Overview of thalassaemia in Myanmar children

* Aye Aye Khaing

Thalassaemia and abnormal hemoglobin are the most common monogenic disorders and it has been estimated that approximately 5.2% of the world population are carriers. (Weatherall, 2005). It has been estimated that the conception of homozygous beta-thalassemia and beta-thalassaemia / Hb E in Southeast Asia is 0.24 and 0.25 per 1000 birth, respectively (Fucharoen S, Winichagoon P, 1987). Moreover, incidence of B thal trait in Myanmar is 0.54% (Aung Than Ba Tu et al 1971), incidence of Hb E in Myanmar is also high, varies from 1% to 28% (Aung Than Batu and Hla Pe, 1971) (Ne Win et al, 2005).

Moreover, in Southeast Asia (SEA) α-thalassaemia, β-thalassaemia, haemoglobin (Hb) E and Hb Constant Spring (CS) are prevalent. The abnormal genes in different combinations lead to over 60 different thalassaemia syndromes, making Southeast Asia the locality with the most complex thalassaemia genotypes. (Fucharoen S, 2011)

Many thousands with beta-thalassaemia disorders are living in SEA. The alpha-thalassaemia is widespread throughout SEA in which 5-15% of the population had some form of alpha-thalassemias. Because of high prevalence of both alpha thalassemia-1 and alpha thalassemia-2 in these populations, HbH disease and Hb Bart’s hydrops fetalis are seen frequently. This causes public health burden for the region. (Fucharoen S, Winichagoon P. 1992). Estimated health burden for Myanmar is calculated as 250 new borns with homozygous beta thalassemia and 260 newborns with E B Thalassemia each year based on total population of -55, 746, 253 (2014) and birth rate 18.6 births/1,000 population. According to the data from TIF publication 2013, expected figure of B thalassaemia syndromes births per year in Myanmar is 2398, in Thailand it is 6983 and in Bangladesh it is 64.

According to above literature, as a country of the SEA, Myanmar is also facing high incidence of thalassaemia. In fact, a national thalassaemia registry is required for the epidemiology which is not available yet.

However few studies have been performed to find out the prevalence in Myanmar. In 1968, Aung-Than- Batu, Khin- Kyi- Nyunt and Hla- Pe, studied 232 Myanmar volunteers, and reported thalassaemia trait in 4.3% (10); out of which 8 have the α-thalassaemia trait, and 2 have β-thalassaemia trait respectively (Aung-Than-Batu, Khin-Kyi- Nyunt and Hla-Pe 1968).
A total of ten cases of thalassaemia major and 31 patients with thalassaemia-HbE were reported in 1971 (Aung-Than-Batu 1971a). The incidence of α-thalassaemia trait is reported to be approximately 10% among the Burmese (Myanmar) patients; and a total of 15 Burmese (Myanmar) with HbH disease were reported in the same year (Aung-Than-Batu, U Hla-Pe, and Khin-Kyi-Nyunt 1971b).

It is well known that genotyping of thalassaemia in SEA is complex, even so, it would be interesting to determine the genotype of thalassaemia in Myanmar as we have quite a lot of ethnicities prone to Thalasaemia ie, Kayin, Mon.

In the year 2006 Ne-Win, Harano and Rai-Mra screened 170 transfusion dependent and unrelated Myanmar thalassaemia patients for three common α-thalassaemia deletions and found out combined α-thalassaemia 1 and haemoglobin H disease represented 8.8% of patients seeking transfusion for refractory anaemia (Ne-Win, 2006).

A study for clinical, haematological, and molecular features of α-thalassaemia in 224 Myanmar patients found the commonest deletional type of α-thalassaemia in Myanmar was α\textsuperscript{3.7}. (Kyaw Shwe 2006). Similar result was observed in a study done in Yangon General Hospital, the commonest genetic abnormality in Myanmar patients with HbH disease was (-SEA/-α\textsuperscript{3.7}) (53 percent) of α-globin genes followed by (-SEA/-α\textsuperscript{4.2}) (30 percent). (Moe Hein 2010).

