The Normal HbA2 Hematological Phenotype of β-Thalassemia Trait. Problems in Detection and Measures to Improve Sensitivity of Screening Tests

Christos Kattamis*
Thalassemia Unit, National Kapodistrian University of Athens, Greece

In population with a high incidence of β-thalassemia trait, the presence of normal HbA2 hematological phenotype (normal HbA2, β-thal) carriers may negatively influence the efficacy of prevention programs. This is mainly due to the fact that the hematological tests recommended for massive screening to detect couples at risk are basically addressed to the high HbA2, β and δβ'-thal traits. Normal HbA2, β-thal carriers necessitate additional, examinations for identification. Thus, in population with remarkable incidence of normal HbA2, β-thal trait, measures to improve sensitivity of screening tests to detect normal HbA2 carriers should be considered [1].

Normal HbA2, β-thal trait was first described in one of the parents of 7 families, with the mild clinical phenotype of Thalassemia Intermedia (TI) [2]. Later 9 Greek families with normal HbA2, β-thal trait in one parent were reported: in six the offspring had TI and in three the severe clinical phenotype of thalassemia major (TM). Based on the clinical, hematological and globins synthesis studies of the family's two subtypes of normal HbA2, β-thal trait were characterized:

a) The mild (type 1), in which normal HbA2, β-thal carriers had minimal red cells changes, mild increase in a-to a non-a chains synthesis ratio and off springs with TI, with low HbF (<20%).

b) The severe or type 2 normal HbA2, β-thal with marked red cell changes similar to those of the l high HbA2, β-thal and off springs with the severe clinical phenotype of Thalassemia Major with high HbF >80% [3].

Extensive hematological studies on 475 homozygous β thalassemia patients and their parents, confirmed the existence of the two types (mild and severe) of normal HbA2, β-thal trait in Greek population. Of 950 β-thal heterozygotes, 35 (3.7%) were heterozygotes for normal HbA2, β-thal, 13 (1.4%) with type 1 and 22 (2.1%) with the severe type 2 [4].

This assumption was confirmed later by extensive molecular studies, which revealed a 7.2 kb deletion partially removing the δ globin gene and a single nucleotide mutation (G → A) in IVS1-5 in cis. The mutation, named Corfu δβ’ mutation, from the place of origin of the first patient [5,6]. The delta beta Corfu mutation seems to be a unique, specific mutation for greek population; up to now it was not detected in other countries.

The hematological findings of the normal HbA2, β-thal carriers with mild or severe changes in red cells morphology and increased osmotic fragility are also common in α- and (δβ) thal heterozygotes and in individuals with iron deficiency anemia; (δβ)’thalassemia carriers are easily recognized from the high HbF levels (>5%) and iron deficiency anemia from the low levels of serum iron, transferrin saturation and ferritin.

Prior the implementation of molecular methods, differentiation between carriers of normal HbA2, β-thal and α-thal was difficult; it necessitated chains synthesis studies, a method available only in expertise hematological laboratories. The diagnosis of normal HbA2, β-thal heterozygote is of major concern, in countries implementing prevention programs using hematological tests to screen and detect couples at risk. Hematological tests frequently fail to detect normal HbA2, β-thal carriers.

A study for identification of causes diminishing the efficacy of Greek prevention programs, showed that a major cause was the failure of detection normal HbA2, heterozygote’s, especially with type 1.

The prevalence and type of normal HbA2, β-thal carriers vary widely in population with high prevalence of β-thal trait. Heterogeneity in the prevalence of normal HbA2 is mainly related to the spectrum of β-thalassemia mutations prevailing in the population. Relative high incidences of carriers with normal HbA2, β-thal are expected in populations with a relatively high incidence of the very mild β++’ (silent), and the mild β++ mutations.
To our knowledge, population’s studies on the prevalence and molecular basis of normal HbA2 β-thal are limited to a few countries. Greece is one of them. In Greece about 30 β-thalassemia mutations have been identified so far. The type and incidence of mutations in 500 patients (50 with TI) with β thalassemia are summarized in Table 1 (separately for TM and TI) [7].

Analyzing the relation of hematological data to the genotypes of parents and patients it was realized that severe type of normal HbA2 β-thal carriers, was mainly due to δ0 β+ Corfu mutation and to a lesser extent to the co-existence of severe β0 or β+ thalassemia mutations combined with a δ0 or δ+ mutation, or to a mild β++ mutation mainly IVSI-6 and -83 [8,9]. On the other hand the mild type 1 mutation was found in all the very mild (silent) mutations mainly -101(C-T) and +1480 (C-T) and the β++ mild mutation IVS1-6 and -83 (Table 1) [10].

To minimize the failure of detection of normal HbA2 heterozygotes applying the recommended hematological tests for screening, a reduction of the HbA2 cut off level of >3.5 to >3.2 and even >3.0% was proposed recently, in parallel to molecular studies not only for the β, but also for the α-globin genes [1]. This was considered necessary when it was realized that few patients with thalassemia intermedia, resulted from worsening of the clinical phenotype of heterozygotes β0 or β+ severe mutations, combined with a triplicate ααα or quadruplicate αααα genes. In contrast it is well known that co-existence of αα αα αα αα genes ameliorates the clinical phenotype of patients with severe TM clinical phenotype. Similar problems are expected to be found in populations with high prevalence of β-thalassemia and a relative remarkable incidence of normal HbA2 heterozygous related to mild and very mild β-thalassemia mutations.

Finally we like to stress that characterization of genotype of β- thal heterozygous as well as that of homozygous patients is most valuable not only for prenatal diagnosis counseling, but also for anticipation of prognosis and treatment. of patients The recent recognition of the severe late complications of pulmonary hypertension, predisposition to venous thrombosis (especially after splenectomy), osteoporosis and others, in non-transfused patients with TI are changing the outlook for antenatal diagnosis, prognosis and treatment of patients with β and δβ thalassemias.

REFERENCES


5. Wainscoat JS, Thein SI, Wood WG, Weatherall DJ, Metaxotou-


