Impact of genotype on endocrinal complications of Children with Alpha-thalassemia in China

Hong-Cheng Luo, Qi-Sheng Luo, Fu-Gao Huang, Chun-Fang Wang & Ye-Sheng Wei

Alpha-thalassemia occurs with high frequency in China. Four common α-globin gene deletion mutations (–SEA, –α3.7, and –α4.2, Haemoglobin Constant Spring (CS) mutation) were identified in Chinese patients. Individuals with alpha-thalassemia syndrome are more often of children. However, report on endocrinal complications in children with alpha thalassemia in China are still absent. The present study aimed to investigate the impact of genotype on endocrinal complications in Chinese children. Association analysis between genotype and endocrinal complication development was conducted on 200 patients with 200 healthy controls. Hypogonadism was found to be the most prominent endocrinal complications (84.0%) leading to the growth retardation, hypogonadism, diabetes mellitus, hypothyroidism and hypoparathyroidism whose incidence were significantly higher in patients. (αCSα/–SEA) was the main genotype of Alpha thalassemia identified in the patients (37.5%), and patients with the (–α4.2/–SEA) genotype had a higher prevalence of hypogonadism, diabetes mellitus and hypoparathyroidism (P = 0.001, P = 0.001, P < 0.001, respectively).

Materials and Methods

General. Two hundred Children (126 males and 74 females) with mean age of 9.64 ± 1.15 years (range, 3–12 years). Who were registered in The Affiliated Hospital of Youjiang Medical College for Nationalities from the period January 2010 to June 2016 were included in this research. α-thalassemia children were characterized with one of the genotype of SEA, –α3.7/–SEA, –α4.2/–SEA or αCSα/–SEA. Which was identified by the DNA sequencing technique. The basic clinical information collected included Average Hematological Parameters of diagnosis, gender, age, age of start transfusion, age of start chelation, frequency of transfusion and related compliance.

A-thalassemia is a serious health problem worldwide, especially in Mediterranean areas, Southeast Asia and Southern China. Guangxi Province is located in the southwest of China where the incidence of thalassemia is 24.51%. However, in the past decades, data on diagnose and treatment of α-thalassemia or related complications in children are still absent. In this study typical physical exam findings growth retardation, hypogonadism, thalassemic bone deformities, diabetes mellitus were included to identify the association between four genotype (SEA, –α3.7/–SEA, –α4.2/–SEA, αCSα/–SEA) and endocrine complications in children with α-thalassemia.

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Normal levels serum ferritin, no one suffering from hemolytic anemia and malnutrition anemia. No cardiovascular and blood infectious disease. Its family without hypertension, diabetes. All the control group also were diagnosed by the DNA sequencing technique, no one suffers from six common α-thalassemia (–SEA, -α3.7, -α4.2, αCS, αeM/N, αCAMP, IntM), which the common type of thalassemia in chinese people.

Physical examination including. Red blood cells (RBC), Hemoglobin (HGBg/l), Mean corpuscular volume (MCV/pg), Mean hemoglobin content (MCH/pg), Mean hemoglobin concentration (MCHC), basal growth hormone, estradiol (in females) and testosterone (in males), thyroid-stimulating hormone (TSH), FT3, FT4, serum calcium concentration, serum phosphate and parathyroid hormone. Alpha globin mutations were analyzed using gap-PCR and reverse-hybridization assay according to the manufacturer.

Classification of patients according to genotype. Children were divided into four groups according to their genotype based on the α-globin gene production. Group1–4: (SEA) deletions, (−3.7 kb merge SEA) deletions, (−4.2 kb merge SEA) deletions and (CS mutations merge SEA) deletions.

Definitions. Short stature was defined as patient height ≥2 standard deviation below the mean for age, gender and ethnicity. Short stature was evaluated by Children’s Health Rehabilitation Center (Affiliated Hospital of Youjiang Medical College for Nationalities, Guangxi, China).

Hypogonadism was defined as low testosterone (in males or oestradiol (in females) level or subjects who had received testosterone or oestradiol therapy.

Patients were diagnosed with diabetes mellitus based on WHO criteria or history of insulin therapy or oral antidiabetic therapy according to American Diabetes Association, World Health Organization Criteria and National Diabetes Health Group 1979.

