Prevalence of β-thalassaemia trait and Haemoglobin E disorders among students aged between 14-17 yrs in Kurunegala district, Sri Lanka

Rohinie Warushahennadi¹, Dhanuja H Wanasundara³, Wasantha Priyadarshani³, Asha M Abeysekara³, Mohamed N Jiffry²

(Index words: Thalassaemia, Prevalence, Kurunegala district)

Abstract

Introduction Knowledge on frequency of carrier status of thalassaemia in a country is very important to form a plan of reducing the disease burden. In Sri Lanka, the amount of research done regarding the topic is insufficient considering the amount of thalassaemia carriers who are being detected island wide, which is increasing in numbers annually. Kurunegala is one of the districts in Sri Lanka where thalassaemia is prevalent.

Objectives To determine the prevalence of and factors associated with β-thalassaemia trait and Hb E thalassaemia among school children aged 14-17 years in Kurunegala district.

Methodology Descriptive cross sectional study. Using probability proportional to size sampling technique, 55 clusters of 30 students in the age range 14-17 years each, were selected from all the schools in Kurunegala district. Within each school, the required number of children were selected randomly.

Results Out of the participants (n=1821), 5.7% (104) were β-thalassaemia carriers and 1.2% (21) were Hb E carriers.

Conclusion The results of this study provided the true burden of β-thalassaemia trait and Hb E thalassemia in Kurunegala district.

The study also revealed the distribution of β-thalassaemia trait and Hb E disorders within the district is not even. The frequency of thalassaemia showed a significant difference across ethnic groups in the district.

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¹Consultant Haematologist, National Thalassaemia Centre, Teaching Hospital, Kurunegala, ²Consultant Paediatrician, National Thalassaemia Centre, Teaching Hospital, Kurunegala, ³Medical Officer, National Thalassaemia Centre, Teaching Hospital, Kurunegala, Sri Lanka.

Correspondence: RW, e-mail: <rgodakandage@yahoo.com>. Received 13 December 2019 and revised version 03 March 2020 accepted 02 July 2020.

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β thalassaemia trait and E thalassaemia and the factors associated with β thalassaemia trait and E thalassaemia among school children aged 14-17 years in Kurunegala district, Sri Lanka.

Methodology

Year of study: 2017-2018.

Study design: Descriptive cross sectional study.

Study setting: Kurunegala district, the capital city of North Western Province, Sri Lanka.

Study population: Children within 14-17 yrs age group studying in schools of Kurunegala district. There were 732 schools in Kurunegala district. The total number of school children within 14-17 yrs age group is 69,665 (statistics of Pro vincial Education office, Kurunegala-2015). Total population of Kurunegala district is 1.7 million (Census 2012).

Sample size determination: Since the expected proportion is not known, in order to arrive at the maximum sample size, it was taken as 50%. When alpha error = 5%, precision of the estimate =5%, cluster size =30, intra-cluster correlation coefficient =0.1, the design effect =3.9, and the non-response rate =10%, the required minimum sample size amounts to 1648.

Sampling technique: Using probability proportional to size sampling technique, 55 clusters of 30 students in the age range 14-17 years each, were selected from all the schools in Kurunegala district. 54 schools were sampled (Two clusters from one school and one cluster from 53 schools). Within each school, the required number of children were selected randomly. If there were less than 30 students in the age range, 14-17 years in a particular school, all the children in the required age group were included in the study.

Inclusion and exclusion criteria: School children within the age group 14-17 yrs studying and residing in Kurunegala district were included into the study. School children within the selected age group, studying in Kurunegala district but not residing there were excluded.

Study instruments:

1. Self-administered questionnaire to get the demographic data.
2. Three ml of venous blood; (2cc EDTA sample and 1cc clotted blood) for Complete Blood Count (CBC) and Hb electrophoresis (EDTA sample) and Serum ferritin (clotted blood sample).

Data collection method

Participation in this study was voluntary.

The self-administered questionnaire was administered on the participants to obtain demographic data. 3ml of venous blood was drawn by qualified Nursing Officers, following universal precautions.

Blood samples were analyzed for CBC and Hb electrophoresis at Thalassaemia Centre, Kurunegala with fully automated hematology analyzer (Sysmex) and Capillary electrophoresis analyzer (Sebia) respectively.

These analyzers undergo quality control procedures to maintain the accuracy and precision of the results.

Serum ferritin was done for samples with low Hb and red cell indices to assess body iron status which may influence the diagnosis of β thalassaemia trait. For participants with low serum ferritin with marginally elevated HbA2, oral iron was prescribed and Hb electrophoresis was repeated after a course of oral iron. Serum ferritin was done at Chemical Pathology Department, Teaching Hospital, Kurunegala with Immulite 1000 machine.

Data analysis

Sample characteristics were analyzed using descriptive statistical methods, and presented as percentages or proportions depending on their scales of measurements.

Ethical issues

The protocol was approved by the Ethical Review Committee, Teaching Hospital, Kurunegala.

