The Hematological and Molecular Spectrum of α-Thalassemias in Turkey: The Hacettepe Experience

Türkiye’de Alfa Talasemilerin Hematolojik ve Moleküler Spektrumu: Hacettepe Deneyimi

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Abstract:

Objective: The spectrum of α-thalassemias correlates well with the number of affected α-globin genes. Additionally, combinations of the several non-deletional types of mutations with a large trans deletion comprising the 2 α-globin genes have an impact on the clinical severity. The objective of this study was to analyze the hematological and molecular data of 35 patients with Hb H disease from a single center in order to identify the genotypes of Hb H disease and genotype-phenotype correlations.

Materials and Methods: Herein, we report the hematological and mutational spectrum of patients with Hb H disease (n=35). Additionally, genotypes of α-gene mutations of 78 individuals, who were referred to our institution for α-gene screening, were analyzed.

Results: Supporting the previous data from Turkey, -α3.7 was the most common mutation among patients with Hb H disease (62.8%) and in the other 78 subjects (39.7%). Of the patients with Hb H disease, the most common genotypes were -α3.7/−20.5, -α3.7/−26.5, and -α3.7/−17.5 in 10 (28.6%), 6 (17.1%), and 6 (17.1%) patients, respectively. Another small deletion, -4.2 alpha, and several non-deletional types of α-gene mutations, namely α (-5nt): IVS-I donor site (GAG.GTG.AGG->GAG.G-----); α (PA-2): AATAAA>AATGGA, and α (cd59): GGC->GAC, were found to be associated with Hb H disease when present at trans loci of one of the large deletions given above. The combinations consisting of 1 non-deletional and 1 of the large deletional types of mutations (α²α/−) at trans loci were found to result in a more severe phenotype compared to the genotypes composed of 1 small trans deletion of a large deletion (-α/−). The combination of α (Cd59) and – in trans was associated with severe phenotype and the disease was associated with an increase in Hb Bart’s level with null Hb H. In spite of the presence of 2 intact α-globin genes, homozygosity for PA-2 mutation resulted in severe Hb H disease.

Conclusion: This study indicated that Hb H disease is not rare in Turkey and its genotype is quite heterogeneous.

Key Words: Molecular, Mutation, α-Thalassemia, Turkey
Özet:

Amaç: Alfa (α) talasemilerin farklı klinik spektrumundan etkilenen α-globin gen sahayı sorumludur. Ayrıca delesyonel olmayan mutasyonların, iki α-globin geninin birden etkilendiği büyük delesyonel mutasyonlarla kombinasyon oluşturmasının da hastalığın klinik şiddetinde etkisi bulunmaktadır.

Gereç ve Yöntemler: Burada Hb H hastalarının (n=35) hematolojik ve mutasyonel spektrumunu sunmaktayız. Buna ek olarak, merkezimize bulunan büyük delesyonel mutasyonların varlığı taraması için merkezimize gönderilen ve α-globin geni mutasyonu taşıyan 78 bireyin bulguları analiz edilmiştir.

Bulgular: Çalışmamızda daha önce bildirilenleri destekle Hb H hastası grubunda (%62,8) ve 78 bireyde (%39,7) en sık mutasyon -α3.7 olarak bulunmuştur. Hemoglobin H hastalarımızda en sık genotipler -α3.7/20.5; -α3.7/26.5 ve -α3.7/17.5 olarak sırasıyla 10 (%28,6), 6 (%17,1) ve 6 (%17,1) sıklıklarda bulundu. Diğer bir küçük delesyon olan -4.2 (Asya tipi), delesyonel olarak, merkezimize α78 bireyin bulguları analiz edilmiştir.

Anahtar Sözcükler: Moleküler, Mutasyon, Alfa talasemiler, Türkiye

Çalışmamız Hb H hastalığının ülkemizde nadir olmadığına ve genotipinin heterojen olduğuna işaret etmektedir.

