diagnosis of the samples collected from “Malaria indicator survey (2015)” to detect asymptomatic malaria

“Malaria indicator survey 2015” (MIS) was conducted by Malaria Consortium under the guidance of National Malaria Control Programme. A total of 13,873 dried blood spot (DBS) samples from 4495 households were collected from four malaria endemic settings (domain 1= annual parasite index, API > 5, domain 2 = API 1 - 5, domain 3 = API < 1,

domain 4 = hard to reach areas). Among these samples, 13,726 DBS were found eligible for molecular analysis and ultra sensitive quantitative PCR method was applied to detect asymptomatic malaria at Parasitology Research Division and Molecular Technology Applications Division in 2106. Overall malaria prevalence rate was 3.83% (526/13,726). Species prevalence in over all samples was *Plasmodium falciparum* was 0.6% (80/13,726, 95% CI (0.5-0.7)), *Plasmodium vivax* was 3.1% (418/13,726, 95% CI (2.8 – 3.4)) and mixed (*P.f* and *P.v*) infection was 0.2% (28/13,726, 95% CI (0.1 – 0.3)). According to endemic setting, total malaria positivity by USqPCR was 10.96% in domain 4, 3.46% in domain 1, 1.24% in domain 3, and 0.19% in domain 3. The finding pointed out the decreasing prevalence of *Plasmodium falciparum* and increasing *Plasmodium vivax*. *Plasmodium vivax* is dominant in all four domains. About three-quarter of the *Plasmodium falciparum* (77%) and most of the *Plasmodium vivax* infections (99.83%) were asymptomatic. These findings also highlighted the asymptomatic malaria burden which is to be considered in formulating malaria elimination strategies.

1.1.1.4 Establishment and standardization of *in vitro* artemisinin sensitivity testing against *Plasmodium falciparum*

Artemisinin resistance in *Plasmodium falciparum* has emerged in Southeast Asia and now poses a threat to the control and elimination of malaria. Malaria is still one of the priority diseases and a re-emerging public health problem in Myanmar. This study was conducted to establish the *in vitro* artesunate sensitivity assay using field isolates and standard strain of *Plasmodium falciparum* and to determine *in vitro* artesunate sensitivity against *Plasmodium falciparum* in Shwe Kyin Township, Bago region. The cross sectional descriptive study was done in Shwe Kyin Township during the malaria transmission season of 2016. A total of 1430 clinically suspected malaria patients were screened for malaria parasite by Rapid Diagnostic Test and Giemsa microscopy. Malaria Parasite positivity rate by Giemsa microscopy was 2.73% (39 out of 1430 cases). As regards to species prevalence, *P. falciparum* was detected in 16 cases (41.03%), *P vivax* was 22 cases (56.41%) and mixed infection was 1 case (2.56%). Only 14 *P.falciparum* isolates were criteria matched for artesunate susceptibility applying WHO Mark III. Collected samples were properly stored in the liquid nitrogen and transported to Department of Medical Research. At the Parasitology Research Division, 14 collected samples had been properly thawed and adapted to culture system. The collection of malaria positive samples is still in progress to obtain the required sample size.

1.1.1.5 K13 gene sequence analysis of the samples collected in Therapeutic Efficacy Study (TES) (Buthidaung, Myitkyinar, Muse)

Artemisinin resistant *P. falciparum* was reported in Cambodia, and now confirmed in several Greater Mekong Subregion (GMS) countries. Mutations in *P. falciparum* Kelch propeller gene, commonly known as K13, were shown be associated with artemisinin resistance in Cambodia and specific regions of GMS. These mutations are expected to predict clinical efficacy of ACT, and the assessment of K13 mutations was recommended by the WHO, as part of studies to routinely monitor therapeutic efficacy of ACTs at the country level. Therapeutic Efficacy Study (TES) of artemether-lumefantrine (AL) and dihydroartemisinin-piperaquine (DHA-PIP) were conducted in 2014-2015 in three sentinel sites in Myanmar: Buthidaung, Rakhine State, Myitkyinar Kachin State and Mu-se Shan State. A total of 293 dried blood spot day 0 samples (59 AL and 39 DHA-PIP from Mu-Se,

50AL and 44 DHA-PIP from Myitkyinar, and 58 AL and 43 DHA-PIP from Buthidaung) were investigated for K13 gene sequence analysis.

