J Clin Haematol. 2022;3(1):9-11.

Journal of Clinical Haematology Case Report

Phlebotomy in Congenital Erythrocytosis and in Sickle Cell Disease HbSC

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Received date: February 18, 2022, **Accepted date:** March 03, 2022

Citation: Testa ER, Masotti A, Valeri P, Geremia L, Brunetta V, Testa R, et al. Phlebotomy in Congenital Erythrocytosis and in Sickle Cell Disease HbSC. J Clin Haematol. 2022;3(1):9-11.

Abstract

The inherited haemoglobin disorders and the clinical laboratory is essential in the diagnosis. Phlebotomy is the therapeutic procedure indicated to reduce blood viscosity and decreasing the risk of thrombotic events in the haemoglobin variants with high affinity for O_2 or vaso-occlusive crises in SCD with double heterozygosity HbSC. We describe how therapeutic bleeding remains the treatment in two variant haemoglobinopathies, *Hb Regina* and the *Hb Trollhattan,* reported for the first time in the Italian population and a double heterozygosity HbSC. In patients with high haematocrit level when other secondary conditions of polyglobulia are excluded, a high liquid chromatography (HPLC) analysis for haemoglobin study should be suggested.

Keywords: Haemoglobinopathy, Sickle cell disease, High liquid chromatography analysis, Polyglobulia, Phlebotomy

Introduction

The inherited haemoglobin disorders, including thalassemias and haemoglobinopathies, are the most common genetic diseases and the clinical laboratory is essential in the diagnosis. Haemoglobinopathies are monogenic disorders in the genes that encode globin chains that can lead to a defective globin production or a variant in the haemoglobin structure.

The replaced amino acid nature and its location are responsible for the stability, solubility and function of the molecule that could lead to clinical manifestations. Some haemoglobinopathies are clinically silent and are discovered accidentally [1]. Congenital erythrocytosis is a clinical manifestation that can be found in patients with structural haemoglobin variants with a high oxygen affinity [2]. Otherwise, some structural alterations of the haemoglobin such as in the sickle cell disease (SCD), in particular in the double heterozygosity HbSC, could have a high haematocrit level with increased risk of haemoglobin polymerization and subsequently falcemic crisis.

Phlebotomy is the therapeutic procedure highly indicated to reduce blood viscosity and improve clinical conditions, decreasing the risk of thrombotic events in the haemoglobin variants with high affinity for O_2 or vaso-occlusive crisis in SCD with double heterozygosity HbSC.

In this report we describe three different haemoglobinopathies found during a haematologic counselling, two of them with high oxygen affinity [3] and one double heterozygosity HbSC disease that needed phlebotomy treatments to reduce hyper viscosity [4]. The rarity of these haemoglobinopathies in our country proved that a high liquid chromatography (HPLC) analysis for haemoglobin study should be required in the diagnostic tests of patients with high haematocrit level, in absence of other secondary conditions of polyglobulia.

Clinical Case 1

A 68-year-old patient born in Italy was treated for a polyglobulia with quarterly phlebotomies of 450 ml. per year without a significant improvement of the haematocrit

level. The patient was a non-smoker without symptoms of hyper viscosity. At the diagnosis the hemochromocytometric analysis showed Hb 19.2 gr/dl, Hct 61.5%, normal levels of leucocytes and platelets. JAK2 test resulted negative whereas EPO level was increased. Other cardiac, nephrological and pneumological causes were excluded. Iron balance was normal. The patient revealed that the mother was affected by an erythrocyte plethora that was never investigated in the past. Therefore, at the counselling, an HPLC analysis was performed to investigate haemoglobin variants as a possible aetiology. The HPLC chromatogram showed an abnormal peak in the P3 area of a 39.7%, HbA 48.8%, HbA2 2.6% and HbF less 0.8%. The HPLC result was suggestive of haemoglobinopathy and in subsequent molecular analysis of nucleotide sequence of the beta globin gene from nucleotide -190 to +530 and from +1120 to +1500; it revealed a mutation C>G at the nucleotide +469 (codon 96), in a heterozygous state that was responsible for an amino acid substitution [ß 96 Leu> Val] characteristic of the variant *Hb Regina*.

Clinical Case 2

Two brothers 57 and 58 years old born in Italy were blood donors in the past, but they were respectively deferred from the donation for repeated values of high haematocrit level. Other cardiac, nephrological and pneumological causes were excluded. Both brothers were non-smokers and were asymptomatic. They were introduced into a therapeutic phlebotomy treatment regime to reduce the haematocrit and put into an anti-aggregation therapy. The haemachrome showed erythrocytosis RBC 5.45 $\times 10^6$/uL, Hb 18-19 gr/dl, Hct of about 58%, normal levels of leucocytes and platelets. JAK2 analysis resulted negative. During counselling, they referred different personal histories of phlebotomy: the first was in therapy since 2004 with phlebotomies of 400 ml with a two months interval, while the second since 2006 with quarterly intervals. During familial anamnesis the patients reported a nonspecific polyglobulia in the parents and daughter.

An HPLC analysis was performed and the chromatogram showed a haemoglobin variant represented by 40-44%, HbA2 2.3-2.2%, HbF <1.0%. The chromatograms required identification by genetic testing that revealed heterozygosity for *Hb Trollhattan*. The analysis of the nucleotide sequence of the beta globin gene from nucleotide 190 to +530 and from +1120 to +1500 showed a mutation T> A at the nucleotide 112 (codon 20), that was responsible for an amino acid substitution [ß 20 Val> Glu].