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Introduction

α-Thalassemia results from a genetic defect in α-globin chain synthesis, often as a consequence of deletional mutations and less frequently due to non-deletional types of mutations [1,2]. α-Thalassemias may occur worldwide; however, they are seen more commonly among populations in South East Asia, the Mediterranean region, and the Middle East [1]. The α-globin gene is located on the short arm of chromosome 16 (16p13.3) and normally there are 4 α-globin gene copies in an individual, with 2 in each allele [3]. The phenotype of α-thalassemias is directly related to the number of α-globin genes affected. α*Thalassemias designate the status of deletion in one of the paired α-globin genes (–α/αα), whereas in α0-thalassemias both of the paired α-globin genes are deleted (–/–αα). Heterozygous α∗α-thalassemia usually causes a silent carrier state. On the other hand, heterozygous α0-thalassemia (–/αα) and homozygous α∗α-thalassemia (–α/–αα) result in hematological findings similar to α-thalassemia trait, except for the Hb A2 value, which is at the normal level or below the normal level in α-thalassemia. The co-existence of both α∗α-thalassemia and α0-thalassemia (–/––) results in hemoglobin H (Hb H) disease [1]. There are also non-deletional types of mutations (α3/αα) resulting in Hb H disease, when a large deletional type of mutation (–/–) co-exists in trans (α5/α–) [4,5].

The most common deletional mutations causing α*thalassemia are -α3.7 and -α4.2, whereas the common deletional mutations causing α0-thalassemias are of 20.5-kb deletion, approximately 17.5-kb deletion (-MED-I), greater than 26.5-kb deletion (-MED-II), and approximately 18-kb deletion (-SEA) [1,4,6]. MED-II has previously been reported in a few Turkish families and from other Mediterranean populations [4].

In this study, the hematological and molecular data of 35 patients with Hb H disease from a single center were analyzed and reported in order to identify the genotypes of Hb H disease and genotype-phenotype correlations, and also to create awareness that Hb H disease is not a rare entity in Turkey.

Materials and Methods

Of the 788 patients who were diagnosed with thalassemia between 1981 and 2014 at our institution, 138 (17.5%) were diagnosed with Hb H disease (Table 1). Unfortunately, from those 138 patients only a total of 35 had genotype data available; those 35 were included in the current study. Splenomegaly was detected at diagnosis, during physical examination, or by ultrasonography in 40% of the patients with Hb H disease. The transfusion histories of patients with Hb H were recorded from patients' files. Of the patients with Hb H disease, 18% received erythrocyte transfusion at least once, and 82% had no transfusion history at diagnosis and received no transfusion during follow-up. The number of transfusions ranged between
1 and 24. One patient was on a chronic transfusion program, whereas the other patients were transfused occasionally. Ethical committee approved this study.

Excluding the patients with Hb H disease, of the individuals screened for α-thalassemia mutations, 78 were found to carry an α-thalassemia mutation. The indications of α-thalassemia mutational screening among those 78 individuals were either having hypochromic microcytic erythrocytes, with normal iron status and Hb A2 below 3.5%, or being the available parent of a patient with Hb H disease.

Results of hematological studies and red cell indices were analyzed. For discussion purposes, values prior to splenectomy or erythrocyte transfusion were taken into consideration. Hemoglobin A2, Hb F, and Hb H values were measured with the previously described methods [7] or high-performance liquid chromatography with the Bio-Rad Variant II system. Supravital stains for Hb H inclusions were examined in all cases [8].

Prior to 2008, α-thalassemia mutations were identified with previously described methods [7,8,9,10,11,12,13]. After 2008, mutation analyses for the α-globin gene were evaluated with the α-Globin Strip-Assay (ViennaLab, Austria), based on the reverse-hybridization technique used for detection of the 21 most common α-thalassemia mutations in the Mediterranean region. Of the 35 patients with Hb H disease, 25 have been reported previously [7].

The obtained data were evaluated with SPSS 21 (IBM Corp., Armonk, NY, USA). Normality test was performed to determine if the data were distributed in a normal fashion. For comparison between groups of more than 2, one-way ANOVA test was used. Statistical significance was determined as p values <0.05.

Results

Of the 35 patients with Hb H disease, the age range was 1.5-50 years at diagnosis (mean: 15.9±12.9 years). The mean values of red blood cell indices at diagnosis are summarized in Table 2a. A total of 10 different genotypes were detected in 35 patients with Hb H disease (Tables 2b and 2c.).

Of the 35 patients with Hb H disease, 22 (62.8%) and 18 (51.4%) were found to have -α3.7 or -α20.5 alleles, respectively (Table 3). The most common genotype was -α3.7/-α20.5 in 10 (28.6%) of the patients, followed by -α3.7/-α26.5 in 6 (17.1%) and -α3.7/-α17.5 in 6 (17.1%). The most common 3 genotypes were distributed among 22 of the 35 patients, representing 62.8% of all genotypes found in patients with Hb H disease. The numbers of Hb H patients having other genotypes were too small to make any statistical analysis; therefore, comparison of the hematological data was made only among the patients with the 3 most common above-mentioned genotypes.