Prevalence of K13 mutations in three study sites Sequence type	Total number of samples and prevalence rate	Number of samples and prevalence rate of Mu-Se	Number of samples and prevalence rate of Myitkyinar	Number of samples and prevalence rate of Buthidaung
Number of readable sequence	293	97	95	101
Wild type	157/293 (53.5%)	26/97 (26.8%)	30/95 (31.6%)	101 (100%)
F 446 I mutation	113/293 (38.56%)	55/97 (56.7%)	58/95 (61.1%)	0
G 453 D mutation	1/293 (0.34%)	1/97 (1%)	0	0
N 458 Y mutation	1/293 (0.34%)	0	1/95 (1.1%)	0
S 485 N mutation	2/293 (0.68%)	2/97 (2.1%)	0	0
R 561 H mutation	1/293 (0.34%)	0	1/95 (1.1%)	0
C 580 Y mutation	5/293 (1.70%)	5/97 (5.2%)	0	0
Q 661 R mutation	1/293 (0.34%)	1/97 (1%)	0	0
A 676 D mutation	7/293 (2.38%)	6/97 (6.2%)	1/95 (1.1%)	0
V 692 L mutation	5/293 (1.70%)	1/97 (1%)	4/95 (4.2%)	0

Nine resistance-associated K13 mutations were observed and overall mutation rate was 46.4% (136/293). In relation to clinical outcomes, 50% of the day 3 and 100% of day 21 parasite positive samples harbored F446I mutation but it was not statistically significant, as small number of failure cases were available in this study. Important K13 mutation, C580Y associated with artemisinin resistance in Cambodia was detected in 5 samples (5.2%) of Mu-Se, Shan State and it was not associated with ACT failure. The study reported the prevalence of K13 mutations together with therapeutic efficacy of different combination of ACT. This finding approved that K13 mutation was not found among ACTs sensitive parasite population circulating along Myanmar-Bangladesh border. Among K13 mutations, F446I was most common among 2 study sites close to Myanmar-China border.

1.1.1.6 Serological analysis of the samples collected from “Malaria Indicator Survey (2015)”

In areas where malaria transmission is low, serological surveillance may provide additional information about the level of transmission and risk factors for exposure. Antibodies are made as part of immune response to infection and they are species specific and their prevalence increases with the age. Therefore seroconversion rate (SCR) reflects transmission intensity. Myanmar malaria indicator survey was conducted during malaria transmission season of 2015, a total of 12210 dried blood spot samples from different malaria

endemic areas (domain 1= annual parasite index, API > 5, domain 2 = API 1-5, domain 3 = API < 1, domain 4 = hard to reach areas) were collected. Out of that, 11760 blood samples were found eligible for serological analysis. Antibodies to *Plasmodium falciparum* merozoite antigen (Pf MSP-1), and apical membrane protein (Pf AMA-1), *Plasmodium vivax* (Pv MSP-1) and (Pv AMP-1) were measured by ELISA method. The findings showed that 17.7% were seropositive for PfAMA-1, 12.4% were positive Pf MSP-1, 8.3% positive for Pv AMA-1, and 6% positive for Pv MSP-1. Age specific sero-prevalence identified 2 sero-conversion rates and age above 15 year had 10 times greater risk of exposure. Area specific conversion rates were correlated with annual parasite index (API). Multiple logistic regression identified older age, low socio-economic status, living in rural areas and working in the forest as major risk factors for malaria. Serological surveillance can be applicable in low transmission settings (especially pre-elimination, elimination, and post-elimination era) when entomological measures and parasitological interventions are difficult to implement. Risk factors identified in this study could be used to formulate malaria interventions.

