

## Comparative molecular spectrum of beta-thalassaemia mutations in Myanmar with neighbouring countries

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### Introduction

Myanmar is composed of 135 ethnic groups in which Kachin, Kayah, Kayin, Chin, Mon, Bamar, Rakhine and Shan are major indigenous races.  $\beta$ -thalassaemia is endemic in all countries of the Southeast Asia region, probably due to the presence of malaria endemicity previously. The frequency of  $\beta$ -thalassaemia carriers had been known since 1968 and reportedly varies from 0.8% to 1.49%<sup>1</sup>. The first report on the molecular basis of  $\beta$ -thalassaemia was published in 1992 where migrant population in Myanmar-Thai border were involved<sup>2</sup>. The second molecular study was conducted by the year 2002 where transfusion receiving patients, both adult and children, were involved as study population<sup>3, 4, 10-12</sup>. First study could describe six mutations and second study reported additional twelve more new mutations and thus altogether 18 mutations. According to these two reports, the most common six mutations could be established in Myanmar in order of allele frequency as follows: IVSI-1 (G > T), CD41/42 (-TCTT), IVSI-5 (G > C), CD17 (A > T), CD35 (TAA) and IVS2 nt654 (C > T).

These six mutations covered 158/209 (78.0%) of the total  $\beta$ -alleles ( $\beta$  E globin i.e., Haemoglobin E not included). The remaining 12 mutations are not common (03 mutations) and rare (09 mutations). Many of thalassaemia genotypic research and studies in Myanmar after 2002 reports have used these six commonest mutations as an initial mutation screen to detect common mutants followed by detection of uncommon and rare mutants individually if common mutants were not detected<sup>5-9</sup>.

According to recently conducted two large scaled studies (One hospital Based<sup>7</sup> and the other community based<sup>9</sup>), no more new mutation was reported and still occurring within the molecular spectrum of previous studies<sup>3,4</sup>. A review of  $\beta$ -thalassaemia mutations types and frequencies reported in Myanmar, so far, is presented here, in an attempt to compare with those of five neighboring countries i.e., China, Laos and Thailand in the east and India and Bangladesh in the west.

### Review of the Literature

A few studies reporting the spectrum of  $\beta$ -thalassaemia mutations in different study populations in Myanmar have been published. Detailed information at clinical, haematological and molecular levels in various different study populations is available. Hospital and laboratory based studies have attempted to find out genotype - phenotype relationship among the thalassaemic victims<sup>3-7, 10-12</sup> and community based studies<sup>2, 8, 9</sup> attempted to describe genotype distribution among the carriers in the general population. All of these studies have detected first previously reported known common  $\beta$ -thalassaemia

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mutants by PCR using specific primers in those thalassaemic patients and in thalassaemia carriers who have osmotic fragility positive and normal ferritin level. Both types of studies attempted to report common types, uncommon types and rare types of mutations by finding the allele frequencies in their study populations. Such different study population constituted with different proportion of seven major ethnic groups (Kachin, Kayah, Kayin, Chin, Mon, Bamar, Rakhine, Shan) but other minor ethnic groups were not included.

The currently available genotypic data of Myanmar beta thalassaemia patients and carriers are shown in Table 1. This review attempted to compare beta thalassaemia mutation frequencies between Myanmar and five neighboring countries, China, Lao and Thailand in the east and India and Bangladesh in the west.

**Table 1. Comparison of beta thalassaemia mutation frequencies between Myanmar and neighboring countries**

Mutation	<sup>1</sup> Myanmar	<sup>2</sup> China	<sup>3</sup> Lao	<sup>4</sup> Thailand	<sup>5</sup> India	<sup>6</sup> Bangladesh
IVS1-1T	+++	+	ND	+++	+++	NR
Cd 41/42	+++	+++	+++	+++	+++	NR
IVS I-5C	+++	+	ND	+++	+++	+++
Cd17T	+++	+++	+++	+++	+	NR
Cd35TAA	++	NR	ND	++	NR	NR
IVS2-654	++	+++	ND	+++	NR	NR
nt-28G	+	+	+++	++	+	NR
Cd71/72+A	+	++	+++	+++	NR	NR
IVS1(-1)C	+	NR	ND	NR	NR	NR
619 bp del	+	NR	ND	+	+++	NR
Cd41(-C)	+	NR	ND	+	NR	NR
Cd44(-C)	+	NR	ND	NR	+	NR
Cd15(TAG)	+	NR	ND	+	++	NR
Cd15(-T)	+	NR	ND	NR	+	NR
Cd16(-C)	+	NR	ND	NR	+	NR
nt-31G	+	+	ND	NR	NR	NR
Cd5(-CT)	+	+	ND	NR	NR	NR
Cd8/9(+G)	+	NR	ND	NR	+++	NR

<sup>1</sup>(Ref: 2 - 12); <sup>2</sup>(Ref: 13, 14, 15); <sup>3</sup>(Ref: 16, 17, 18); <sup>4</sup>(Ref: 19, 20); <sup>5</sup>(Ref: 21, 22, 23, 24); <sup>6</sup>(Ref: 25);  
 +++ (Common mutant); ++ (Uncommon mutant); + (Rare mutant); NR (No Report); ND (No Data)

## Findings

A total of 18 beta thalassaemia mutations have been reported in Myanmar, reflecting the heterogeneity of its population. As expected, Myanmar has its own characteristic spectrum of mutations, with a handful of frequent mutations (four common and two uncommon) and several rare ones (12 rare). The number and frequency of different mutations varies from one study population to another, depending on the type of studies and size and ethnicities involved of the study population. While some mutations appear in most study population, others seem to be rare to a region or even to the whole country. Several observations can be made, discussed here below.