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Statistical analyses of the mean values of red cell indices showed no significant difference among these 3 common genotypes. Hemoglobin F level was found significantly higher in -α3.7/-α17.5 patients (p=0.041), whereas Hb H levels were significantly lower among patients with this genotype compared to the -α3.7/-α20.5 and -α3.7/-α26.5 genotypes (p=0.036). Hemoglobin A2 levels were similar among these 3 genotypes.

Of the patients with Hb H disease, 26 (74.3%) were found to have deletional types of mutations, whereas 9 (25.7%) were found to have non-deletional types of mutations. Comparison of the hematological data of the Hb H patients showed that the group of patients with a genotype consisting of non-deletional types of mutations with a large trans deletion (ααT/-) had statistically lower hemoglobin values (p=0.007) compared to those who had deletional types of mutations with a large trans deletion (-α/-) (Table 4). On the other hand, the mean of Hb H levels was significantly higher in the former patients (18.1±8.3 vs. 7.4±4.7; p=0) than the latter (Table 4). In the examination of the 78 individuals with α-thalassemia mutations other than Hb H disease, the most common genotype was -α3.7/-αα in 31 patients (39.7%) (Table 5). The most common non-deletional genotype was α(PA-1)/αα in 5 of the individuals (6.4%). Of the 78 subjects, 34 (43.5%) and 21 (26.9%) were found to have -α3.7 or -α20.5 alleles, respectively (Table 5).

Discussion

The incidence of deletional α-thalassemia (-α/αα) among newborns screened by globin gene mapping from samples obtained from cord blood at birth has been reported to be 3.6% in Turkey [14]. In other reports, the chromatographic analyses of cord blood samples of newborns in Turkey suggested that -α/αα or (αTαα) thalassemia incidence was between 2.9% and 4.1% [15,16].

In a recent report from Antakya-Hatay, a city in the southern part of Turkey, 300 individuals with moderate anemia, microcytosis, and normal iron levels were tested for α-thalassemia by the aid of α-globin strip assay; of these, 97 were found to have at least 1 mutation in 4 of the α-globin genes [17]. Of these patients, the most common mutation was -α3.7 (57.3%) [17]. Similarly, Öner et al. and Çürek reported -α3.7 as the most common α-thalassemia gene

| Disease               | n (%)       |
|-----------------------|-------------|
| β-thalassemia major/intermedia | 650 (82.5)  |
| Hb H                  | 138 (17.5)  |
| Total                 | 788 (100)   |
**Table 2a.** The age and hematological data of patients with Hb H disease with molecular diagnosis.

| Patients with Hb H | Age at diagnosis (years) | Hb (g/dL) | MCV (fL) | MCH (pg) | MCHC (g/dL) | RBC (x10^{12}/L) | RDW | Hb A2 (%) | Hb F (%) | Hb H (%) |
|---------------------|--------------------------|-----------|-----------|-----------|-------------|------------------|-----|-----------|-----------|-----------|
| Total               | n=35                     |           |           |           |             |                  |     |           |           |           |
| Mean ± SD           | 15.9±12.9                | 9.3±1.6   | 63.1±9.7  | 17.7±1.8  | 30.9±2.3    | 4.7±0.8          | 22.5±8.5 | 1.2±0.4   | 1.3±0.9   | 10.3±7.5  |
| Range               | 1.5-50                   | 6.7-13.7  | 48-98     | 15.3-20.9 | 28.2-35.8   | 2.8-6.4          | 9.5-34.9 | 0.5-2     | 0-4.3     | 1.4-34    |