Keiko Harano, Ne Win and Teruo Harano (2000) reported the results of β-thalassaemia mutations in transfusion dependent thalassaemic children in Myanmar [21 cases with CD 41/42 (-TCTT), 15 cases with IVS 1-1 (G\textsuperscript{→}T), 13 cases with IVS 1-5 (G\textsuperscript{→}C), 8 cases with CD 17 (A\textsuperscript{→}T) and 1 case with IVS II-654 (C\textsuperscript{→}T)]

In 2010, a study Yangon General Hospital found 4 common mutation of Beta Thalassaemia in Myanmar patients which is compatible to the results of the other studies done in South East Asia Region. (Sein Win 2010).

### Common Beta mutation in Myanmar

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<tr>
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<tbody>
<tr>
<td>E/CD 41/42</td>
<td>/β\textsuperscript{+}</td>
<td>(21) 36.21%</td>
<td>(2, 22) .2%</td>
<td>(27), 28.72%</td>
</tr>
<tr>
<td>E/CD 17 (A→T)</td>
<td>/β\textsuperscript{0}</td>
<td>(8) 13.79%</td>
<td>(2, 22) .2%</td>
<td>(19), 20.21%</td>
</tr>
<tr>
<td>E/IVS 1-1 (G→T)</td>
<td>/β\textsuperscript{0}</td>
<td>(15) 25.86%</td>
<td>(2, 22) .2%</td>
<td>(25), 26.60%</td>
</tr>
<tr>
<td>E/IVS 1-5 (G→C)</td>
<td>/β\textsuperscript{-}</td>
<td>(13) 22.41%</td>
<td>(2, 22) .2%</td>
<td>(6), 6.38%</td>
</tr>
<tr>
<td>E/unknown</td>
<td>?β/β</td>
<td>-</td>
<td>(1, 11) .1%</td>
<td>(17), 18.09%</td>
</tr>
<tr>
<td>E/IVSII-654 (C→T)</td>
<td></td>
<td>(1) 1.72%</td>
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</table>
B mutations compare to the other countries

<table>
<thead>
<tr>
<th>Dr Ne Win 2002</th>
<th>Newin, Harano 2000 58 cases EB YCH</th>
<th>Common mutation in Thailand</th>
<th>Sri Linka</th>
</tr>
</thead>
<tbody>
<tr>
<td>IVS 1-1 (G&gt;T)</td>
<td>26% IVS 1-1 (G→T)</td>
<td>IVS I-1 G &gt; T</td>
<td>12% IVS 1-1 G &gt; A</td>
</tr>
<tr>
<td>CD 41/42 (-TCTT)</td>
<td>36% CD 41/42 (-TCTT)</td>
<td>Codon 41/42 - TCTT</td>
<td>2% CD 41/42 (-TCTT)</td>
</tr>
<tr>
<td>IVS 1-5 (G&gt;C)</td>
<td>22% IVS 1-5 (G→C)</td>
<td>IVS I-5 G &gt; C</td>
<td>39% IVS I-5 G &gt; C</td>
</tr>
<tr>
<td>CD 17 (A&gt;T)</td>
<td>14% CD 17 (A→T)</td>
<td>Codon 17 A &gt; T</td>
<td></td>
</tr>
<tr>
<td>IVS II-654 (C&gt;T)</td>
<td>2% IVS II-654 (C→T)</td>
<td>IVSI1-654 C &gt; T</td>
<td>1% IVS II-1 G &gt; C</td>
</tr>
<tr>
<td>nt-28 G (A&gt;G)</td>
<td></td>
<td>ATA-28 A &gt; G</td>
<td>1% IVS II 613 C &gt; T</td>
</tr>
<tr>
<td>CD 35 (TAA)</td>
<td></td>
<td>Codon 35 C &gt; A</td>
<td></td>
</tr>
<tr>
<td>CD 71/72 (+A)</td>
<td></td>
<td>Codon 71/72 +A</td>
<td></td>
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<tr>
<td>IVS1 (-1) (G&gt;C)</td>
<td></td>
<td>1% IVS 1-129 A &gt; C</td>
<td></td>
</tr>
<tr>
<td>CD 41(-C)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>619 bp deletion</td>
<td></td>
<td>3% 619 bp deletion</td>
<td></td>
</tr>
<tr>
<td>CD 5 (-CT)</td>
<td></td>
<td>3% CD6A &gt; Hb S</td>
<td></td>
</tr>
<tr>
<td>CD 8/9 (+G)</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>CD 15 (-T)</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>CD 15 (TAG)</td>
<td></td>
<td>2% CD15 G-A</td>
<td></td>
</tr>
<tr>
<td>CD 16 (-C)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CD 44 (-C)</td>
<td></td>
<td>Codon 19 A &gt; G</td>
<td>3% CD121Hb D</td>
</tr>
<tr>
<td>nt -31 (A&gt;G)</td>
<td></td>
<td>Codon 26 (Hb E) G &gt; A</td>
<td>5% CD26G (Hb E)</td>
</tr>
</tbody>
</table>

