Cas Clinique

TRANSFUSION-INDUCED HEMOGLOBINOSE C IN A BETA-THALASSEMIC MAJOR PATIENT: A CASE REPORT.
HEMOGLOBINOSE C INDUITE PAR UNE TRANSFUSION CHEZ UN PATIENT BETA-THALASSEMIQUE HOMOZYGOTE: A PROPOS D'UN CAS.

Dr NECHAR Maya, Pr DJENOUHAT Kamel.
Laboratory of medical biology, Public Hospital, Rouiba, Algiers. Pharmacist university of Algiers. nechar.maya@hotmail.com
Laboratory of medical biology, Public Hospital, Rouiba, Algiers. Medecin university of Algiers. djenouhatkamel@gmail.com

Résumé

L'électrophorèse capillaire de l'hémoglobine est une technique utilisée depuis de nombreuses années dans le diagnostic des hémoglobinopathies. Elle a l'avantage d'être une technique à la fois qualitative et quantitative, permettant de ce fait, d'identifier et de quantifier différentes fractions hémoglobiniques.

Les hémoglobinoses induites par la transfusion restent un diagnostic rare. Cependant, elles doivent être discutées devant un taux anormalement bas d'un variant hémoglobinique.

Abstarct

The hemoglobin capillary electrophoresis is a common technique used in the diagnosis of hemoglobinopathies, which have the advantage to separate and quantify the different fractions of hemoglobin. Transfusion-induced hemoglobinopathy is an uncommon diagnosis that needs to be discussed when the percentage of variant hemoglobin is abnormally low.

Introduction

Transfusion-induced hemoglobino-pathy may be defined as the transfusion of a unit of non-AA blood leading to the consideration of an alternative hemoglobin (Hb) variant in the recipient.(1) Generally, the blood donor carrying the hemoglobin variant is heterozygous, has no significant abnormalities and is clinically asymptomatic.

This can have many consequences not just increasing the cost of analysis by repeating the test, misdiagnosis and improper counseling, but also causing delay in the diagnosis, because of the diagnosis dilemma. Rarely, the complications related to the variant Hb, e.g. Hb S, have also occurred in the recipients.(2)

Here is the case of Hb C hemoglobinopathy due to transfusion.

Case Report

We report the case of an 15-month-old male child from a first degree consanguineous marriage, with Beta-thalassemia major, where capillary electrophoresis of hemoglobin, on Capillarys flex piercing 2 Sebia, performed for a second time, in a family investigation, revealed a peak of 9.6% in the Hb C zone (second zone) creating a diagnostic dilemma. **Figure (1A).**The parents were heterozygous for beta-thalassemia with 5.2% of Hb A2 each one. **Table (1)** However, there was no abnormal peak in Hb C zone, in their Hb profile. The patient had received 100 ml of red blood cells (RBCs) 08 days prior. In his old records, no Hb C was mentioned in a capillary electrophoresis done 07 months back, which had revealed two different fractions of hemoglobin: 98.8 % Hb F and 1.2 % Hb A2.

A repeated Hb capillary electrophoresis and hemoglobin HPLC (BIO-RAD D10, Dual-program) on patient's blood was carried 6 months later, which revealed no peak in the C zone (C window). **Figure (1B, 1C)**

It's well know that the percentage of Hb C in heterozygotes for Hb C approximates 35-40 % (3), unless there is an associated alpha gene deletion or an iron deficiency. A transfusion acquired hemoglobin C was suspected based on the unexpectedly low percentage of Hb C, it's absence in the followed tests and the pattern of the hemoglobin capillary electrophoresis of the parents.

To confirm the involvement of transfusion we used the identification number of transfused red blood cell units noted in the medical record, to contact donor and finish investigations.

The blood donor was a 54 years-old male, from B negative cc ee K negative blood group. The Hb capillary electrophoresis reveled a heterozygous composite HbC/ beta-thalassemia with 90,8%% Hb C, father of two beta-thalassemic major females, from B negative cc ee K negative blood group.

Discussion

In 1950 Itano and Neel, described hemoglobin C properties including its electrophoretic one. (4) The clinical description accompanying Hb C in the heterozygous state or in association with hemoglobin S were reported by Kaplan, Zuelzer and Neel, one year later.(5)

Hb C ($\alpha 2\beta 2^{6Glu \rightarrow Lys}$) can be present in the heterozygous state (hemoglobin C trait), in the homozygous state (hemoglobin C disease) and in a variety of compound heterozygous states such as sickle cell/hemoglobin C disease and hemoglobin C/β thalassemia.

