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Prevalence of Anaemia, Iron Deficiency Anaemia and Haemoglobinopathies among Pregnant Mothers Attending 300-bedded Pyin Oo Lwin General Hospital

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This hospital- and laboratory-based, cross-sectional descriptive study was done at 300-bedded Pyin Oo Lwin General Hospital from September 2014 to February 2015. The purpose of this study was to determine severity of anaemia and types of abnormal haemoglobin among pregnant women who attended antenatal clinics of study hospital. A total 500 pregnant women were enrolled in which hematological parameters were assessed by using Pentra-60 haematology auto-analyzer and serum ferritin level by Mini-vidas, fully-automated Immunology analyzer. HbH inclusion detection by Brilliant cresyl blue dye test and HbE by NESTROFT (Naked Eye Single Tube Red Cell Osmotic Fragility test) were done and agarose gel electrophoresis by SAS-MX Alkaline Hb-10 kits for qualitatively. The overall prevalence of anaemia was 64.2% and third trimester group was significantly higher than other groups ($p<0.05$). Moreover, 108 out of 321 anaemic cases (21.6%) were iron deficiency anaemia and 89 cases (17.8%) had haemoglobinopathy in which 43 cases (8.6%) haemoglobin E trait (HbAE), 6 cases (1.2%) β-thalassaemia trait ($HbAA_2$), 2 cases (0.4%) haemoglobin E β-thalassaemia (HbEF), 36 cases (7.2%) carrier of α-thalassaemia trait (HbAH) and 2 cases were HbH disease ($HbAA_2H$), respectively. There was positive correlation between haemoglobin level, haematocrit, RBC count, mean corpuscular volume and mean corpuscular haemoglobin with haemoglobinopathy cases ($p=0.001$, $p=0.016$). Therefore, this study recognizes the high prevalence rate and highlights that anaemia remains a common health problem among pregnant women and it is also required for screening programs and clinical management of haemoglobinopathies in this area.

Key words: Anaemia, Iron deficiency anaemia, Haemoglobinopathy pregnant mother

INTRODUCTION

Haemoglobinopathy is the common single gene disorder and it causes severe clinical signs and symptoms as well as clinically asymptomatic. The severity of haemoglobinopathies can vary. Screening is afford able and an accessible way to detect carriers, and can be offered in a range of settings in different societies: in high school, before marriage, or in antenatal clinics. At present, at least 6.5% of children worldwide are carriers.[1] It is estimated that over 300,000 affected children are born each year and most are with sickle cell disease; while about 60,000-70,000 are born with β-thalassaemia. Most affected children are born in countries with limited resources, where priority tends to be given to tackling high rates of infant and child mortality from infection and malnutrition.[2]

The Southeast Asia Region (India, Thailand and Indonesia) accounts for about 50% of the world's β-thalassaemia carriers: about

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40 million people and almost half of homozygote births.[3] The carrier rate of the abnormal haemoglobin is varied among the different races in Myanmar. Carrier couples have 25% risk in each pregnancy that the child will have a serious haemoglobin disorder, and timely information allows them informed choice among reproductive options, including prenatal diagnosis.[4]

The haemoglobin disorders, in particular, are often regarded as incurable and, therefore, "hopeless" and they are expensive to treat. Thalassaemia patients may be left untreated (indeed, they often die without any diagnosis) or grossly under-treated. At the same time, quality of treatment is firmly linked to both survival rates and quality of life. There are serious consequences including limited prolongation of life, with death usually occurring in adolescence or early adulthood, increased complications: major organ damage, transfusion-transmitted infections, poor quality of life and increased psychosocial burden.[2]

Anaemia is also one of the public health problems and iron supplementation is an effective measure for anaemia control. National Anaemia Initiative Project supplied the Ferrous Sulphate tablet and syrup to the population but the prevalence of anaemia is still medium magnitude of public health problem according to the public health significance of anaemia (WHO/ UNICEF/ UNU 2001).[5] Thus, haemoglobinopathy can cause the therapeutic problem with ferrous sulphate tablet and syrup for the anaemia patients.

Therefore, early diagnosis, effective treatment and prevention are important for the patients. Preventing thalassaemia is based on identifying individuals at risk through carrier screening programs or family history and providing adequate information to reduce that risk. The approach to deal with the thalassemic problem is to prevent and control births of the new cases. This requires an accurate identification of couple at high risk to have a thalassemic child. To design such a preventive program and to assess the public health burden in the population, accurate population frequency data on α- and β-thalassaemia in this area are required.

