CASE REPORT

A case of late diagnosis of compound heterozygosity for Hb Adana (HBA2:c.179G>A) in trans to an α+- thalassemia deletion: guilty or innocent

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Abstract

Background: Hemoglobin Adana is a non-deletional alpha chain mutation, particularly rare, and to date, it is mostly described in coinheritance to other a-thalassemia mutations. Such interactions result in various phenotypes depending on the underlying genotype. Since routine hematological tests do not detect the aforementioned unstable variant, it is quite likely a diagnosis to be missed or delayed, with any complications this may have for a patient.

Description of the case: A case report of late mutation identification in a 64-year-old woman of Greek origin is described. The importance of conducting not only molecular studies to confirm common mutations, such as the –a3.7 kb deletion, but also DNA studies in patients whose phenotype and results of standard tests are not consistent or who present with severe, late complications is highlighted.

Conclusion: The awareness of the necessity for accurate diagnosis is raised, especially in populations that thalassemia prevails and is attributed to numerous mutations. HIPPOKRATIA 2020, 24(1): 38-42.

Keywords: Hemoglobin Adana, thalassemia, extramedullary hematopoiesis

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Introduction

Mutation of the globin genes can occur due to either deletion, most frequently, or point mutations resulting in a deficient synthesis of the globin chains or abnormal hemoglobin (Hb) molecules. HbH disease is the most common form of α- thalassemia (α-thal) syndrome, and although it is basically caused by deletional mutations, it may also arise from coinheritance of α^0- thalassemia with non-deletional mutations or with abnormal Hb variants such as Hb Constant Spring[1,2]. In recent years, the amount of unstable α-chain hemoglobin variants being identified is constantly increasing[1,3]. Patients with HbH disease generally present with mild symptoms, normal growth and development, and bearable anemia without requiring regular blood transfusions. Moreover, based on previous reports, patients with non-deletional α-thal mutations almost always present with a more severe phenotypic expression than those with large deletion ones[2,4].

Hb Adana (HBA2:c.179G>A or HBA1) is a highly unstable and rare hemoglobin (Hb) variant caused by a point mutation in codon 59 of either the alpha 1 or alpha 2 globin gene resulting in a Gly → Asp substitution. This compromises the stability of the Hb molecule by disrupting its folding and creates a structural modification, which can steer towards an α-thal phenotype.

This non-deletional alpha chain mutation was first identified in two Turkish patients by Curuk et al[5]. In literature to date, Hb Adana has been reported to interact with both deletional and non-deletional α-thalassemia mutations mainly observed in populations of Southeast Asia, Philippines, Indonesia, and the Mediterranean[3,6-13]. It is noteworthy to mention that highly unstable α-globin variants, like Hb Adana, in the heterozygotic state do not lead to remarkable clinical features and hematological findings[1,8]. Consequently, all the cases mentioned above recognize individuals carrying hyper-unstable Hb variants along with other α-thal mutations demonstrating mild to severe phenotypes. More specifically, Hb Adana carriers present asymptomatic, while in compound heterozygosity to other α-chain mutations, there is a diversity of phenotypes[13]. This is related to the excess of stable, functional α-globin polypeptide chains produced by the affected α-globin genes and is also dependent on which of the α-globin genes (the α1 or the more dominantly expressed α2) has the molecular defect.

It is interesting that Hb Adana, when associated with α^0- thal alleles (--FIL)[10,14] or (--SEA)[3] leads to very severe Hb H hydrops fetalis that can provoke intrauterine fetal

death. Couples at risk with such combinations should be informed and counseled about the risk in their pregnancies, and prenatal diagnosis should be offered.

Therefore, the clinical manifestations vary from moderate anemia to typical HbH disease, involving Hydrops fetalis, or α- thal intermedia (a-TI)[7,13]. Herein, a case report of Hb Adana in coinheritance with the –α3.7 gene deletion in a subject of Greek origin is presented. This is the third case diagnosed in the Greek population[11,12]. The patient gave consent to the publication of this report.