Clinical Case 3

In 2013 a 29-year-old young man born in Ghana was known to be affected by a sickle cell disease with double heterozygosity *HbSC*. He was admitted to the hospital six times in a year with a painful vaso-occlusive crisis, due to constant hyper viscosity, but in the absence of anaemia and pathologies [5,6]. Due to an unacceptable response to hydroxyurea [7], he was maintained solely with folic acid treatment. Pre- bleed values were: Hb 14.5-16.0 g/dL, iron dosage normal. An HPLC study was performed to evaluate the pathologic haemoglobin. The chromatogram showed two haemoglobin variants *HbS 46%, HbC 43.7%*, HbA2 4%, HbF 0.9% and HbA 3% that did not need a molecular biology study. The patient started a treatment of phlebotomies of about 400 ml every three months to reduce blood viscosity achieving and maintaining haemoglobin value of 10-12 gr/dl. The clinical condition of the patient improved and no longer required hospital admission for pain due to vaso-occlusive crisis.

Discussion

In our report we describe how therapeutic bleeding remains the treatment of choice to reduce hyper viscosity that manifests itself in pathological situations with different etiopathogenesis such as haemoglobinopathies.

Hb Regina and the *Hb Trollhattan,* first described in a Scandinavian family and in a Swedish one respectively, are identified among the variants of about 100 haemoglobins; they are characterized by a compensatory erythrocytosis of genetic origin, related to oxygen release by haemoglobin. To our knowledge this is the first report in the Italian population; individuals with haemoglobin disorders were observed in various countries, however finding them outside the first described geographic area shows that the people left the original regions for various reasons in the past [8,9].

The reduction of the haematocrit remains effective with phlebotomy. The experience of congenital erythrocytosis suggests that it is sufficient to reduce the Hct to 52% with quarterly blood drawing; in the case of comorbidity for thrombotic risk, it is suggested to maintain the Hct value at 50%.

Interestingly, in the patient with double heterozygosity *HbSC,* the sickle cell crisis are due to hyper viscosity in the absence of anaemia: an unusual clinical picture compared to homozygous sickle cell disease (*HBSS*) in which phlebotomy therapy is necessary to maintain Hb levels between 10-12 g/dL for reducing vaso-occlusive crisis [10].

To date patients with haemochromatosis are not denied as blood donors. Persons with high haematocrit levels and carrying a haemoglobin variant with a high oxygen affinity with congenital erythrocytosis are unlikely to be suitable for blood donation. Indeed, they will not be able to maintain a good therapeutic haematocrit range with regular donation intervals of 90 days and it is strongly possible that transfusion of blood cells with haemoglobin variant having a high oxygen affinity would not have any clinical benefit.

Conclusion

Our report wants to focus the attention on these

rare hemoglobinopathies in our country; a high liquid chromatography (HPLC) analysis for Haemoglobin study should be suggested in the diagnostic tests of patients with high haematocrit level when other secondary conditions of polyglobulia are excluded. The reduction of the haematocrit remains effective with phlebotomy, to think about in some genotypes of Sickle cell diseases like HbSC or in rare Hemoglobinopathies with high oxygen affinity.

References

1. Kalotychou V, Tzanetea R, Konstantopoulos K, Papassotiriou I, Rombos I. Erythrocytosis due to a combination of the high oxygen affinity hemoglobin variant, Hb Olympia [β20 (B2) Val→ Met] with β-and α-thalassemia mutations: first case in the literature. Hemoglobin. 2010 Aug 1;34(4):383-8.

2. Bard H, Peri KG, Gagnon C. The biologic implications of a rare hemoglobin mutant that decreases oxygen affinity. Pediatric research. 2001 Jan;49(1):69-73.

3. Wajcman H, Galactéros F. Hemoglobins with high oxygen affinity leading to erythrocytosis. New variants and new concepts. Hemoglobin. 2005 Jan 1;29(2):91-106.

4. Markham MJ, Lottenberg R, Zumberg M. Role of phlebotomy in the management of hemoglobin SC disease: case report and review of the literature. American journal of hematology. 2003 Jun;73(2):121-5.

5. Scheifer C, Lionnet F, Bachmeyer C, Stankovic-Stojanovic K, Georgin-Lavialle S, Alamowitch S, Marro B, Mattioni S. Cerebral fat embolism in hemoglobin SC disease. The American Journal of Medicine. 2017 May 1;130(5):e187-9.

6. Colella MP, de Paula EV, Machado-Neto JA, Conran N, Annichino-Bizzacchi JM, Costa FF, et al. Elevated hypercoagulability markers in hemoglobin SC disease. haematologica. 2015 Apr;100(4):466-71.

7. Luchtman-Jones L, Pressel S, Hilliard L, Brown RC, Smith MG, Thompson AA, et al. Effects of hydroxyurea treatment for patients with hemoglobin SC disease. American Journal of Hematology. 2016 Feb;91(2):238-42.

8. Percy MJ, Butt NN, Crotty GM, Drummond MW, Harrison C, Jones GL, et al. Identification of high oxygen affinity hemoglobin variants in the investigation of patients with erythrocytosis. Haematologica. 2009 Sep;94(9):1321-2.

9. Orvain C, Joly P, Pissard S, Badiou S, Badens C, Bonello-Palot N, et al. Diagnostic approach to hemoglobins with high oxygen affinity: experience from France and Belgium and review of the literature . In Annals of Clinical Biology 2017 Jan 1 (Vol. 75, No. 1, pp. 39-51).

10. Summarell CC, Sheehan VA. Use of hydroxyurea and phlebotomy in pediatric patients with hemoglobin SC disease. Experimental Biology and Medicine. 2016 Apr;241(7):737-44.