1.1.2 Field Research on Malaria

1.1.2.1 Efficacy and safety of artemether-lumefantrine (A-L), and dihydroartemisinin-piperaquine for the treatment of uncomplicated *Plasmodium falciparum* malaria in sentinel sites (2015 - 2016) (Paletwa, Buthidaung, Beelin, Thandaung)

Paletwa, Chin State

The study was conducted during transmission seasons of 2015 and 2016 in Paletwa. A total 36 uncomplicated falciparum malaria cases in artemether-lumefantrine combination trial and 13 cases in dihydroartemisinin-piperaquine (DHA-PPQ) trial were enrolled. Enrolled patients were monitored clinically and parasitologically upto day 28 in AL and day 42 in DHA-PIP treated group. Day 3 parasite positive cases were detected in 15.4% (2/13) of DHA-PIP treated group and none of AL treated group. One case of Late Parasitological Failure (LPF) was detected in AL treated group. Adequate Clinical and Parasitological Response (PCR corrected) ACPR was 97.2% in AL group and 100% in DHA-PIP. In parallel to falciparum malaria case recruitment, a total of 7 vivax cases were enrolled chloroquine trial. There was no treatment failure case and ACPR was 100%.

Therapeutic outcome of antimalarials in Paletwa

	AL in falciparum malaria	DHA-PIP in falciparum malaria	CQ in Vivax malaria
Early Treatment Failure (ETF)	0	0	0
Late Clinical Failure (LCF)	0	0	0
Late Parasitological Failure (LPF)	1	0	0
PCR-corrected Adequate Clinical and Parasitological Response (ACPR)	35 (97.2%)	13 (100%)	7
Total analysis	36	13	7
Day 3 (+)	0	2 (15.4%)	0

1.2 Parasitic Diseases

1.2.1 Molecular verification of *Schistosoma* spp. infection in Shwe Kyin Township, Myanmar

Helminth infections such as nematodiasis, trematodiasis and cestodes are among the most prevalent NTDs in low and middle income countries. Helminth infections are most commonly found in Southeast Asia, and the flukes are endemic in all countries of the Southeast Asian peninsula, particularly in communities where raw fish food consumption is culturally rooted. Similar ecological setting with agro-based economy, being a member of GMS, Myanmar is no exception. However scientific documentation on these flukes is limited in Myanmar. With the aim to explore the diverse helminthic population, a cross sectional descriptive study was conducted during June-August 2016, in Shwe Kyin Township, Bago region. A total of 204 subjects were included in the study. Stool samples were collected from the study participants and were examined by Kato-Katz method for detection of helminth's eggs. The stool samples were stored at -20°C for further molecular verification. By Kato-Katz method, 39% (n=80/204) of the study population were found to be infected with at least one species of helminth. A diverse helminth infection among study population was noted. The most prevalent one was *Ascaris lumbricoides* 20%, (n=41/204). *Schistosoma* spp. infection was detected in 2.9% (n=6/204). Molecular verification of schistosome species infection was carried out at National Institute PD, Chinese CDC during July, 2016. A total of 31 stool samples with microscopically positive for at least one helminth, were verified by conventional PCR method. By molecular method, 8 samples were diagnosed as *Schistosoma mekongi* infection. The findings of the study reported the presence of schistosomiasis in Myanmar and also confirm *Schistosoma* species prevailing in Myanmar. The results provide some ideas to choose potential study sites with favourable geographical and risky dietary habit for future research.