Case report

The patient, a 64-year-old woman, was referred to the outpatient hematology clinic for hemoglobinopathies due to chronic hemolytic anemia and significant splenomegaly. In particular, the patient suffered from hypochromic, microcytic, hemolytic anemia, and distinguishable splenomegaly, also affecting the platelet life-expectancy to the level of thrombocytopenia. According to her medical history, the patient's anemia first presented during her childhood, and it had been progressively worsening, while, in the meantime, it was characterized as a non-transfusion-dependent anemia. Regarding her medication, she had received folic acid regularly and denosumab injections, a receptor activator of nuclear factor-kappa B ligand (RANKL) inhibitor, for postmenopausal osteoporosis. Through a period of more than ten years, the patient had undergone various tests to confirm or rule out a diagnosis. There had always been inadequate data from her family history. Nonetheless, the possibility of a pathologic hemoglobinopathy causing the symptoms was excluded by previous hematologists. She had three bone marrow aspirations in search of a possible myeloproliferative disorder, a bone marrow infiltration from neoplastic cells or, even a diffuse lymphoma. The karyotype from the bone marrow sample was also normal, and the test for potential paroxysmal nocturnal hemoglobinuria (PNH) disease was negative. What is also important, the delay to the diagnosis in combination with the persistent anemia stimulated the extramedullary hematopoiesis, a hostile hematopoietic environment for stem and progenitor cells in the bone marrow. Therefore, this case is notable since the disease had already progressed in a harmful way for the patient, and it was of great importance to recognize the pathological background. That was the time when she got referred to our clinic for a search of a hemoglobinopathy.

Clinical examination revealed increased pigmentation of skin exposed to light, mild jaundice, icteric sclera, and a palpable spleen was noted, as well. Her hematological laboratory findings were as follows: Hb 10.9 g/dL, Ht 35.9%, mean corpuscular volume 72.7 fl, mean corpuscular hemoglobin content 22.1 pg, red cell distribution width 19.5 %, reticulocyte count 2.7 %, platelet count 95 10^9/L, ferritin 701.44 ng/mL, total bilirubin 2.20 mg/dL, direct bilirubin 0.7 mg/dL, LDH 188 U/L, Vitamin B12 298 pg/mL, G6PD 2.3 U/gHb (partial deficiency, reference range: adults: 5-14 U/gHb), haptoglobin <8 mg/dL. The blood film showed anisocytosis, poikilocytosis, polychromasia, and basophilic stippling, while inclusion bodies were absent after incubation. High-performance liquid chromatography (HPLC) analysis detected hemoglobin A, A2 (1.3 %), and F (4.3 %), and no abnormal Hb fractions were observed with gel electrophoresis (SDS-PAGE). The radiological findings confirmed the enlargement of the spleen with a cephalocaudal diameter of 21 cm. Moreover, CT imaging revealed significant extramedullary hematopoiesis in para-vertebral regions of the lower thoracic spine with a maximal diameter of 3,5cm. Finally, DNA genetic sequencing featured compound heterozygosity for the HB Adana variant on the α2-globin gene in trans to the α3.7 kb deletion, whereas no abnormal mutations of β-globin genes were detected. The patient was advised to start transfusion due to extramedullary hematopoiesis, and she refused. Splenectomy was then performed after prophylactic vaccination for pneumococcus and meningococcus. She now maintains a Hb level near 13 gr/dl and receives chelation therapy and Hydroxycarbamide.

Discussion

This is a case report of α-thalassemia intermedia with a compound heterozygosity genotype for Hb Adana and $–α^{3.7}$ kb deletion in a Greek patient. This same interaction has been previously detected in this population and reported in only two more families, both individuals being diagnosed in their childhood and both presented with a very mild phenotype[11,12]. Unlike those cases, this one differs regarding both the age of the diagnosis and its effect on the patient's clinical outcome. This report underlines the necessity to consider a rare and unstable Hb variant, Hb Adana, in patients who present with symptoms reminding thalassemia; even if inclusion bodies are negative, the Hb electrophoresis comes out normal, or the routine hematology studies are not alarming. This uncommon molecule isn't visualized by them[6]. And above all, this need is increased, since delayed diagnosis might have a negative impact to the patient's organs and even lead to extramedullary hematopoiesis. Last but not least, another reason for high suspicion of non-deletional Hb variants is that heterozygotes for hyper-unstable α chain Hb variants often have no abnormal hematological profile. Consequently, prevention programs cannot easily facilitate their detection since accurate diagnosis is often only achieved through DNA analyses and could easily be misdiagnosed.

Nowadays, more and more studies regarding the HbH disease tend to evidence that not only HbH disease could be more damaging and not as benign a form of α- thalassemia syndrome as most expected, but also that more patients with non-deletion HbH disease appear with a great clinical diversity[2]. The molecular and phenotypic expression of HbH disease in Greece appears to vary with a wide spectrum of gene mutations and multiple clinical manifestations, only a few of which are affiliated with an adverse outcome[4]. Thus, we highlight the significance of

correct evaluation of clinical, hematological, biochemical, and figurative findings to patients with anemia and splenomegaly and the association to common genotypes, or when undetected, to more rare ones.

Conflict of interest

The authors report no conflicts of interest. The authors alone are responsible for the content and writing of the paper.

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