Hypothyroidism was defined according to TSH/FT3, FT4 or based on the history of treatment with levothyroxine for previously diagnosed hypothyroidism. Hypoparathyroidism was defined as low serum calcium and low serum parathyroid hormone concentration, with increased serum phosphate.

A hemoglobin level of less than 90 (g/L) was the standard for initiating transfusion in children with severe thalassemia. Infection, growth retardation, diabetes mellitus, hypogonadism, hypothyroidism, hypoparathyroidism or other complications in thalassemia children, were the indications for transfusion at a relatively high level of haemoglobin.

Statistical analysis. SPSS13.0 (SPSS, Inc., IL, USA) was used to conduct statistical analysis. χ² test or Fisher’s exact test was used for comparison between different groups. Measurement data were represented as mean ± standard deviation (X ± s), and categorical data were represented as χ². P < 0.05 and P < 0.001 were considered to indicate statistically significant differences.

Results

Patient characteristics. All the patients were recruited from Affiliated Hospital of Youjiang Medical College for Nationalities, Guangxi, China. The patients (126 males and 74 females) had a mean (SD) age of (9.64 ± 1.15) years. Hypogonadism was the most prominent endocrinical complications in patients (84.0%), followed by growth retardation (68.5%) and hypoparathyroidism (14.5%). A total 70.5% of patients start to use chelation in 3 years old. There was no significant difference in RBC, MCV, MCH and MCHC among the four groups (P > 0.05). Clinical Average Hematological Parameters were summarized in (Table 1).

Genotype of Thalassemia and endocrinical complications. Two major genotype identified in the Alpha thalassemia patients were αCS/− SEA (37.5%, 38.1% of males and 36.5% of females) and SEA (32.5%), followed by -α3.7/−SEA (18.5%) and -α4.2/−SEA (11.5%). A total of 94.1% of patients with the αCS/−SEA genotype started earlier transfusion (≤3 year), 77.3% of patients received frequent transfusion (every 4–5 weeks) and 68.0% started earlier iron chelators (>3 years). In addition, patients with the αCS/−SEA genotype had a higher

### Table 1. Clinical Average Hematological Parameters of the Study Population (X ± s). RBC, red blood cell; HGB, haemoglobin; MCV, mean corpuscular volume; MCH, mean corpuscular haemoglobin; MCHC, mean corpuscular concentration; SEA: the Southeast Asian deletion; -α3.7, rightward deletion; -α4.2, leftward deletion; CS, Hb Constant Spring. a:compared with -α3.7/–SEA; CS, p < 0.001; b:compared with -α3.7/–SEA, P < 0.001. Note: RBC, HGB, MCV, MCH and MCHC were the average hematological parameters level before the first blood transfusion.

| Genotype     | n  | RBC (x10ⁱ²/L) | HGB (g/L) | MCV (fl) | MCH (pg) | MCHC (g/L) | Serum ferritin (ng/ml) |
|--------------|----|--------------|----------|----------|----------|------------|-----------------------|
| SEA          | 65 | 5.21 ± 1.08  | 85.69 ± 23.44 | 62.43 ± 7.31 | 19.11 ± 2.29 | 305.11 ± 14.70 | 356.17 ± 25.76 |
| -α3.7/–SEA   | 37 | 5.09 ± 0.82  | 84.43 ± 15.63 | 56.28 ± 6.56 | 16.70 ± 1.67 | 299.61 ± 21.01 | 976.58 ± 79.11 |
| -α4.2/–SEA   | 23 | 5.32 ± 0.85  | 80.88 ± 14.67 | 52.49 ± 2.44 | 16.74 ± 2.90 | 294.50 ± 15.02 | 997.37 ± 78.69 |
| αCS/–SEA     | 75 | 4.11 ± 0.96  | 79.17 ± 18.89 | 68.63 ± 8.38a | 18.64 ± 2.03 | 270.00 ± 25.01 | 1023.69 ± 81.55 |
| Reference    | 350–550 | 110.00–160.00 | 80.00–100.00 | 27.00–34.00 | 320.00–360.00 | 15.00–250.00 |
prevalence of growth retardation (92%), and patients with the −α4.2/−SEA genotype had a higher prevalence of hypogonadism and diabetes mellitus (100% and 73.9%, respectively) (Table 2).