1.2.2 Infection with soil-transmitted helminthes and related knowledge, attitude and practices in Phyu Township in Myanmar

Monitoring of the chemotherapeutic STH control is needed especially in the plains and coastal areas, and epidemiological situation should be assessed for integrated STH control with water supply, sanitation and hygiene promotion (WASH) component for sustainable results. Thus, the study was conducted with the objectives to (1) assess the prevalence and intensity of STH infection with *Ascaris lumbricoides*, *Trichuris trichiura* and hookworm in schoolchildren attending the fifth grade in Phyu (the plains area) (2) identify the knowledge, attitudes and practices regarding STH infection, STH treatment, hygiene behaviors, sanitation and water supply; and (3) identify the potential risk factors related to STH infections. It was a cross-sectional study, consisting of a parasitological survey and a questionnaire survey on STH infections and WASH. A total of 363 fifth graders in 10 randomly selected schools in Phyu Township were invited to participate in this study, and 279 of them (aged 9-16 years) completed the survey questionnaires with the guidance from the researcher, and 274 of them provided a stool sample each. Stool samples were examined for the presence of eggs of any of the STH parasites stated above. Stool examination results showed that 33% (90/274) of study participants were infected with any STH, 14% (38/274) with *Ascaris lumbricoides*, and 25% (69/274) with *Trichuris trichiura*. About one-third to half of the participants did not know that intestinal worms can be transmitted through eating fly-infested food (46%, (128/279), walking barefoot (32%, 89/279), eating unwashed raw vegetables (53%, 148/279) and playing with soil (38%, 106/279). About three-quarters of the participants believed that they are likely to have intestinal worms. Although the reported

toilet use (98%) and washing hands with soap after toilet use (46% always and 35% most of the time) were high, 93% reported eating food by hand and only 15% reported always using soap before eating and 34% most of the time. The results of the study have shown that, despite the targeted deworming, STH infections remain prevalent among schoolchildren in the plains-area sentinel site. The findings of the study also highlighted the areas for health education among schoolchildren for integrated STH control.

2. TRADITIONAL MEDICINE

2.1 *In vitro* anti-malarial activity of ethanol extract of *Zingiber cassumunar* Roxb. (Meik-tha-lin-oot) on *Plasmodium falciparum*

This study was carried out in collaboration with Chemistry Department, MawLaMyaing University. The aim of this study is to evaluate the *in vitro* anti-malarial activity of ethanol extract of *Zingiber cassumunar* Roxb. (Meik-tha-lin-oot). *Zingiber cassumunar* Roxb. (Meik-tha-lin-oot) were collected from Chaungzone Township, Mon state. After cleaning and drying at room temperature, the dried samples were ground into powder and stored in air-tight containers. Crude extracts (Petroleum-ether and 95% Ethanol) were prepared by successive solvent extraction method. The extracts were concentrated by vacuum rotatory evaporator under reduced pressure to yield Petroleum-ether and 95% ethanol extracts. In this study, ethanol extract and watery extracts were used for *in vitro* anti-malaria sensitivity test. 20 blood samples from *P.falciparum* infected patients which fulfilled the selection criteria were investigated for *in vitro* drug sensitivity test applying *in vitro* micro technique (Mark III) (WHO 1987). Chloroquine was used as standard drug. The test dose of ethanol extracts and watery extracts of *Zingiber cassumunar* Roxb (Meik-tha-lin-oot) were used ranging from 40, 80, 160, 320, 640, 1280, 2560 µg/ml and 45, 90, 180, 360, 720, 1440, 2880 µg/ml. Effective doses were determined by using Probit Calculus software provided by WHO. It was found that 95% ethanol extracts and watery extracts show *in vitro* anti-malaria activity against *P.falciparum* ($EC_{50}=398.1\mu\text{g/ml}$) and ($EC_{50}=501\mu\text{g/ml}$). From these results, it can be concluded that 95% ethanol extracts and watery extract of *Zingiber cassumunar* Roxb. (Meik-tha-lin-oot) have no degree of *in vitro* anti-malaria activity on *Plasmodium falciparum*. These two extracts have less *in vitro* anti-malaria activity than that of chloroquine ($EC_{50}=21.1\mu\text{g/ml}$).