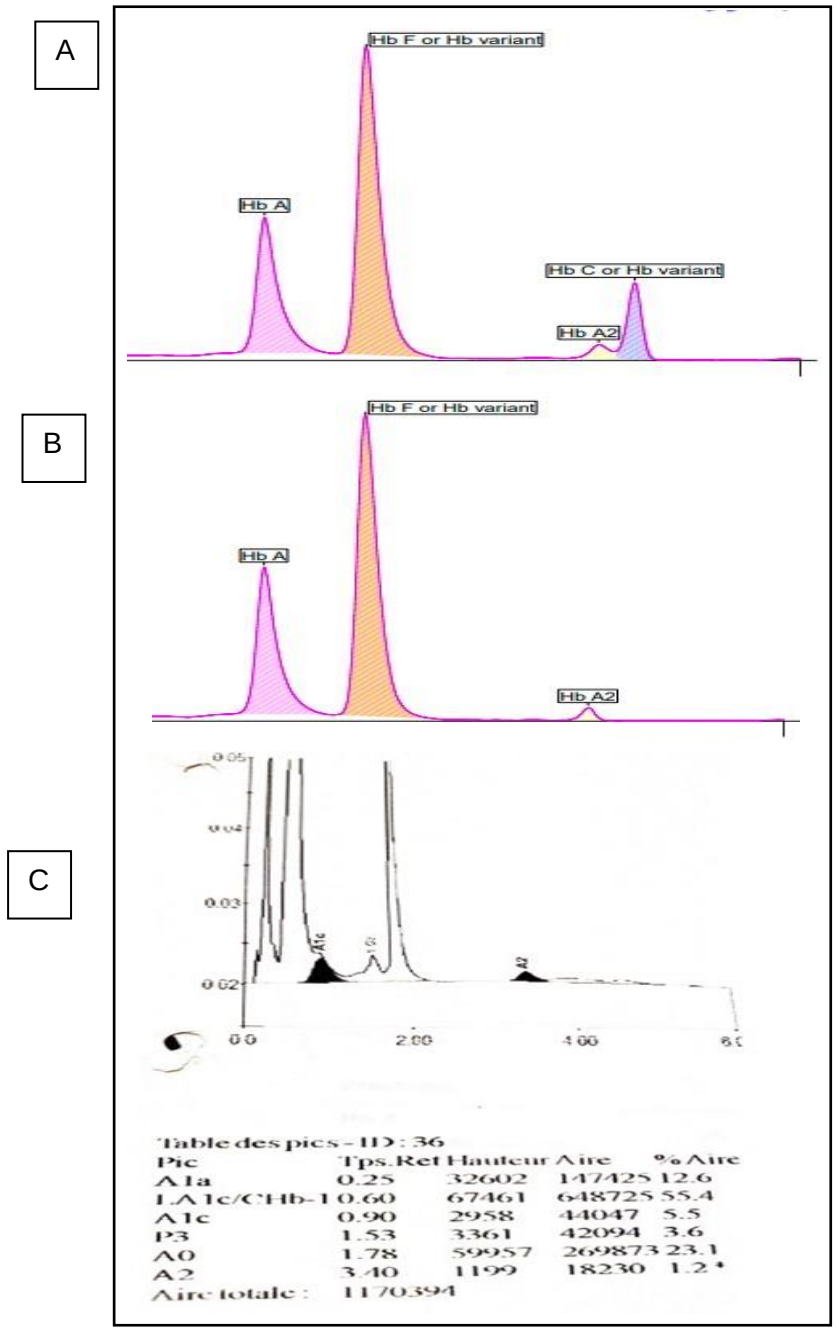

Figure (1): Hb fractions in HPLC and capillary electrophoresis of propositus. (A) at presentation. (B) and (C) 6 months later.

Hb C is present with a significant incidence in north Africa and with approximately 1-10% in Algéria.(6) However it should be noted that some early reports of the presence of hemoglobin C, based only on electrophoresis at alkaline pH, may have been a misidentification of hemoglobin O-Arab as hemoglobin C.

Hemoglobin C trait is the heterozygous condition with no clinical significance. Hemoglobin electrophoresis, HPLC and capillary electrophoresis show hemoglobin

A (Hb A) to constitute somewhat more than 50% of hemoglobin and hemoglobin C (Hb C) slightly less.

The proportion of hemoglobin C is lower in those with coexisting α thalassemia trait.
Huisman *et al*, reported a mean percentage of hemoglobin C (with Hb A2 fraction) around 44%, in patients thought likely to have four α genes, around 37.5% in those thought likely to have three α genes and around 32% in those likely to have two α genes (7). When Couto *et al,* reported a mean of 37% and 32% in those with four and three α genes, respectively.(8)
In hemoglobin C disease, the pathological gene β^c is in the homozygous state. This is why, hemoglobin C is about 95% of total hemoglobin. Clinically the patient presents a mild, chronic hemolytic anemia.
In the case of heterozygous composite hemoglobin C/β thalassemia there is, generally, a moderately severe anemia with splenomegaly, the clinical picture can resemble thalassemia intermedia, if the Hb C is coinherited with β0 thalassemia or severe β+thalassemia.