### Growth retardation in patients

Growth retardation was identified in 75.2% of patients (≥6 years old) and 61.1% of patients (<6 years old), and no significant difference was identified between males and females. A total of 40.9% of patients with growth retardation started earlier blood transfusion (≤3 years), 69.3% received frequent transfusion (every 4–5 weeks), 89.8% started iron chelation (>3 years) and 17.5% were poor compliant (Table 3).

### Hypogonadism in patients

Hypogonadism was identified in 83.8% of patients (≥6 years old) and 84.2% of patients (<6 years old), and there was significant difference between males and females (P < 0.001). A total of 67.3% of patients with hypogonadism started earlier transfusion (≤3 years), 51.8% of them received frequent transfusion (every 2–3 weeks), 76.2% of patients with hypogonadism started iron chelation (>3 years) and 12.5% had a poor compliance (Table 4).

### Diabetes mellitus in patients

Diabetes mellitus was identified in 14 patients and 71.4% of them were ≥6 years old with no significant difference identified between males and females. 92.9% of them received frequent transfusion (every 2–3 weeks), and 85.7% of patients with hypogonadism started iron chelation (>3 years) (Table 5).

### Hypothyroidism in patients

26 patients (15 males and 11 males) were diagnosed with hypothyroidism and no significant difference was identified between males and females. All of these patients started earlier transfusion (≤3 years). Most of the patients (88.3%) were more than 6 years older and 96.2% had a poor compliant (Table 6).

### Hypoparathyroidism in patients

Hypoparathyroidism was identified in 29 patients and 82.8% of them were ≥6 years old, no significant difference was observed between males and females. All of these patients started earlier transfusion (≤3 years) and most of them had a poor compliant (Table 7).

### Endocrine complication between case group and control group

There was no significant difference in the incidence of endocrine complication between male and female in case group and control group, alpha thalassemia patients are significantly more likely to have growth retardation, hypogonadism, diabetes mellitus, hypothyroidism, and hypoparathyroidism. 

#### Table 2. Association between patient genotype and endocrinal complications.

| Characteristics                  | Patients (n = 65) | −α3.7/−SEA (n = 37) | −α4.2/−SEA (n = 23) | αCS/−SEA (n = 75) | P-value |
|----------------------------------|-----------------|---------------------|---------------------|------------------|---------|
| **Gender**                       |                 |                     |                     |                  |         |
| Male                             | 126             | 31.7%               | 19.0%               | 11.1%            | 38.1%   | 0.98    |
| Female                           | 74              | 33.8%               | 17.6%               | 12.2%            | 36.5%   |         |
| **Age of start transfusion (years)** |               |                     |                     |                  |         |
| ≤3                               | 117             | 22.2%               | 13.7%               | 10.3%            | 53.8%   | <0.001  |
| >3                               | 83              | 47.0%               | 25.3%               | 13.3%            | 14.5%   |         |
| **Frequency of transfusion (weeks)** |             |                     |                     |                  |         |
| Every 2–3                        | 98              | 45.9%               | 19.4%               | 17.3%            | 17.3%   | <0.001  |
| Every 4–5                        | 102             | 19.6%               | 17.6%               | 5.9%             | 56.9%   |         |
| **Age of start chelation (years)** |           |                     |                     |                  |         |
| ≤6                               | 59              | 27.1%               | 15.3%               | 16.9%            | 40.7%   | 0.31    |
| >6                               | 141             | 34.8%               | 19.9%               | 9.2%             | 36.2%   |         |
| **Growth retardation**           |                 |                     |                     |                  |         |
| Negative                         | 63              | 50.8%               | 25.4%               | 14.3%            | 9.5%    | <0.001  |
| Positive                         | 137             | 24.1%               | 15.3%               | 10.2%            | 50.4%   |         |
| **Hypogonadism**                 |                 |                     |                     |                  |         |
| Negative                         | 32              | 62.5%               | 15.6%               | 0.0%             | 1.9%    | 0.001   |
| Positive                         | 168             | 26.8%               | 19.0%               | 13.7%            | 40.5%   |         |
| **Diabetes mellitus**            |                 |                     |                     |                  |         |
| Negative                         | 186             | 33.9%               | 18.3%               | 9.1%             | 38.7%   | 0.001   |
| Positive                         | 14              | 14.3%               | 21.4%               | 42.9%            | 21.4%   |         |
| **Hypothyroidism**               |                 |                     |                     |                  |         |
| Negative                         | 174             | 36.8%               | 14.4%               | 8.6%             | 40.2%   | <0.001  |
| Positive                         | 26              | 3.8%                | 46.2%               | 30.8%            | 19.2%   |         |
| **Hypoparathyroidism**           |                 |                     |                     |                  |         |
| Negative                         | 171             | 38.0%               | 16.4%               | 9.4%             | 36.3%   | <0.001  |
| Positive                         | 29              | 0.0%                | 31.0%               | 24.1%            | 44.8%   |         |
hypothyroidism and hypoparathyroidism compared with controls \((P < 0.001)\) (Table 8). The HGB level lower in patients \((81.17 ± 15.23 \text{ g/L, range, 13~95 g/L})\) than control subjects \((126.21 ± 17.65 \text{ g/L, range, 55~167 g/L})\). We also identified a significant difference between RBC and MCV indices in case group and control group \((P < 0.001)\).