(1) The common  $\beta$ -thalassaemia mutations were detected in nearly all countries under study, albeit at different frequencies. IVS1 nt1 (G > T), the most frequent mutation in Myanmar, was also found in all neighboring countries without exception. It is found at higher frequencies in India (13.7%), whereas Thailand and China show much lower frequencies (1.7% and 0.5% respectively).

(2) Frame shift mutant Codon 41/42 (-TCTT) is an Asian-Indian origin and has high frequencies in all of the countries under study, highest in Thailand (50.9%) and China (46.7%) and second most common mutation in Myanmar (23%). It was reported 11.8% in India.

(3) The same is true for IVSI-5 (G > C), an Asian-Indian mutation, highest in India and Myanmar (15.3%) down to 5.2 % in Thailand and Codon 17 (A > T), highest in China (17.6%), Thailand (10.3%) and 9.1% in Myanmar but very rare in India. On the other hand, 619 bp deletion of Indian origin was detected as rare mutation in Myanmar (0.5%) and Thailand, none in all other countries except India (20%).

(4) Codon 35 (TAA) mutation was 5<sup>th</sup> most common at a 3.3% frequency in Myanmar similar to Thailand but no report and no data obtained from other neighbours under this review.

(5) IVS2 nt654 (C > T), 6<sup>th</sup> most common mutation of Myanmar (2.4% frequency) was found in China and Thailand with relatively at higher frequencies, highest in China (13.9%) but not detected nor reported in India, Bangladesh and Laos.

(6) IVS1 (-1) (G > C) mutation and -28 (A > G) mutation, were detected in Myanmar 1.4% each in frequency. The former is reported in none of other countries but the latter is reported in other countries at high frequencies such as Laos and Thailand (10.3%), China (11.1%), rare in India. No report in Bangladesh studies.

(7) Codon 71/72 (+A) frame shift mutation at a 1.4% frequency in Myanmar was found to be of 7.4% frequency in China and 0.8% in Thailand and Laos but none in India and Bangladesh.

(8) Codon 8/9 (+G) frame shift mutation (Asian-Indian origin) and Codon 16 (-C) frame shift mutation are rare mutants (0.5% each frequency) and was not reported from other countries except India where at a frequency of 19.6% and 1.0% respectively.

(9) Codon 44 (-C) (Kurdish origin) is rarely reported in Myanmar but none from other countries.

(10) Codon 15 (TAG) is rarely reported in Myanmar and Thailand but not commonly reported in India.

(11) nt-31 (A > G) mutant and Codon 5 (-CT) mutant are reported rarely in Myanmar, China but not reported and detected in other countries under study.

(12) Codon 15 (-T) mutation is rarely reported in Myanmar and India, not from others.

(13) Codon 41 (-C) mutation is rarely reported only in Myanmar and Thailand. No report from other neighbors yet.

(14) -88 (C > T), a mutation of Asian-Indian origin, reported in India (2% in frequency) is not found yet in Myanmar.

(15) Codon 19 (A > G) reported in Thailand (1.7%) is not detected in Myanmar. There are another two mutations reported in Thailand only, -86 (C > G) and codon 35 (+A), in low frequencies are also not reported yet in Myanmar.

## **Discussion**

The countries compared in this study encompass a wide region including East Asia, Southeast Asia and South Asia. In addition to geographic differences, each country has experienced admixtures from various populations throughout history. Furthermore, migration between these countries has been common until the present time. The heterogeneity of the Myanmar people is reflected in the 18  $\beta$ -thalassaemia mutations detected. These mutations are mainly of Asian origin with a very few exception of Mediterranean origin, and, although some countries have unique mutations, no specific mutation seems to be confined to these countries.

The most widespread and common mutations are presumably not the oldest. This is true of IVS1-1 (G > T), IVS1-5 (G > C), Codon 41/42 (-TCTT) and Codon 17 (A > T), former three are believed to be of Asian-Indian origin and the latter one is Chinese origin, not of Roman (Mediterranean) origin. However, IVS1-1 (G > T) mutation reaches a highest frequency in Myanmar, a fact that may be explained by gene flow and founder effect. Similarly, IVS1-5 (G > C), which is believed to have arisen in India, reaches its highest frequencies in the Southeast Asian countries like Thailand and Myanmar and in South Asia like Bangladesh and rare in China; Codon 41/42 (-TCTT) which was found highest in all neighbouring countries but no report from Bangladesh; Codon 17 (A > T) mutation, believed to be Chinese origin was detected highest in Myanmar, Laos, and Thailand and rare in India. Such a pattern of mutations may have been introduced to other countries by a variety of settlers and migration from its origin.

In general, the distribution of the other mutations reflects the geographical location of each country. In each country, the spectrum of mutations could be well explained by looking at its geographical location and its individual history of wars, invasions, migrations, and settlements.

Mutations of Asian-Indian origin tend to be found at higher frequencies around Southeast Asian countries like Myanmar, South Asian countries like Bangladesh whereas Chinese mutations tend to be clustered around countries closer to it like Laos, Thailand and Myanmar. However, population migration may explain the flow of some of these mutations towards other distant countries.

Recent thalassemia genotypic studies in Myanmar<sup>(5-10)</sup> also could have described almost similar pattern of the distribution of mutation frequencies as previously reported, namely common mutations are common and rare mutations are rare, although exact percentage of frequency were different which may be due to differences in size, type and demographic characteristics of study population. This review could also describe that there is no genetic epidemiological changes of thalassaemia in Myanmar due to population migration and settlement from neighboring countries, at least for the past 30 years (1990 to 2020).

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