The occurrence of low percentage of variant hemoglobin with transfusion creates a diagnostic dilemma, because it becomes not easy to make the difference between this condition and the dilutional effects of transfusion on the variant hemoglobin in the recipient.
There are two ways to distinguish between these two different diagnoses. The first, can be done by the identification of the variant hemoglobin in the transfuse red blood cell units, or directly in the blood donor. However, when it isn't possible, the second way is to prove that hemoglobin variant decreases or disappears with time or was absent in previous results in the transfused patient blood.
Finally, family study can help in ruling out a true hemoglobinopathy in such cases.

Table (1): Detailed hematological profile of both propositus, parents and donor.

Parameters	**Propositus (at presentation)**	**Father**	**Mother**	**Donor**	**Propositus (6mounths later)**
Age	15 mounths	32years	27years	54years	21 mounths
RBC count (10^6/µl)	3.57	6.64	5.04	5.13	3
Hb (g/dL)	9. 3	12.7	11.2	10.7	8.5
MCV (fl)	76.7	64.2	72.1	67.1	81.3
MCH (pg)	26	19.2	22.2	20.8	28.3
MCHC (g/dl)	33.9	29.9	30.8	31	34.7
Capillary electro-phoresis	Hb A=26.4% Hb F=61.4% Hb A2=2.6% **Hb C=9.6%**	HbA=94.8% HbA2=5.2% No Hb C	HbA=91.2% HbA2=5.2% HbF=3.6% No Hb C	Hb C=90.8% Hb F=3.2% Hb A2=6%	Hb A=31.7% Hb F=66.5% Hb A2=1.8% No Hb C
HPLC	ND	ND	ND	ND	Hb A=29% Hb F=69.4% Hb A2=1.6% No Hb C

In our case we report Hb C at 9.6%, in a Beta-thalassemic major patient
with no Hb C in prior hemoglobin capillary electrophoresis and in both hemoglobin HPLC and capillary months later. **(Figure 1)** The low concentration of the Hb variant (Hb C), the hemoglobin patterns of both parents and blood donor confirm the transfusion origin of the Hb C.

It's important to diagnose the transfusion-acquired hemoglobinopathies to avoid misdiagnosis, as reported by Wong *et al.* Where a diagnosis of heterozygous beta-thalassemia was made in a normal individual after transfusion with blood of a donor carrying Hb E.(9)

Many authors reported the occurrence of transfusion-induced hemoglobinopathies. The largest series was described by Kazarski *et al.* with 52 transfusion associated peaks in 32 recipients, where Hb C was the most frequent hemoglobin variant (46/52), present at a percentage between 1.1 and 14% of all hemoglobin, Hb S was described
in 4 recipients at 1.4 to 7.1%, and Hb 0-Arab detected in 2 recipients with respectively 0.8 and 3.4%.(1) In the case reported by Suarez *et al.* the concentration of the acquired Hb C was 10.9%.(10)
Many others hemoglobin variants induced by transfusion were described: like the three cases of Hb S, ranged from 9.9 to 18%, that were described by Gupta *et al*. in their study, of 1530 Hb HPLC.(11)

10.2% of Hb D were detected in a case of post-transfusion peak in a juvenile myelomonocytic leukemia by Jain *et al*.(12)
Chauhan *et al*. had reported a case of post-transfusion peak of 9.1% Hb Q India.(13)
In all these observations, we can note that the percentage of the hemoglobin variant induced by transfusion is much lower than in the case of their heterozygous state. Fact that can help in differential diagnosis.

Generally, there is no clinical consequences in the recipient du to transfused Hb C or other hemoglobin variant, it's unlikely to adversely affect transfusion efficacy. Exception is made, for the case of higher Hb S in pediatric patients
which can increase the risk of sickling complications. This can happen easily when the donor is an asymptomatic heterozygous for Hb S. Especially in neonates in whom multiple renal and splenic infarcts following transfusion with sickle trait blood has been reported.(14) Transfused Hb S may also create complications during anesthesia in these patients. (11)
On the other side, blood from sickle trait donors can lead to blockage of leukodepletion filters.(15)
In our case, no complications were seen due to transfused Hb C in the recipient. Indeed, there was no need to sooner transfusion.