### Discussion

Thalassemia is a well-known inherited hematologic disorder caused by reduced or absence of globin production\(^{11}\). In China, this disease is prevalent in areas near the southern bank of the Yangtze River, such as Guangdong, Guangxi, Fujian and Yunnan Provinces\(^{12–14}\). Endocrine dysfunction is a frequent complication in thalassemic
patients who are on regular blood transfusions. Iron overload has been considered to be the major cause of endocrine abnormalities of α-thalassemia. Growth retardation, hypogonadism, diabetes mellitus and hypoparathyroidism represent the most common endocrinopathies in thalassemic patients. In this study, we evaluate the impact of genotype on endocrinal complications of children with α-thalassemia in China and demonstrate that hypogonadism is the most frequent endocrine complication (84.0%), followed by growth retardation (68.5%) and hypoparathyroidism (14.5%).

| Characteristics | Patients, n | Diabetes mellitus (n = 200) | P-value |
|-----------------|-------------|-----------------------------|---------|
|                 | Negative (n = 186) | Positive (n = 14) |
| Gender          |             |                             |         |
| Male            | 126         | 95.2%                       | 4.8%    | 0.11 |
| Female          | 74          | 89.2%                       | 10.8%   |
| Age (years)     |             |                             |         |
| ≥6              | 105         | 90.5%                       | 9.5%    | 1.14 |
| <6              | 95          | 95.8%                       | 4.2%    |
| Frequency of transfusion (weeks) |
| Every 2–3       | 98          | 99.0%                       | 1.0%    | 0.001 |
| Every 4–5       | 102         | 87.3%                       | 12.7%   |
| Age of start transfusion (years) |
| ≤3              | 117         | 95.7%                       | 4.3%    | 0.07 |
| >3              | 83          | 89.2%                       | 10.8%   |
| Age of start chelation (years) |
| ≤6              | 59          | 96.6%                       | 3.4%    | 0.32 |
| >6              | 141         | 91.5%                       | 8.5%    |
| Compliance, %   |             |                             |         |
| <60             | 43          | 88.4%                       | 11.6%   | 0.32 |
| ≥60             | 157         | 94.3%                       | 5.7%    |

**Table 5.** Association between diabetes mellitus and each of the demographic, frequency of transfusion, age of start transfusion, Age of start chelation, compliance.