**Table 2b.** The age and hematological data of patients with Hb H disease with the 3 most common genotypes.

| Genotype | Age at diagnosis (years) | Hb (g/dL) | MCV (fL) | MCH (pg) | MCHC (g/dL) | RBC (x10^{12}/L) | RDW | Hb A2 (%) | Hb F (%) | Hb H (%) |
|----------|--------------------------|-----------|-----------|-----------|-------------|------------------|-----|-----------|-----------|-----------|
| -α^{3.7/} 20.5 | n=10                    | 14.8±9.6  | 9.8±1.6   | 64.6±9.6  | 16.6±1.2    | 29.5±1.1         | 4.9±0.4 | 23.9±2.4  | 1.4±0.4   | 0.9±0.6   | 9.9±5.2  |
| Mean ± SD  | 14.8±9.6                | 9.8±1.6   | 64.6±9.6  | 16.6±1.2  | 29.5±1.1    | 4.9±0.4          | 23.9±2.4 | 1.4±0.4   | 0.9±0.6   | 9.9±5.2   |
| Range     | 1.5-30                   | 8.1-12.3  | 51.4-77   | 15.3-17.5 | 28.2-30.3   | 4.5-3            | 21.2-25.7 | 0.9-2     | 0.5-2     | 2.9-17    |
| -α^{3.7/} 26.5 | n=6                    | 18±6.5    | 9.9±1.5   | 61.5±7.3  | 17.5±1.7    | 30.5±1.5         | 5.4±1.1 | 21.8±11.7 | 1±0.2     | 0.7±0.4   | 8±4.8    |
| Mean ± SD  | 18±6.5                  | 9.9±1.5   | 61.5±7.3  | 17.5±1.7  | 30.5±1.5    | 5.4±1.1          | 21.8±11.7 | 1±0.2     | 0.7±0.4   | 8±4.8     |
| Range     | 8-28                     | 7.5-11.6  | 52-72     | 15.8-19.2 | 29.1-32.1   | 3.9-6.4          | 13-32.8 | 0.9-1.2   | 0-1.1     | 1.5-15.4  |
| -α^{3.7/} 17.5 | n=6                    | 13.6±15.3 | 9.5±0.3   | 56.3±5.3  | 19.2        | 35.8            | 4.9±0.5 | 11.5      | 1.5±0.3   | 2.4±1.3   | 3.3±1.7  |
| Mean ± SD  | 13.6±15.3               | 9.5±0.3   | 56.3±5.3  | 19.2       | 35.8        | 4.9±0.5          | 11.5   | 1.5±0.3   | 2.4±1.3   | 3.3±1.7   |
| Range     | 2-43                     | 9-9.9     | 48-63     | 19.2       | 35.8        | 4.2-5.8          | 11.5   | 1.2-1.9   | 0.6-4.3   | 1.4-6     |
| p         | >0.05                    | >0.05     | >0.05     | >0.05      | >0.05       | >0.05            | >0.05 | >0.05     | >0.05     | 0.041     | 0.036    |
Table 2c. The age and hematological data of patients with Hb H disease associated with rare mutations.

| Genotype                        | Age at diagnosis (years) | Hb (g/dL) | MCV (fL) | MCH (pg) | MCHC (g/dL) | RBC (x10^{12}/L) | RDW | Hb A₂ (%) | Hb F (%) | Hb H (%) |
|--------------------------------|--------------------------|-----------|----------|----------|-------------|-------------------|-----|-----------|----------|----------|
| α (-5nt*)/α (--20.5)           | 13±2.6                   | 8.4±0.6   | 68.3±2.5 | NA       | 4.6±0.8     | NA                | NA  | 0.8±0.2  | 1.5±0.9  | 23.2±9.6 |
| Mean ± SD                      | 10-15                    | 7.9-9     | 66-71    |          | 4-5.6       | NA                | NA  | 0.6-0.9  | 1-2.5    | 15.5-34  |
| Range                          |                          |           |          |          |             |                   |     |           |          |          |
| α (-5nt*)/α (--20.5)           | 27±29.6                  | 9.7±2.3   | 64.8±5.4 | 20.9     | 4.4±1.4     | 19                | NA  | 0.9±0.5  | 0.8±0.3  | 6.5±3    |
| Mean ± SD                      | 6-48                     | 8.1-11.4  | 61-68.6  | 20.9     | 3.4         | 19                | NA  | 0.6-1.3  | 0.6-1.3  | 4.4-8.6  |
| Range                          |                          |           |          |          |             |                   |     |           |          |          |
| α (PA-2**)/α (--20.5)          | 2.3±0.4                  | 7.8±0.4   | 53±2.8   | NA       | 4.3±0.1     | NA                | NA  | 0.7±0.1  | 1.1±0.6  | 13.8±2.5 |
| Mean ± SD                      | 2-2.5                    | 7.5-8.1   | 51-55    |          | 4.2-4.3     |                   |     | 0.6-0.7  | 0.7-1.5  | 12-15.5  |
| Range                          |                          |           |          |          |             |                   |     |           |          |          |
| α (PA-2**)/α (PA-2)            | 38.5±16.3                | 9.1±1.3   | 65.5±2.1 | NA       | 4.2±0.6     | NA                | NA  | 1.5±0.5  | 1.2±0.9  | 10.7±3.8 |
| Mean ± SD                      | 27-50                    | 8.2-10    | 64-67    |          | 3.7-4.6     |                   |     | 1.1-1.8  | 0.5-1.8  | 8-13.4   |
| Range                          |                          |           |          |          |             |                   |     |           |          |          |
| α (cd59 ***)/α (--20.5)        | 8±2.8                    | 6.8±0.2   | 68±0     | NA       | 3.6±0.6     | NA                | NA  | 0.8±0.1  | 1.2±0.4  | 14.1±8   |
| Mean ± SD                      | 6-10                     | 6.7-6.9   | 68-68    |          | 3.2-4.1     |                   |     | 0.7-0.9  | 1-1.5    | 8.4-19.7 |
| Range                          |                          |           |          |          |             |                   |     |           |          |          |
| α (-5nt*)/α (--17.5)           | 10±2.5                   | 8.2       | 64       | NA       | 4.8         | NA                | NA  | 0.5      | 2.9      | 28       |
| Mean ± SD                      | 8-12                     |           |          |          |             |                   |     |           |          |          |
| Range                          |                          |           |          |          |             |                   |     |           |          |          |