So far, all above studies found out the commonest genetic abnormality in Myanmar patients with HbH disease was (-SEA/-α^3.7) (53 percent) of α-globin genes followed by (-SEA/-α^4.2). In Myanmar, six β-thalassaemia mutations (three, IVS 1-1 (G-T), IVS 1-5 (G-C), CD 41/42 (-TCTT)) have been characterized population since 1992 (Brown et al, 1992). Recently, eighteen different β-thalassaemia mutations were characterized, out of which 12 different mutations were totally new to Myanmar and three mutations were also new to South East Asia region and thus new to other parts of the world where thalassaemia is endemic. Novel genotype for either Hb E-β thalassaemia or β-thalassaemia major has also been discovered in Myanmar population. (Ne Win et al, 2002; Ne Win, 2002). Similar genotypes are observed in Sri Linka although their commonest mutation is IVS I-5 G > C which is 3rd commonest B mutation in Thailand and Myanmar (Thematic Group Meeting on Management and Prevention of Thalassemia 7-8 August 2014, New Delhi, India). Compared to other SEA countries, B Mutation in Myanmar is mostly similar to the pattern of mutation in Thailand.
Profile of thalassaemia in Yangon Children’s Hospital (YCH)

Types of thalassaemia

YCH has dedicated Thalassaemia Day Care Room (DCR) since 1970. Diagnosis of thalassaemia in Myanmar is mainly done by cellulose acetate method of hemoglobin electrophoresis, although HPLC and IEF are readily available in Department of Medical Research. Alpha thalassaemia was usually detected by the presence of H inclusion body.

According to the Thalassaemia DCR registry, up to end of 2014, 2334 patients have been registered. Among them data can be analyzed for 1134 patients. It was found that 46% (524) are E Beta thalassaemia, 22% (248) are B thalassaemia major, 17% (193) are not of established type of thalassaemia, 7% (79) are Hb E disease, 6% (65) have Hb H disease, Alpha thalassaemia, 2% (25) are B thalassaemia minor.

Compared to the data from Bangladesh, where E B thalassaemia is the 2nd most commonest after B thalassaemia minor; in YCH, B thalassaemia minor cases may not attend as they may not have symptoms; so E B thalassaemia becomes the commonest type in Myanmar Children. In Thailand E B thalassaemia is the 2nd commonest after Hb H disease. Again, in Myanmar Hb H may be under-diagnosed unless population screening was done. (WHO, SEARO, Group Meeting on Management and Prevention of Thalassemia 7-8 August 2014, New Delhi, India)

Age distribution

Among the patients in YCH, 1-5 years group is the largest, 46% of total attendees followed by 5-12 years group 41%, followed by under 1 year is 13% which is comparable to the data from the thalassaemia center in Jakarta, where they also have the largest populations of thalassaemia in 5 - 13 years age group. Also in Sri Linka where the biggest group is 5 - 9 years and 15 to 19 years, majority of thalassaemia major and intermedia presented in childhood. (WHO, SEARO, Group Meeting on Management and Prevention of Thalassemia 7-8 August 2014, New Delhi, India)
Approximately 100 to 150 new cases of thalassaemia are registered in YCH yearly which is 7-8% of estimated thalassaemic syndromes birth per year according to TIF (Thalassaemia International federation 2013).

Management

When to start transfusion

In DCR, the newly diagnosed patients are usually monitored over a period of time as shown in the following flow diagram. Usually anaemia threshold is kept at < 7 gm/dl (Haemoglobin on two occasions consecutively).

![Decision to initiate lifelong transfusion](image)

The available transfusion therapy in YCH include packed red cell, matched for patient’s ABO and Rh (D) Ag, < 4 weeks usually. If alloantibody is suspected then extended matching for other Ag (C,c,E,e,kell) in National Blood Center can be done. Although red cell phenotype is available in National Blood Center in presence of complication, full red cell phenotype is not accessible for all the newly diagnosed patients. Neither pre storage leucocyte depletion, bed side leucocyte depletion nor irradiated blood is available in our setting.