| Characteristics | Patients | Hypothyroidism (n = 200) | P-value |
|-----------------|----------|--------------------------|---------|
|                 | Negative (n = 174) | Positive (n = 26) |
| Gender          |             |                          |         |
| Male            | 126        | 88.1%                    | 11.9%   | 0.70 |
| Female          | 74         | 85.1%                    | 14.9%   |
| Age (years)     |             |                          |         |
| ≥6              | 105        | 78.1%                    | 21.9%   | <0.001 |
| <6              | 95         | 96.8%                    | 3.2%    |
| Frequency of transfusion (weeks) |
| Every 2–3       | 98         | 79.6%                    | 20.4%   | 0.004 |
| Every 4–5       | 102        | 94.1%                    | 5.9%    |
| Age of start transfusion (years) |
| ≤3              | 117        | 77.8%                    | 22.2%   | <0.001 |
| >3              | 83         | 100.0%                   | 0.0%    |
| Age of start chelation (years) |
| ≤6              | 59         | 67.8%                    | 32.2%   | <0.001 |
| >6              | 141        | 95.0%                    | 5.0%    |
| Compliance, %   |             |                          |         |
| <60             | 43         | 41.9%                    | 58.1%   | <0.001 |
| ≥60             | 157        | 99.4%                    | 0.6%    |

**Table 6.** Association between hypothyroidism and each of the demographic, frequency of transfusion, age of start transfusion, Age of start chelation, compliance.

Our survey showed that the MCV levels in group (α CS/α SEA) were higher than those in group (α 3.7/–SEA) and group (α 4.2/–SEA) (P < 0.001, P < 0.001, respectively), there were no significant differences in RBC,
HGB, MCH and MCHC levels among the four groups ($P > 0.05$), similar to the previous study by Zhu et al.\textsuperscript{16}. Compared with the other three groups ($\alpha\text{-CS}/-\text{SEA}, -\alpha 3.7/-\text{SEA}, -\alpha 4.2/-\text{SEA}$), the group SEA had a significant lower serum ferritin levels ($P < 0.001$, respectively), this may be due to patients with SEA genetype generally do not receive blood transfusion therapy frequently unless combined with iron deficiency anemia, vitamin D deficiency, infection caused by long-term malnutrition anemia. In consistent with report by Zhou Y. U. et al.\textsuperscript{17} no significant difference was observed among the three group ($\alpha\text{-CS}/-\text{SEA}, -\alpha 3.7/-\text{SEA}, -\alpha 4.2/-\text{SEA}$) in Serum ferritin levels ($P > 0.05$, respectively).

In the present study, we found that the patients with the genetype of ($\alpha\text{-CS}/-\text{SEA}$) had significant higher prevalence of growth retardation, hypogonadism ($P < 0.001$, $P = 0.001$, respectively). Just like previous report\textsuperscript{18–20} hypogonadism was identified as the most common endocrine complication in the patients (84.0%).

### Table 7. Association between hypoparathyroidism and each of the demographic, frequency of transfusion, age of start transfusion, Age of start chelation, compliance.

| Characteristics | Patients | Hypoparathyroidism | $P$-value |
|----------------|---------|-------------------|-----------|
|                |         | Negative (n = 171) | Positive (n = 29) |
| Gender         | Male    | 126               | 87.3  12.7 | 0.35 |
|                | Female  | 74                | 82.4  17.6 |     |
| Age (years)    | ≥6      | 105               | 77.1  22.9 | <0.001 |
|                | <6      | 95                | 94.7  5.3  |     |
| Frequency of transfusion (weeks) | Every 2–3 | 98 | 80.6 | 19.4 | 0.05 |
|                | Every 4–5 | 102 | 90.2 | 9.8  |     |
| Age of start transfusion (years) | ≤3 | 117 | 75.2 | 24.8 | <0.001 |
|                | >3      | 83                | 100.0 0.0 |     |
| Age of start chelation (years) | ≤6 | 59 | 69.5 | 30.5 | <0.001 |
|                | >6      | 141               | 92.2  7.8  |     |
| Compliance, %  | <60     | 43                | 39.5  60.5 | <0.001 |
|                | ≥60     | 157               | 98.1  1.9  |     |