Permission for the study was obtained from the Provincial Director of Education, North Western Province. Zonal Directors of Education were informed about the schools selected for the research. Permission was obtained from all the Principals of selected schools. Written consent was taken from parents of the students and the students as well after reading out the information sheet and answering participants’ questions by the researcher. A witness (a teacher/principal) confirmed that the individual has given the consent freely.

Result

A total of 1839 students aged between 14-17 yrs were initially selected for the present study. They were from 54 schools situated within Kurunegala district 2 clusters × (1school + 1 cluster × 53 schools). Of those, 1821 students were eligible to participate in the present study. 18 students were not eligible for the study as they did not reside in Kurunegala.

The study group was predominantly female (59.9%). Of the participants, 57.4% were 14-15 yrs old. Most of the participants were Sinhalese (93.6%), while 5.8% were Muslim and 0.4% were Tamil and 0.2% were other ethnicities. In Kurunegala, main ethnic group is Sinhalese (91.9%). There are 6.5% of Muslims and 1.4% of Tamils (Census 2012).
In the sample, higher participation was noted from schools situated in Kurunegala (22.1%), Kuliyapitiya west (7.7%), Mawathagama (7.6%) and Ibbagamuwa (7.5%) Educational Divisions. When these figures were compared with the statistics of Education Department, Kurunegala district, highest number of students within 14-17 yrs age were noted in schools situated in same educational divisions [Kurunegala (22.1%), Ibbagamuwa (7.2%), Kuliyapitiya west (6.1%) and Mawathagama (6.4%)].

When the sample was analyzed according to the residence of participants, highest participation was from Rideegama (9.1%) and Kurunegala (8.9%) MOH areas. Lowest percentage of participants were from Ambanpola (0.5%) and Maspotha (0.9%) MOH areas.

In Kurunegala district, highest population was seen in Pannala (7.68%), Panduwasnuwara (5.94%), Rideegama (5.48%) MOH areas. Lowest population was seen in Ambanpola (1.41%) MOH area.

Prevalence of thalassaemia carrier state

Great majority of participants (83.7%) showed normal results in both CBC and Hb capillary electrophoresis (CE) (Table 1).

<table>
<thead>
<tr>
<th>Diagnosis</th>
<th>No</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal</td>
<td>1524</td>
<td>83.7</td>
</tr>
<tr>
<td>Iron deficiency</td>
<td>29</td>
<td>1.5</td>
</tr>
<tr>
<td>β trait (Heterozygous β thalassaemia)</td>
<td>104</td>
<td>5.7</td>
</tr>
<tr>
<td>Hb E trait (Heterozygous Hb E)</td>
<td>21</td>
<td>1.2</td>
</tr>
<tr>
<td>Hb D trait (Heterozygous Hb D)</td>
<td>10</td>
<td>0.5</td>
</tr>
<tr>
<td>Hb S trait (Heterozygous Hb S)</td>
<td>1</td>
<td>0.1</td>
</tr>
<tr>
<td>Delta β trait (Heterozygous Delta β Thalassaemia)</td>
<td>2</td>
<td>0.1</td>
</tr>
<tr>
<td>Possible Hb Constant Spring trait</td>
<td>1</td>
<td>0.1</td>
</tr>
<tr>
<td>Possible α thalassaemia trait</td>
<td>129</td>
<td>7.1</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>1821</strong></td>
<td><strong>100.0</strong></td>
</tr>
</tbody>
</table>

Out of the participants, 5.7% (95% CI-4.65-6.78) showed CBC and Hb electrophoresis results suggestive of β thalassaemia trait. β thalassaemia trait was identified by quantifying Hb A2 level in the absence of a variant Hb. Hb A2 level of >3.5% in the presence of high RBC count with low MCV, MCH were the diagnostic criteria for β thalassaemia trait.

Hb E trait was identified by relevant variant Hb band in CE. In the sample, 1.2% (95% CI-0.66-1.64) showed Hb E band in electrophoresis (Hb E trait).

There were few other haemoglobinopathies identified in the study sample. There were 10 participants (0.5%) with Hb D trait (Heterozygous Hb D). One participant (0.1%) with Hb S trait (Heterozygous Hb S). Two participants (0.1%) showed low red cell indices with high Hb F band in electrophoresis, suggestive of Delta β Thalassaemia trait (Heterozygous Delta β thalassaemia).

One participant (0.1%) showed a small Hb Constant Spring band suggestive of Hb Constant Spring trait which needs to be confirmed with molecular studies.

There were 129 (7.1%) participants showed low red cell indices with normal electrophoresis pattern, suggestive of α thalassaemia trait which needs to be confirmed with molecular studies.

There were 29 participants (1.5%) with Iron deficiency (Low serum ferritin).

Factors associated with β thalassaemia trait

Out of the 104 participants with β thalassaemia trait, 59.6% were in the age group of 14-15y.

Majority of β thalassaemia trait (62.9%) were females.

Great majority (85.6%) of β thalassaemia trait participants were Sinhalese while 14.4% were Muslims. There were no Tamil participants with β thalassaemia trait. (There were only 8 Tamil participants in the study sample).