MATERIALS AND METHODS

This hospital- and laboratory-based, cross-sectional descriptive study was done at Pathology Research Division, Department of Medical Research (Pyin Oo Lwin Branch) and 300-bedded Pyin Oo Lwin General Hospital. A total of 500 apparently healthy pregnant women were recruited from the antenatal clinic of Outpatient Department using systematic sampling procedure from September 2014 to January 2015.

Any pregnant woman, who had history of hypertension, diabetes mellitus, acute and chronic infection, tuberculosis, was excluded from this study and their relevant history regarding age, gravida, and general health information was taken. Then, five milliliters of venous blood samples were taken after getting informed consents from the participants after explaining the purpose, risks and benefits of the research. All the samples were collected by using the sterile and disposable materials.

Hematological parameters were assessed by using Pentra-60 haematology auto analyzer for all cases. For biochemical parameters, serum ferritin level was determined by Mini-vidas, fully automated immunology analyzer. HbH inclusion detection by Brilliant cresyl blue dye test and Hb E by NESTROFT (Naked Eye Single Tube Red Cell Osmotic Fragility Test) were done for screening and then agarose gel electrophoresis by SAS-MX Alkaline Hb-10 kits for qualitatively.

Data entry and analysis was done by SPSS software 20.0 version. Frequency charts were defined and continuous variables' averages and standard errors (SE) were calculated. The prevalence rates were calculated with 95% Confidence Interval.

Regarding the ethical consideration, this study was approved by the Institutional

Ethical Committee of the Department of Medical Research.

RESULTS

A total of 500 pregnant women who attended the antenatal clinic during the first, second and third trimesters were 29 cases (5.8%), 193 cases (38.6%) and 278 cases (55.6%), respectively. Age distribution in these subjects was from 16 years to 47 years and the mean age was 27.7 years (SE= 6.1 years). Body mass index (BMI) of all cases were 23.35-49.22 and the mean BMI was 24.98 (SE=3.86). Eighty-four percentages of pregnant women were taken folic acid tablet regularly, 77% took multi-vitamin and minerals and only 31% took iron tablets. Most of the people were Myanmar (72.6%) and educational level was secondary and above (57.2%). In this study, 291 cases (58.2%) had moderate income and their family numbers were not crowded.

Table 1. Basic characteristics of 500 pregnant women

Characteristics	Number of cases	Percent
Ethnics		
Myanmar	363	72.6
Shan	46	9.2
Indian	33	6.6
Gorakha	30	6.0
Others	28	5.4
Gravida		
Primigravida	246	49.1
2-3	213	42.6
≥4	41	8.2
Gestational period (Trimester)		
First	29	5.81
Second	193	38.6
Third	278	55.6
Income (kyats/month)		
<100,000	40	8
100000-200000	291	58.2
>200,000	169	33.8
Education		
Illiterate	9	1.8
Read & Write	6	1.2
Primary	199	39.8
Secondary & Above	286	57.2
No of family members		
≤4	330	66
>4	170	34
History of consanguineous marriage in her parents		
Yes	18	3.6
No	482	96.4

Among 500 women, 246 cases (49.1%) were primigravida, 213 cases (42.6%) were normal gravida and 41 cases (8.2%) were multi gravida. The basic characteristics of enrolled participants are shown in Table 1.

The prevalence of anaemia distributed in this study are described in Table 2.

Table 2. Prevalence of anaemia among pregnant women

Variables	WHO (Hb below 11 g/dl)		
	Percent	Mean	SD
First trimester (n=29 women)	1.8	11.348	1.1639
Second trimester (n=193 women)	27	10.413	1.1743
Third trimester (n=278 women)	35.4	10.584	1.2241
Total (n=500 women)	64.2	10.562	1.2178

According to trimester, the prevalence of anaemia by WHO (Hb<11 g/dl) criteria were 1.8%, 27% and 35.4% in the first, second and third trimesters, respectively. Results showed that the overall prevalence of anaemia among 500 pregnant women was 64.2% and the third trimester group was significantly higher than the other groups (p=<0.05). According to medical history taking of anaemia of these pregnant women, 57.5% of cases reported tiredness and weakness in present pregnancy. In clinically, 53% of the subjects had sign and symptoms of anaemia. The proportion of severity of anaemia in this study is described in Table 3.

Table 3. Proportion of severity of anaemia among pregnant women

Trimester	Severity of anaemia				Total
	Mild	Moderate	Severe	No	
First	8	1	0	20	29
Second	123	10	2	58	193
Third	150	27	0	101	278
Total (%)	281	38	2	179	500
	(56.2)	(7.6)	(0.4)	(35.8)	

On microscopic examination, out of 321 anaemic pregnant women, 57 cases were found to be normochromic normocytic anaemia and 264 cases were hypochromic microcytic anaemia. Serum ferritin level was measured in the 321 potential cases of anaemia, 108 cases (21.6%) were iron deficiency anaemia (By WHO, serum ferritin level <15.0 µg/l).