### Table 8. Comparison of endocrine complications in patients with alpha thalassemia and control group.

| Characteristics | Alpha thalassemia (n = 200) | Control group (n = 200) | $P$-value |
|----------------|-------------------------------|-------------------------|-----------|
| Gender         | Male 126 (63.0%) 113 (56.5%) | Female 74 (37.0%) 87 (43.5%) | 0.22 |
| Growth retardation | Negative 137 (68.5%) 195 (97.5%) | Positive 63 (31.5%) 5 (2.5%) | <0.001 |
| Hypogonadism | Negative 168 (84.0%) 197 (98.5%) | Positive 132 (16.0%) 3 (1.5%) | <0.001 |
| Diabetes mellitus | Negative 186 (93.0%) 198 (99.0%) | Positive 14 (7.0%) 2 (1.0%) | 0.05 |
| Hypothyroidism | Negative 174 (87.0%) 193 (96.5%) | Positive 26 (13.0%) 7 (3.5%) | 0.001 |
| Hypoparathyroidism | Negative 171 (85.5%) 199 (99.5%) | Positive 29 (14.5%) 1 (0.5%) | <0.001 |
of start transfusion or start Chelation had a significant impact on hypogonadism development. However a lower prevalence of hypogonadism was found in some study21–23, which were mainly attributed to difference in the economic status of patients, Physicians’ strategies to optimize chelation therapy, promoting compliance, educating patients and different ethnic23–26. The patients with the genotype of (-α/-SEA) had a significantly higher prevalence of diabetes mellitus (P = 0.001). And there was no significant differences in the incidence of genotypes between males and females (P = 0.98).

Compared to the present study 68.5% of patients identified with growth retardation, Hattab, F. N. et al.27 found a higher prevalence of growth retardation (75.9%). This may be attributed to the difference in economy, most of the patients come in the latter study from poor families, received poor health care treatment, which resulted in multiple infections, thereby aggravating growth retardation or other potential endocrine complications development in Alpha-thalassemia during childhood. Futhermore, the discrepancy of clinical manifestations may be impacted by genetic and environmental factors24–30. There was significant association between growth retardation and older year (≥6 years), earlier age of start transfusion, chelation, frequency of blood transfusion or poor compliance (P = 0.03, P < 0.01, P < 0.01, P < 0.01, P = 0.04, respectively). But there was no significant association between growth retardation and gender (P = 0.83).

In the present study, 7.0% of patients were diagnosed with diabetes mellitus, similar to 8.0% in report by Ong, C. K. et al.24. Several previous study have report a lower prevalence of diabetes mellitus, which ranged from 2.5% to 4.9%32–34, while Other had report a higher prevalence of diabetes mellitus, reaching 13% to 17.0%35–37. These discrepancies can be attributed to differences in the age of patients and severity of Hepatitis C virus infection, transfusion rates and chelation therapies, male sex, liver iron concentration38,39. There was significant association between diabetes mellitus and frequency of blood transfusion (P = 0.001), but there was no significant association between diabetes mellitus and gender, age, age of start transfusion, chelation, frequency blood transfusion or compliance (P = 0.11, P = 1.14, P = 0.07, P = 0.32, P = 0.32, respectively).

Hypothyroidism was identified in 26 patients (13.0%), which was similar to the result reported by Eshragi, P. et al.40. While, other studies reported a lower prevalence of hypothyroidism, which ranged from 1.0% to 10.0%41–43. The results of different studies vary widely, these discrepancies can be attributed to differences in genotype of thalassemia, the age of patients or treatment protocols.

Hypogonadism (84.0%), growth retardation (68.5%) and hypoparathyroidism (14.5%) were the first and the most frequent endocrine complications diagnosed in our present study. Today, many patients can benefit from modern treatment, improve the quality of life of patients due to adopting in early and regular chelation therapy. Therefore, prevention of the endocrine complications may be influenced by the improvement of medical diagnosis and treatment. Monitoring compliance is essential in such conditions. There are a few limitations need to be mention here. Firstly, the sample size is small, and the age of these patients too early which may result in limited power. Secondly, the type of iron chelation used could not be figured out, rare genotype of α-thalassemia were not included in our study. Thirdly, none of the analyses take into account the age effect properly. The incomplete medical records could prevent us from identifying predictive complication. Further studies are needed on the complications of all α-thalassemic and older patients in the region.

In conclusion, our present study show that α/α–SEA, SEA, -α–SEA, -α-SEA, -α-SEA, -α.4.2–SEA, and -α-4.2–SEA are the main genotype identified in α-thalassemia children in Guangxi Province, and hypogonadism, growth retardation and hypoparathyroidism are the most common endocrine complications in children with α-thalassemia.