In the sample, 5.2% of the Sinhalese participants were β thalassaemia trait positive while 14.1% of Muslim participants were β thalassaemia trait positive. This difference was statistically significant (p<0.001) (Figure 1).

![Figure 1. Distribution of participants with β thalassaemia trait and others by ethnicity.](image-url)
Highest prevalence of \( \beta \) thalassaemia trait was from Galgamuwa (0.12%), Narammala (0.11%), Polgahawela (0.11%) MOH areas. With regards to MOH areas of residence, the prevalence of \( \beta \) thalassaemia trait was not statistically significant between areas.

### Factors associated with Hb E trait

Out of the 21 participants with Hb E trait, 11 (52.4%) were males. 19 participants (90.5%) with Hb E trait were Sinhalese while 2 (9.5%) were Muslims.

Highest prevalence of Hb E thalassaemia trait was from schools situated in Kobeigane (3.0%), Kotawehera (2.9%), Giribawa (2.9%), Galgamuwa (2.9%) educational divisions.

There was no statistically significant difference regarding the prevalence of Hb E thalassaemia trait between Educational divisions.

### Discussion

Thalassaemia was first identified in Sri Lanka in 1951[27]. Since then there have been reports of further cases and the occurrence of HbE and HbE/\( \beta \) thalassaemia in the population.
Knowledge on the true frequencies of thalassaemia patients and carriers in the country is still limited. In a recent nationwide survey on hospital based thalassaemia patients, 1774 patients with thalassaemia were identified in 23 different centers in the country. Out of them, 68.7% had β thalassaemia major and 20.3% had Hb E/β thalassaemia. Majority (755) were from Teaching Hospital, Kurunegala [22].

In the initial studies done in Sri Lanka, the carrier rates were described as α thalassaemia (+), β thalassaemia (2.2%) and HbE (0.5%) [6]. In another early study directed mainly at the different forms of severe thalassaemia and their molecular basis in Sri Lanka suggested that the thalassaemia mutations were unevenly spread. Although 23 different β-thalassaemia mutations were found, three accounted for the thalassaemia phenotype in about 70% of the patients. Overall, 15.5% patients were carriers for deletion forms of α+ thalassaemia. The study also included a small survey of school children which showed the highest frequency of thalassaemia carriers in the Kurunegala district [4].

In a recent islandwide study done among school children, the prevalence of different types of thalassaemias were determined in all the districts of the country by sampling 3 schools in one district. In that study, prevalence of β thalassaemia trait in Kurunegala district was 4.33% and Hb E trait was 1.67%. Prevalence of β thalassaemia trait and Hb E trait for the country was 2.0% and 0.5% [23, 15].

In a survey done among university students of Sri Jayawardhanapura, the carrier rate for β thalassaemia was found as 5.5% which is higher than the figures of previous studies in Sri Lanka [9].

The present study revealed the prevalence of β thalassaemia trait and Hb E trait in Kurunegala district as 5.7% and 1.2% respectively. Our findings are similar to the findings of Premawardhana et al done in 2016. The number of studies done on frequency of thalassaemia carriers in Kurunegala district is limited. Therefore it is difficult to comment on the trend of β thalassaemia trait in Kurunegala district. Evaluation of frequencies of thalassaemia carriers in the district in every 5 yrs may be necessary.

Since 2005/2006 a national thalassaemia program to screen individuals and increase awareness with the goal of reducing new births of children with severe thalassemia has been in place. Whether these efforts have impacted on numbers of births of severe thalassemia needs evaluation. Premawardhana et al (2019) evaluated the number of births of patients with β-thalassaemia from 1996 to 2014, and found the number of new births, between 45-55 per year, had remained fairly constant.

In the present study, there was a significant difference in frequencies of β and E thalassaemia in Muslims compared to the others (p<0.01). In contrast, Premawardhana et al (2016) found that there was no difference in the gene frequencies for the common haemoglobin variants between Sinhalese and Tamil populations.

This is the first study which analyzed the frequencies of thalassaemia trait in Educational Divisions and MOH areas within Kurunegala district. The frequency of β thalassaemia trait in Galgamuwa Educational Division was high compared to the other divisions (p<0.05). These findings show to whom and where to give more attention when screening.

In conclusion, the results of this study provides the true burden of β thalassaemia trait (5.7%) and Hb E thalassaemia trait (1.2%) in Kurunegala district.

The study has revealed the distribution of β thalassaemia trait and Hb E thalassaemia within the district is not even. The prevalence of β thalassaemia trait varied from 0.0% to 13.2% and prevalence of Hb E thalassaemia trait varied from 0.0% to 3.0% among the Educational divisions within the Kurunegala district.

The study has provided strong evidence on significant differences of frequency of thalassaemia in Muslims compared to the other ethnic groups of Kurunegala.

These findings will be important for the future planning in prevention of thalassaemia in Kurunegala district.

Acknowledgements

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Conflicts of interest

There are no conflicts of interest.

References