SERVICES PROVIDED

ACADEMIC

Sr.	Name	Course	Responsibility
1.	Dr. Kay Thwe Han	M.Med.Sc (Microbiology)	Teaching and Thesis Supervision
2.	Daw Kyin Hla Aye	M.Med.Sc(Microbiology)	Demonstration
3.	Dr. Hnin Wai Lwin	M.Med.Sc(Microbiology)	Teaching

PARASITOLOGY RESEARCH DIVISION (POL)

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	...	Dr. Ei Ei Swe MBBS (UMM)
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Nurse (3)	...	Daw Khaing Khaing Lin BSc (Zoology) (MU)
Research Assistant (4)	...	Daw Myint Myint Khin
	...	U Lai Lian Maung BA (Geography)(Pakokku University)

Parasitology Research Division is mainly involved in research activities relating malaria research such as therapeutic efficacy studies and malaria epidemiology studies. Moreover, the division also conducts the studies on vector borne diseases like dengue infections, and studies on tropical acute febrile illnesses.

RESEARCH PROJECTS

1. COMMUNICABLE DISEASES

1.1 MALARIA

1.1.1 Efficacy and safety of artemether-lumefantrine and dihydroartemisinin-piperquine phosphate for the treatment of uncomplicated *Plasmodium falciparum* malaria, and chloroquine for *P. vivax* in Thabeikyin township (Mandalay Region) and Ta-Mu township (Sagaing Region)

This study aims to assess the efficacy and safety of artemether-lumefantrine and dihydroartemisinin-piperquine phosphate for the treatment of uncomplicated *Plasmodium falciparum* malaria, and chloroquine for *P. vivax*. Study Sites are Thabeikyin township (Mandalay Region) and Ta-Mu township (Sagaing Region). The study is conducted during the malaria transmission season, from June to December 2016. This surveillance study is a one-arm prospective evaluation of clinical and parasitological responses to directly observed treatment for uncomplicated malaria. Febrile patients aged 6 years and above with uncomplicated malaria attending the study health clinic, who meet the study inclusion criteria are enrolled, treated on site with artemether-lumefantrine or dihydroartemisinin & piperquine phosphate for the treatment of uncomplicated *P. falciparum* malaria and chloroquine for *P. vivax* and monitored as 28 days for artemether-lumefantrine and 42 days for dihydroartemisinin & piperquine. The findings revealed that Adequate Clinical and Parasitological Response (ACPR) among artemether & lumefantrine, dihydroartemisinin & piperquine phosphate combination for *falciparum* and chloroquine for *vivax* malaria were: 96.2% .100% and 100% respectively in Thabeikyin township, and 98.1%, 100% and 100%, respectively in Ta Mu township. There was no serious side effects among the patients treated by the tested two regimes, thus they are safe for treatment of uncomplicated *falciparum* and

vivax malaria in the area. The study showed that the anti-malarials recommended for treatment of uncomplicated malaria in Myanmar are quite effective with high ACPR status.

1.1.2 Therapeutic efficacy study using Artemisinin-based Combination Therapies for *Plasmodium falciparum* at Myanmar-India and Myanmar-China border