By means of screening and confirmatory investigations, 89 cases (17.8%) had haemo-

globinopathy in which 43 cases (8.6%) were haemoglobin E trait (HbAE), 6 cases (1.2%) were beta thalassaemia trait ($HbAA_2$), 2 cases (0.4%) were haemoglobin E beta thalassaemia (HbEF), 36 cases (7.2%) were carrier of alpha thalassaemia trait (HbAH) and the rest 2 cases were Hb H disease ($HbAA_2H$). The results of prevalence of haemoglobinopathy with severity of anaemia distributed in this study are described in Table 4.

Table 4. Severity of anaemia and prevalence of haemoglobinopathy among pregnant women

Severity of anaemia	A	B	C	D	E	Total
Mild	6	34	2	35	0	77
Moderate	0	9	0	1	2	12
Severe	0	0	0	0	0	0
Total (%)	6(1.2)	43(8.6)	2(0.4)	36(7.2)	2(0.4)	89(17.8)

A = Beta thalassaemia trait
B = Haemoglobin E trait
C = Haemoglobin E beta thalassaemia
D = Alpha thalassaemia trait
E = HbH disease

The mean haemoglobin level and absolute red cell values as well as other haematological parameters of pregnant women were described in Table 5.

Table 5. Comparison of haematologicalparameters among pregnant women with haemoglobinopathy

Variables	A	B	C	D	E	p value (ANOVA)
Hb (g/dl)	9.8 ±0.2	9.7 ±0.7	9.7 ±0.7	10.2 ±0.5	8.8 ±1.5	**0.001**
Hct (%)	32.4 ±1.0	31.8 ±2.1	32.2 ±1.9	33.0 ±1.9	30.3 ±3.1	**0.016**
RBC (10^9/l)	4.1 ±1.0	4.2 ±0.5	4.2 ±1.6	4.4 ±1.1	4.0 ±1.2	**0.016**
MCV (fl)	71.3 ±8.0	70.5 ±5.0	61.5 ±3.5	72.9 ±4.8	69.5 ±1.2	**0.001**
MCH (pg)	21.7 ±2.6	21.7 ±2.0	18.5 ±0.7	22.6 ±1.9	20.0 ±4.7	**0.001**
MCHC (%)	30.3 ±0.6	30.7 ±1.0	30.1 ±0.6	31.0 ±1.0	28.7 ±2.0	0.464
WBC (10^9/l)	253.3 ±43.1	281.1 ±65.3	237.0 ±58.0	246.9 ±78.2	209.0 ±41.3	0.528
Platelet (10^9/l)	9.8 ±2.6	9.8 ±2.5	9.8 ±1.2	9.1 ±1.6	10.0 ±1.4	0.247

A = Beta thalassaemia trait
B = Haemoglobin E trait
C = Haemoglobin E beta thalassaemia
D = Alpha thalassaemia trait
E = HbH disease

There were significant associations between the haemoglobin level, haematocrit (Hct), RBC count, mean corpuscular volume (MCV) and mean crpuscular haemoglobin (MCH) with haemoglobinopathy cases (p=0.001 and p=0.016). However, there was no significant relation between mean corpuscular haemoglobin concentration (MCHC) (p=0.464) as well as WBC (p=0.528) and platelet count (p=0.247).

DISCUSSION

Anaemia is a global public health problem affecting both developing and developed countries with major consequences for human health as well as social and economic development. It occurs at all stages of the lifecycle, but is more prevalent in pregnant women and young children.[6] In 2002, iron deficiency anemia (IDA) was considered to be among the most important contributing factors to the global burden of anemia.[6]

The prevalence rates of the anaemia were 321 of 500 women (64.2%) and prevalence rate of iron deficiency anaemia (IDA) was 108 of 321 women (21.6%), respectively. Out of anaemic pregnant women, 281 were mildly anaemic, 38 showed moderate anaemia and 2 got severe anaemia. The prevalence rate of anaemia were 1.8%, 27% and 35.4% in first, second and third trimester, respectively. Thus, anaemia is a severe public health problem (>40%) among pregnant women which is consistent with WHO recognition of anaemia as a problem of public health significance.[7]

According to the data from World Health Organization, the global prevalence rate of anaemia among pregnant women was (41.8%, 95% CI=39.9% - 43.8%) according to the national surveys conducted between 1993 and 2005. There were about 31.7 million (41%) of pregnant women suffering anaemia in Asia. In South East Asia Region, the prevalence rate was (48.2%, 95% CI=43.9% - 52.5%) and the highest percentage was noted in Africa Region 19.3 millions (57.1%) (47.5% - 67.6%).[7]

Therefore, the prevalence rate among pregnant women was still high in Myanmar compared to global as well as the regional data. In a hospital-based, descriptive study conducted in Thailand showed the prevalence of iron deficiency anaemia in pregnant women was highlighted the prevalence of anaemia among pregnant women to be 14.1% using the WHO criteria (Haemoglobin below 11 g/dl).[8] This depicted that the prevalence of anaemia in Thai pregnant women was lower than that of the present study.