Splenectomy

Splenectomy is done in 15 to 30 patients per year for the patients with transfusion requirement 150 - 200 ml/kg/yr, Age > 5 years, symptomatic splenomegaly and hypersplenism leading cytopenia. All the patients undergoing splenectomy will receive at least HiB and Pneumococcal vaccine.
given 3 - 4 weeks prior to operation. Post-operatively oral antibiotic (Penicillin V prophylaxis) thromboprophylaxis: peri-operatively in patients with thrombocytosis (oral low dose aspirin is) prescribed. Annual influenza vaccination for all post splenectomy patients is encouraged.

Current guideline is suggested as follows: (TIF 2013). Splenectomy is less commonly required nowadays in thalassemia major children who have been transfused appropriately since early childhood; however, it is sometimes still indicated. There is also an increased chance of the adverse effects of splenectomy on blood coagulation, pulmonary hypertension and infection. In general, splenectomy should be avoided unless absolutely indicated. However, in our setting, splenectomy is still indicated as most of our patients do not have appropriate transfusion and timely chelating therapy, ending up with massive splenomegaly.

**Iron Overload assessment and chelation therapy**

In our practice, serum ferritin is usually checked for those have had > 20 units of PRBC transfusion and every 6 months. Oral chelation therapy is started when serum ferritin > 2000 ng/dl in view of likelihood of swinging to iron deficiency anemia, and non-affordability of the patients. (Although current guidelines indicate chelation once serum ferritin level reaches more than 1000 ng/dl).

While patient is on oral chelation (Deferiprone), full blood count (FBC) is checked especially if the patient is febrile, or having sore throat. Patients are informed of the possible side effects and patient alert card is also issued. Serum ferritin level is monitored 6 monthly. MRI T2* and Superconducting Quantum Interference Device (SQUID) are not available in Myanmar yet.

Arthropathy was observed in about 25% of our patients while on deferiprone, sometimes requiring to withhold the chelation or reduce the dose. Majority of them are reversible although a few cases have developed significant joint deformity. Nausea is also a common side effect of Kelfer. Patients also complained of the taste of the drug which sometimes cannot be tolerated.

Although deferoxamine (Desferal) is available, it is even more expensive and compliance is usually poor. Deferasirox (Exjade) availability is still limited in Myanmar.

**Transfusion transmitted infection (TTI)**

Retrovirus, hepatitis B and hepatitis C infection are annually checked for all follow up patients. Okada et al (2000) reported the high prevalence of Hepatitis C virus (HCV) in patients with thalassaemia in Myanmar. The incidence of HCV infection among blood recipients at the Haematology Department of the Yangon General Hospital and at the Yangon Children’s Hospital was found to be 55.5% and 46.7% respectively, which is comparable to the incidence of hepatitis B infection (66.7% and 46.7%, respectively) among these patients.

After 2000, screening for all blood donors are routinely done and up to now a total of 12 transfusion dependent patients are hepatitis B infected and 8 patients are hepatitis C infected, (over the
last 15 years). The incidence of TTI is dramatically reduced after routine screening for all blood donors.

**Cardiac assessment**

Cardiac assessment was done for those who become symptomatic. Routine cardiac assessment available are ECG, CXR and echocardiography. Paediatric cardiologist is referred when indicated.

**Alloantibodies and auto antibodies detection**

5% of our transfusion dependent thalassaemia in YCH are found to have alloantibody formation. Of these 5 (100%) are found to have anti-E and 3 (60%) had both anti-c and anti-E. (Khin Soe Soe 2005). Currently we have found a patient with anti-M and anti-Le$^a$.

Our finding was comparable with the study done in Yangon General Hospital. Anti-E is the most common alloantibody detected in Myanmar followed by anti-c. In their study Red cell alloantibodies were identified in nine patients (8.9%). Altogether six types of alloantibodies were identified: anti-E, anti-c, anti-Jk$^a$, anti-Jk$^b$, anti-Le$^a$, and anti-M. Out of nine patients, anti-E was identified in 7 (77%) and anti-c was identified in 5 (55%).

Autoantibodies were also detected in 9 girls with Thalassaemia presenting with shortened transfusion interval in YCH. A study in Jakatar center found 19% had alloantibody formation and 2.4% had auto anibody formation among 162 patients with shortened transfusion interval. (Thematic Group Meeting on Management and Prevention of Thalassemia 7-8 August 2014, New Delhi, India)

For those with alloantibodies, extended matched blood can be available from National Blood Center and for those with AIHA, immunosuppressant therapy is prescribed.