References

1. De Sanctis, V., Eleftheriou, A. & Malaventura, C. Prevalence of Endocrine Complications and Short Stature in Patients with Thalassaemia Major: A Multicenter Study by the Thalassaemia International Federation (TIF). Pediatr Endocrinol Rev. 2(Suppl 2), 249–255 (2004).
2. Weatherall, D. J. Thalassemia as a Global Health Problem: Recent Progress Toward its Control in the Developing Countries. Ann N Y Acad Sci. 1202, 17–23, doi:10.1111/j.1749-6632.2010.05346.x (2010).
3. Kachroo, S. S. & Winichagoon, P. Thalassemia in Southeast Asia: Problems and Strategy for Prevention and Control. Southeast Asian J Trop Med Public Health. 23, 647–655 (1992).
4. Li, B. et al. High Prevalence of Thalassemia in Migrant Populations in Guangdong Province, China. BMC Public Health. 14, 905, doi:10.1186/1471-2458-14-905 (2014).
5. Xiong, F. et al. Molecular Epidemiological Survey of Haemoglobinopathies in the Guangxi Zhuang Autonomous Region of Southern China. Cite J genet. 78, 139–148, doi:10.1111/j.1399-0004.2010.01430.x (2010).
6. De Sanctis, V. et al. Endocrine Profile of Beta-Thalassemia Major Patients Followed From Childhood to Advanced Adulthood in a Tertiary Care Center. Indian JEndocrinol Metab. 20, 451–459, doi:10.4103/2230-8210.183456 (2016).
7. De Sanctis, V. et al. Acquired Hypogonadotropic Hypogonadism (AHH) in Thalassemia Major Patients: An Underdiagnosed Condition? Medit J Hematol Infect Dis. 8, e2016001 (2014).
8. Dornoykitchaiyaphon, S. et al. Abnormalities in Bone Mineral Density and Bone Histology in Thalassemia. J Bone Miner Res. 18, 1682–1688, doi:10.1359/jbmr.2003.18.9.1682 (2003).
9. Bahar, A. et al. Insulin Resistance, Impaired Glucose Tolerance and Alpha- Thalassemia Carrier State. J Diabetes Metab Disord. 14, 2, doi:10.1186/40200-015-0129-2 (2015).
10. Najafipour, F. et al. Evaluation of endocrine disorders in patients with thalassemia major. Int J Endocrinol Metab 2, 104–113 (2008).
11. Giardina, P. J. “Thalassemia syndromes”, in Hematology: Basic Principles and Practice, R. Hoffman, E. J. Benz, and S. S. Shattil Eds, Elsevier/ChurchillLivingstone, Philadelphia, Pa, USA, 5th edition (2008).
12. Xiong, F. et al. Molecular Epidemiological Survey of Haemoglobinopathies in the Guangxi Zhuang Autonomous Region of Southern China. Clin Genet. 78, 139–148, doi:10.1111/j.1399-0004.2010.01430.x (2010).
13. Yin, A. et al. The Prevalence and Molecular Spectrum of Alpha- and Beta-Globin Gene Mutations in 14,332 Families of Guangdong Province, China. PLOS ONE. 9, e89855, doi:10.1371/journal.pone.0089855 (2014).
14. Huang, H. et al. Molecular Spectrum of Beta-Thalassemia in Fujian Province, Southeastern China. Hemoglobin. 37, 343–350, doi:10.3109/03630269.2013.792274 (2013).
15. Abdulwahid, D. A. & Hassan, M. K. Beta- and alpha-Thalassemia Intermedia in Basra, Southern Iraq. Hemoglobin. 37, 553–563, doi:10.3109/03630269.2013.825841 (2013).
43. Al-Akhras, A.

41. Karamifar, H., Karimi, M., Amirhakimi, G. H. & Badiei, M. Endocrine function in thalassemia intermedia.

40. Eshragi, P., Tamaddoni, A., Zarifi, K., Mohammadhasani, A. & Aminzadeh, M. Thyroid function in major thalassemia patients: Is it...

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Author Contributions
H.C.L. designed and wrote the manuscript. Q.S.L., F.H.H. and C.F.W. collected clinical data. Y.S.W. directed the writing of manuscript. All authors have reviewed and approved the final version of this manuscript.

Additional Information

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