In a nation-wide, population-based study in India described that 70% of pregnant women in the country are mildly anaemic and 42.6 percent are moderately anaemic.[9] In the present study, however, only 56.2%, 7.6% and 0.4% of pregnant women had mild, moderate and severe degree of anaemia. It may be concluded that the presence of anaemia in this study was mainly mild anaemia and it was as similar as India study.

National Nutrition Centre, Myanmar survey from 2001 to 2003 reported the prevalence rates of anaemia among non-pregnant reproductive age women (in 2001), among adolescent girl (in 2002), among pregnant women (in 2003) and among under five children (in 2003) were 45%, 26.4%, 71% and 75%, respectively.[10] In spite of various programmes started by government, there is no significant decline in the prevalence of anaemia in this study. In this study, only 31% (54 of 500 cases) consumed regular iron tablet and the rest did not. Different population background such as income, education, lack of compliance as well as premature discontinuation of iron tablets were the likelihood of occurrence of anaemia.

A study done in Pyinmana Township, Myanmar, 2010, reported the prevalence of anaemia and iron deficiency anaemia among pregnant women 60.1% and 40.6%, respectively.[11] Although the prevalence rate of anaemia was more or less similar to the present study, the prevalence rate of iron deficiency anaemia was lower in the study compared to other studies. This could be due to different diagnostic criteria for the diagnosis of anaemia.

Moreover, the prevalence rate of anaemia obviously increased from first to third trimester ($p=<0.05$). This finding was the same as many previous studies in pregnant women at Maharaj Nakorn Chiang Mai Hospital.[12- 14] Both haemodilution as well as the increased demand of nutritional status are the common causes of anaemia during pregnancy worldwide. The haemodilution reaches the peak at 32 weeks of pregnancy and hence, the prevalence rate is significantly higher in third trimester period.

Serum ferritin level (below 15 µg/l) was used as the cut-off point to diagnose IDA in the present study according to Iron deficiency anaemia: assessment, prevention, and control. A guide for programme managers, Geneva, WHO, 2001.[15] The prevalence rate of IDA in this study was 21.6% and this result is in accordance with the results of study conducted in Chiang Mai Hospital.[12, 13]

Haemoglobinopathies are the commonest genetic defect worldwide with an estimated 269 million carriers. Certain populations are particularly at risk of having a haemoglobinopathy, for example, in South East Asia, there are 90 million carriers, about 85 million in sub-Saharan Africa and 48 million in the West Pacific region.[16] In this study, there were Myanmar people 72.6% and the rest were different ethnic groups.

In this study, 89 cases (17.8%) had haemoglobinopathy among 500 pregnant women. Other study in Thailand revealed that the prevalence of thalassaemia carrier in pregnant women 138 of 519 cases was 26.6%.[8] It may be concluded that the presence of carrier was lower than Thailand study. This could be due to different diagnostic criteria for the diagnosis of haemoglobinopathy. Therefore, future prospective study is needed the tests to determine the presence of main and minor components of adult haemoglobin which

enables quantitative measurement of HbA, HbA_2 and other abnormal haemoglobin HbE, HbH and also HbF by advance laboratory technology such as HPLC and molecular genetic tests.

Ne Win and co-workers also assessed that two hundred and nine β-thalassaemia alleles of 158 unrelated Myanmar patients (107 HbE β-thalassaemia and 51 β-thalassaemia major) were analysed for β-globin gene mutations.[3] Brown JM, *et al*.[17] working with 85 unrelated Burmese patients found that 14 with homozygous β-thalassaemia, 70 with HbE/ β-thalassaemia and one with HbS/ beta thalassaemia. Six mutations have been identified of which three, the G-T at IVS-1 position 1, the G-C at IVS-1 position 5 and the deletion of TCTT in codons 41/42, accounted for 85% of the alleles.[17]