**Screening for hepatic dysfunction**

Every six months, bilirubin, AST (SGOT), ALT (SGPT) and alkaline phosphatase are checked. Pediatric hepatologist is also consulted when the liver function is impaired. The combination of hepatitis and iron overload increases the risk of liver damage. Rapid removal of iron and treatment of viral hepatitis should be considered.

**Gall bladder disease**

Up to two-thirds of thalassemia patients develop gallstones. Thalassemia intermedia patients may be at greater risk. Most of our patients remain asymptomatic and do not have cholecystitis or cholangitis. Surgical removal of gallstones is reserved for the symptomatic patient.
Growth monitoring

Weight measurement is done in every visit and height measurement is done in every 6 months. Bone age is assessed when there is concern and hormonal study is not available locally.

Diabetes Mellitus

Two hour post prandial sugar (2HPP) is baseline screen for all transfusion dependent patients. OGTT is proceeded if indicated.

Up to now, diabetes mellitus was diagnosed in 4 girls with transfusion dependent Thal major 2 expired with uncontrolled disease. 1 presented with DKA in 2014, now insulin dependent and diabetes is well controlled.

Hypothyroidism

Study of thyroid function in multitransfused children with Beta Thalassaemia Major was done in 2013 by Thandar Maung Win. Result showed 16.4% (12) of 73 patients had (compensated and decompensated) primary hypothyroidism.

We plan to start routine thyroid function test screening in 2015 for those who are above 12 years of age.

Hypoparathyroidism

From the beginning of 2015, Calcium and Phosphate level is checked 6 monthly for those above 10 years and parathormone level assessment is not available locally.

Bone disease

X-ray and MRI spine is done for those with symptoms.
DXA scan is prescribed if indicated.

Arthrogryposis due to multiple pathological fractures

Calcium and vitamin D is prophylactically prescribed for the patients of 10 years and above.

Hydroxyurea therapy

Hydroxyurea is known for increasing HbF formation.
In addition, it is also known to augment β globin gene thereby increasing β globin chains. It is one of the recommended drugs in NTDT (non transfusion dependent thalassaemia) patients. Favorable response is achieved in following set of patients:

- β thalassemia intermedia (homozygous for Xmn1 polymorphism)
- Lepore / δβ thalassemia

**Dose:** 10mg/kg/day, **Max dose:** 20mg/kg/day

**Response evaluation after 3 - 6 months of therapy**

We are prescribing Hydroxyurea for EB Thalassaemia NTDT and following up with regular FBC. Up to now we have about 100 patients on hydroxyurea and outcome will be analyzed in the future.

**Current researches**

In 2014, we have started collaboration with Cure 2 Children foundation (C2C) Italy and looking forward to allogenic bone marrow transplantation in the future. Currently our patients (250) have been registered online [http://www.bmtplus.com](http://www.bmtplus.com) and HLA typing will be done for those eligible for the allogenic transplant.

South East Asia is the locality with the most complex thalassaemia genotypes. Myanmar is one of them. In 2014, the study on phenotypic and genotypic characterization of thalassaemia syndrome in YCH has been conducted by collaboration with Mahidol University, Thailand. It is hoped that the exploration of genotypes in our people will help future prevention and genetic counseling of thalassaemia not only in Myanmar but also in South East Asia region.

**Recommendation**

A dedicated thalassaemia center in Myanmar is required for both children and adults at least in the hospitals affiliated to Universities of Medicine, or all the state and divisional hospitals in order to provide comprehensive care for patients with thalassaemia, to conduct research, to study the epidemiology and to develop the National Thalassaemia Registry.

All the newly diagnosed thalassaemia patients to have red cell phenotyping in National blood center so as to diminish the alloantibody formation.

To emphasize on bone care of thalassaemic patients which can be done in our setting.

**Conclusion**

In conclusion, by collaboration with the countries in the SEA region, we hope management of thalassaemia in Myanmar can extend up to sibling screening, genetic counseling and thalassaemia control programme for the preventive aspect as well as bone marrow transplant for the curative aspect of thalassaemia in the future.
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WHO SEARO, group meeting for thalassaemia management guideline (2014), New Delhi, India