*: α (-5nt): IVS-1 donor site (GAG.GTG.AGG>GAG.G-----); **: α (PA-2): AATAAA>AATGGA; ***: α (cd59): GGC>GAC; ****: This value indicates Hb Bart's but not Hb H for this particular patient.
that was associated with Hb H disease in 25 and 32 patients, respectively [7,18]. Our study is compatible with the above stated previously published data pointing out that -α3.7 has been the most common genotype among patients with Hb H disease (62.8%).

In our study, among Hb H patients, the second most common allele was --20.5 (51.4%). This finding is in accordance with the other reports from Turkey [7,18,19]. A hydrops fetalis case due to α-thalassemia associated with homozygosity of --20.5 was also previously reported from Turkey [20].

In the current study, the -MED-II deletion (--26.5) was found as the third most common allele among patients with Hb H disease (25.7%), which was followed by --MED-I deletion (--17.5) at 17%. Contrary to our observation, the --MED-I mutation (--17.5) has been reported as the second most common type of allele by Guvenc et al. with 15.11% frequency among the population of Adana, a city in the southern part of Turkey [21]. This is probably related to the homogeneity of the population studied in that publication.

The -MED-II deletion has been known as a genotype more common among Turkish populations [4], and it was found as the third most common allele in our study.

All of these studies suggest that the molecular pathology of Hb H disease is heterogeneous and, according to our study, the most common genotypes associated with Hb H in 35 patients who were referred to us from all over Turkey are as follows: -α3.7/--20.5 (28.6%), -α3.7/--26.5 (17.1%), and -α3.7/--17.5 (17.1%) (Table 2b).

In the current study, 25.7% of the patients with Hb H disease who had a combination of large deletional and non-deletional (ααT--) mutations were found to have statistically significantly lower Hb and higher Hb H levels compared to those of patients having combinations of large and small deletional (α/--) types of mutations (Table 4). This finding was compatible with the previously published data [1,2,3]. This study revealed the presence of 3 different non-deletional types of mutations, namely the (-5nt), PA2, and C59 mutations. It seemed that the most common non-deletional type of combination involved in Hb H was (-5nt/--), which was found in 3 patients (8.6%) in the current study. Contrary to this, α (PA-2)/--MED-II was the most frequent non-deletional combination in a regional study by Çürük [18]. It was interesting that in spite of the presence of 2 intact α-globin genes, homozygosity for PA-2 mutation (αPA-2/αPA-2) resulted in severe Hb H disease in 2 patients (Table 2c); this was discussed elsewhere [7].