Regarding the absolute red cell values, the indicators were significantly correlation between the haemoglobinopathy groups (Table 5 and p=<0.05). Furthermore, a significant positive correlation between the haemoglobin level (8.8±1.5) and HbH disease was found in the present study (p=0.007). Other study in Thailand revealed that the best cut-off level of MCH in predicting the thalassaemia carrier was 26.5 picograms. Positive MCH (<26.5 picograms) gave the sensitivity of 95.2% and specificity of 82.3% in screening α-thalassaemia 1 trait and β-thalassaemia trait.[18]

The present study showed a high prevalence of anaemia (64.2%) and 86.5% (77 of 89 cases) were mild degree of anaemia and 13.5% (12 of 89 cases) were moderate anaemia on screening haemoglobinopathy among pregnant women at Pyin Oo Lwin Township in which many races such as Myanmar, Shan, Indian, Gorakha, Ka Chin, Chin, Ka Yin and also Mon are resided and is situated at hilly region above 1000 m sea level. A nationwide study should be conducted to collect or update information on the prevalence and severity of anaemia in various age groups as well as different ethnic groups and socioeconomic groups of the country.

Moreover, public health measures integrated into maternal and child health care programme have to establish to reduce the maternal morbidity due to iron deficiency anaemia. Thus, the results of present study are useful for screening programs and clinical management of haemoglobinopathies in this area.

REFERENCES

1. Reports on update of epidemiology of haemoglobin disorders with special reference to thalassaemia, Department of Obstetrics and Gynaecology, University College London Medical School.
2. Weatherall DJ & Clegg JB. Inherited haemoglobin disorders: An increasing global health problem. *Bulletin of the World Health Organization* 2001; 79(8): 704-12.
3. Ne Win, Harano T, Harano K, Thein Thein Myint, Ra Mra, Okada S, *et al*. A wider molecular spectrum of beta thalassaemia in Myanmar. *British Journal of Haemotology* 2002; 117: 988-992.
4. Reports on Thalassaemia and other haemoglobinopathies. World Health Organization, 2006.
5. Reports on community-based anaemia control project, Department of Health, Ministry of Health, Myanmar, 2007.
6. WHO Report. Reducing risks, promoting healthy life. France: WHO Press: 2002.
7. Worldwide prevalence of anaemia, 1993-2005. WHO Global Database on Anaemia, WHO.
8. Sukrat B, Suwathanapisate P, Siritawee S, Poungthong T & Phupongpankul K. The prevalence of iron deficiency anaemia in pregnant women in Nakhonsawan, Thailand. *Journal of the Medical Association of Thailand* 2010; 93(7): 765-770.
9. Chellan R & Paul L. Prevalence of iron-deficiency anaemia in India: Results from a large nationwide survey. *Journal of Population and Social Studies* 2010; 19(1): 59-80.
10. Reports on community-based anaemia control project, Department of Health. Ministry of Health, Myanmar, 2007.
11. Phyu Synn Oo. Prevalence of anaemia among reproductive age women in urban and rural Areas of Pyinmana Township. Department of Medical Research (Central Myanmar), Research Project Report submitted to WHO Country Office, Yangon, Myanmar, 2010.

12. Wanapirak C, Muninthorn W, Sanguansermsri T, Dhananjayanonda P & Tongsong T. Prevalence of thalassaemia in pregnant women at Maharaj Nakorn Chiang Mai Hospital. *Journal of the Medical Association of Thailand* 2004; 87:1415-8.

13. Chotnopparatpattara P, Limpongsanurak S & Charnngam P. The prevalence and risk factors of anaemia in pregnant women. *Journal of the Medical Association of Thailand* 2003; 86: 1001-7.

14. Sukrat B & Sirichotiyakul S. The prevalence and causes of anaemia during pregnancy in Maharaj Nakorn Chiang Mai Hospital. *Journal of the Medical Association of Thailand* 2006; 89(Suppl 4): S142-6.

15. WHO, UNICEF, UNU. Iron deficiency anaemia: Assessment, prevention, and control. A guide for programme managers. Geneva, World Health Organization, 2001. WHO/NHD 01.3.

16. Angastiniotis M & Modell B. Global epidemiology of haemoglobin disorders. *Annals of the New York Academy of Sciences* 1998; 850: 251-9.

17. Brown JM, Thein SL, Weatherall DJ & Mar KM. The spectrum of beata thalassaemia in Burma. *British Journal of Haemotology* 1992; 81(4): 574-8.

18. Pranpanus S, Sirichotiyakul S, Srisupundit K & Tongsong T. Sensitivity and specificity of mean corpuscular haemoglobin (MCH): For screening α-thalassaemia 1 trait and β-thalassaemia trait. *Journal of the Medical Association of Thailand* 2009; 92(6): 739-43.