In Myanmar, three artemisinin-based combination therapies (ACTs) are recommended as first-line treatment of uncomplicated *falciparum* malaria: artemether-lumefantrine (AL), artesunate-mefloquine (AS+MQ), and dihydroartemisinin-piperaquine (DP). Resistance to both artemisinins and ACT partner drugs has been reported from the Greater Mekong subregion, and regular efficacy monitoring of the recommended ACTs is conducted in Myanmar. This paper reports on results from studies to monitor the efficacy of the three ACTs in sentinel sites in Northern Myanmar, and investigations of mutations in the *Kelch13* (*K13*) propeller domain. Seven therapeutic efficacy studies were conducted in 2011-12 and 2014 in three sentinel sites in Myanmar (Tamu, Muse and Thabeikkyin). Three studies were done for the evaluation of AL (204 patients), two studies for AS+MQ (119 patients) and two studies for DP (147 patients). These studies were done according to the 2009 standard WHO protocol. Polymorphisms in the *K13* propeller domain were examined in dried blood spots collected on day 0. The primary end-point was adequate clinical and parasitological response (ACPR) on day 28 for AL and on day 42 for DP and AS+MQ, corrected to exclude reinfection using polymerase-chain reaction (PCR) genotyping. Safety data were collected through self-reporting. PCR-corrected ACPR was 97.2 - 100% for AL, 98.6 - 100% for AS+MQ and 100% for DP across the study sites and years. All studies found a prevalence of *K13* mutations (>440) above 23% in the day 0 samples. The F446I mutation was the most common mutation making up 66.0% of the mutations found. Seven out of nine day 3 positive patients were infected with *K13* wild type parasites. The remaining two cases with day 3 parasitemia had the P574L mutation. The efficacy of AL, AS+MQ and DP remains high in Northern Myanmar despite widespread evidence of *K13* mutations associated with delayed parasite clearance. This study showed that already in 2012 there was a high frequency of *K13* mutations in Myanmar on the border to India. The high efficacy of the recommended ACTs gives confidence in the continual recommendation of the use of these treatments in Myanmar.

1.1.3 Knowledge of high school students about malaria prevention and control in malaria endemic area of Pyin Oo Lwin township

A community based cross sectional descriptive study was conducted with the high school students (9th and 10th standard) from selected three schools situated in malaria endemic rural area of Pyin Oo Lwin Township to assess knowledge about malaria prevention and control and to find out bed-nets utilization among them. A total of 183 students were selected by systematic random sampling method and self-administered questionnaire was used in this study. Out of all students, female occupied (65.5%) and most of students, (78.1%) were at the grade nine levels. Regarding knowledge on cause of malaria, majority of students (93.4%) reported that cause of malaria was due to mosquito bite. Some of students claimed that malaria was caused by bathing in stream (35.0%), eating bamboo shoot, mushroom and banana, (~12.0%) respectively. The students who correctly answered that cause of malaria was due to mosquito bite at night time were only (47.0%). In respect of signs and symptoms of malaria, almost all students (92.9%) had known that fever with chill were the major symptom of malaria. With regard to prevention of malaria, the students reported that using the bed net is a way of preventing malaria was 95.1% and those answered that taking chloroquine drug as preventing malaria was (82.5%). About (80.0%) of all students had known that spraying could prevent malaria infection. The students, (83.6%) answered that

well drainage of water is important to reduce mosquito breeding. Relation to diagnosis and treatment of malaria, the proportion of students reported that malaria could be diagnosed by microscope and Rapid Diagnosis Test were (62.8%) and (77.6%), respectively. Of all students, (76.0%) perceived that malaria disease can be cured by early diagnosis and effective treatment of health workers. They also revealed that malaria was curable by (79.2%) of all. Moreover, they, (92.9%) had perceived threats of severity as malaria is a deadly disease if there is no proper diagnosis and treatment. Although the students, (74.3%) had known that malaria could not be treated effectively by taking drugs from drug shop, about half of all students misunderstood that malaria can be cured without taking complete treatment. The maximal knowledge score was 37 out of 43 total scores while the minimal was 14. The students who got the favorable score which is over average score of 21 was 88.5%. In conclusion, most of the high school students had higher level of good knowledge on causes of malaria and its prevention in this study.

SERVICES PROVIDED

ACADEMIC

Sr.	Name	Course	Responsibility
1.	Dr. Moe Kyaw Myint	Interviewer training on 2016 Facility Assessment for Reproductive Health Commodities and Services	Trainer