### Table 3.

| Genotype     | Number of chromosomes affected |
|--------------|--------------------------------|
| -α3.7        | 22                             |
| -α4.2        | 4                              |
| α (PA-2)     | 6                              |
| α (-5nt)     | 4                              |
| α (cd59)     | 1                              |
| --20.5       | 18                             |
| --17.5       | 9                              |
| --26.5       | 6                              |
| Total        | 70                             |

### Table 4.

| Genotype                       | Hb (g/dL)  | RBC (x10¹²/L) | MCV (fL) | Hb A₂ (%) | Hb F (%) | Hb H (%) |
|-------------------------------|------------|---------------|----------|-----------|----------|----------|
| Combination of deletional mutations* (n=26) | 9.7±1.3    | 4.9±0.7       | 61.9±7.8 | 1.3±0.4   | 1.2±0.9  | 7.4±4.7  |
| Combination of deletional and non-deletional mutations** (n=9) | 8.4±2      | 4.1±0.8       | 67.6±13.1| 0.7±0.1   | 1.5±0.7  | 18.1±8.3 |

*pOf these 26 patients, 8 were below 10 years of age.
**Of these 9 patients, 4 were below 10 years of age.
The distribution of deletional and non-deletional types of α-thalassemia mutations in 78 individuals.

| Genotype                  | n (%) |
|---------------------------|-------|
| -α3.7/α                   | 31 (39.7) |
| --20.5/αα                 | 21 (26.9) |
| --26.5/αα                 | 8 (10.3) |
| α (PA-1)/αα               | 5 (6.4) |
| α (Cd59 G>A)/αα           | 4 (5.1) |
| α (IVS 1-5 nt)/αα         | 3 (3.8) |
| -α3.7/-α3.7              | 2 (2.6) |
| α (PA-2)/αα               | 1 (1.3) |
| α (Cd14 G>A)/αα           | 1 (1.3) |
| α (Cd14 G>A)/α-3.7       | 1 (1.3) |
| -α17.5/αα                | 1 (1.3) |
| Total                     | 78 (100) |

In this study, we did not find any of the previously described α-gene mutations from Turkey, such as -THAI, --FIL, init.cd, Cd 19, Hb Icaria, Hb Pakse, or Hb Koya Dora [14,16,17,18,19,21]. In a previous study from our center, the rate of unidentifiable mutations among individuals with α-thalassemia mutations was reported to be 2.72% [22]. In this study, all of the mutations among patients with Hb H disease were known mutations. In the previous study from our center, among individuals with α-thalassemia major, the most common 3 mutations were distributed among 69.39% of the patients [22]. In this study, it was shown that the most common 3 genotypes associated with Hb H accounted for almost 63% of the study group.

In the previous reports by Altay and by Akar and Altay, related to National Hemoglobinopathy Registry data, Hb H was reported to be 3.6% (n=103) of all hemoglobinopathies in Turkey [22,23]. In our cohort study from a single center, it was shown that Hb H disease was diagnosed in 17.5% of the total 650 thalassemic patients (Table 1). The latest figure for α-thalassemia major in Turkey was reported to be 3.6% (n=103) of all hemoglobinopathies [24,25]. Therefore, according to the data of our center as stated above, the total number of Hb H patients in Turkey should be around 550. The discrepancy in the rates of Hb H between 2002 data and the current study may derive from the higher awareness of the disorder in some centers in recent years, more accurate diagnoses, and/or developments in the diagnostic tools of Hb H disease and/or an increase in referral rates of anemic patients from peripheral to tertiary centers like ours. Therefore, if the figure of the current study reflects a more accurate value of the number of Hb H cases, we may expect to diagnose more patients in the near future.

In conclusion, as our center is a referral center in the mid-Anatolia region with a patient profile from all over the Turkey, the results of our study may represent the Hb H disease rates among the overall Turkish population. Some of the data of this study were in agreement with previous reports [7,8,9,16,17,18,19,20], and our current study also indicated that the molecular spectrum of α-thalassemias is quite heterogeneous in Turkey, as all together 9 deletional and non-deletional mutations and 10 combinations of them were found to be associated with Hb H disease. In previous reports, the mutational spectra were reported to be less heterogeneous among smaller populations, such as among Cypriots and Iraqi Turks [26,27]. Although in this study the molecular pathology of Hb H disease has been addressed, the frequencies of rare genotypes associated with α-thalassemia requires more patients and further population studies, since most of the individuals screened for that purpose in the current study were parents of the patients with Hb H disease, a limiting factor in prediction of the population frequencies of several genotypes. This study also showed that Hb H disease is not uncommon in Turkey; therefore, this disease should be kept in mind in discussion of microcytic anemias and all efforts should be made for correct diagnosis of α-thalassemias. Detection of new cases will be helpful in determining the allele frequencies of different α-thalassemia mutations.

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Conflict of Interest Statement

The authors of this paper have no conflicts of interest, including specific financial interests, relationships, and/or affiliations relevant to the subject matter or materials included.

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