Habitually, transfusion-induced hemoglobinopathy occurs when the donor is an asymptomatic, generally, heterozygous for a hemoglobin variant with no significant abnormalities donates blood, escaping to the blood donor screening process, especially when blood bank criteria for donor selection do not include any screening tests for detection of such carriers. In our case, the donor was heterozygous compound Hb C/β^0 thalassemia, with sub-icterus and no need to transfusion (Hb=10.7g/dl), who has escaped to the donor screening process, probably, because of his dark complexion. The donor has been summoned; explanations were given about his pathology, and his definitive exclusion from blood donation. He was, also, referred to a hematologist for management.

To avoid this problem some authors have suggested screening of blood donors for hemoglobinopathies.(16) This will increase the cost of biological qualification of blood donation, independent on the fact that it is a relatively rare problem. It may have more benefit, in populations with an important frequency of hemoglobinopathies, especially those with no systematic screening program. Like in our case, many donors are not aware that they are carriers of a hemoglobinopathy.

Conclusion

With this case, we want to highlight that transfusion-induced hemoglobinopathy should be considered in the differential diagnosis of patients having unusually low levels of the variant hemoglobin.
The good interpretation of a hemoglobin profile depends on preanalytical conditions, with an important place for the information
sheet which must report all the transfusion history of patient.
It is recommended to avoid prescribing Hb-HPLC or capillary electrophoresis in a recent transfused patient and in unavoidable situations, performing the chromatography along with parental study can prevent such diagnostic dilemmas.

References

1. Kozarski TB, Howanitz PJ, Howanitz JH, Lilic N, Chauhan YS. Blood transfusions leading to apparent hemoglobin C, S, and O-Arab hemoglobinopathies. Arch Pathol Lab Med. déc 2006;130(12):1830-3.
2. Gupta SK, Sharma M, Tyagi S, Pati HP. Transfusion-induced hemoglobinopathy in patients of beta-thalassemia major. Indian J Pathol Microbiol. 2011;54(3):609-11.
3. Cook CM, Smeltzer MP, Mortier NA, Kirk SE, Despotovic JM, Ware RE, et al. The Clinical and Laboratory Spectrum of Hb C [β6(A3)Glu→Lys, *G* AG> *A* AG] Disease. Hemoglobin. févr 2013;37(1):16-25.
4. Itano HA, Neel JV. A New Inherited Abnormality of Human Hemoglobin. Proc Natl Acad Sci U S A. nov 1950;36(11):613-7.
5. Kaplan E, Zuelzer WW, Neel JV. A new inherited abnormality of hemoglobin and its interaction with sickle cell hemoglobin. Blood. déc 1951;6(12):1240-9.
6. Uddin DE, Dickson LG, Brodine CE. Screening of military recruits for hemoglobin variants. JAMA. 25 mars 1974;227(12):1405-7.
7. Huisman TH. Trimodality in the percentages of beta chain variants in heterozygotes: the effect of the number of active Hbalpha structural loci. Hemoglobin. 1977;1(4):349-82.

8. Couto FD, De Albuquerque ABL, Adorno EV, De Moura Neto JP, De Freitas Abbehusen L, De Oliveira JLB, et al. alpha-Thalassemia 2, 3.7 kb deletion and hemoglobin AC heterozygosity in pregnancy: a molecular and hematological analysis. Clin Lab Haematol. févr 2003;25(1):29-34.
9. Wong P, Tapprom A, Jermnim N, Charoenporn P, Kanthiyawong S. A blood transfusion leading to misdiagnosis of beta-thalassaemia carrier status. Blood Transfus Trasfus Sangue. janv 2010;8(1):69-70.
10. Suarez AA, Polski JM, Grossman BJ, Johnston MF. Blood transfusion-acquired hemoglobin C. Arch Pathol Lab Med. juill 1999;123(7):642-3.
11. Gupta S, Sharma M, Tyagi S, Pati H. Transfusion-induced hemoglobinopathy in patients of beta-thalassemia major. Indian J Pathol Microbiol. 2011;54(3):609.
12. Jain S, Dass J, Pati HP. Transfusion associated peak in hb HPLC chromatogram - a case report. Mediterr J Hematol Infect Dis. 2012;4(1):e2012006.
13. Sharma S, Chandra J, Chauhan R, Rai P. Transfusion associated unknown peak on HPLC chromatogram: A diagnostic dilemma. J Appl Hematol. 2014;5(1):19.
14. Novak RW, Brown RE. Multiple renal and splenic infarctions in a neonate following transfusion with sickle trait blood. Clin Pediatr (Phila). avr 1982;21(4):239-41.
15. Beard MJ, Cardigan R, Seghatchian J, Krailadsiri P, Williamson LM. Variables determining blockage of WBC-depleting filters by Hb sickle cell trait donations. Transfusion (Paris). mars 2004;44(3):422-30.
16. Lippi G, Mercadanti M, Alberta C, Franchini M. An unusual case of a spurious, transfusion-acquired haemoglobin S. Blood Transfus Trasfus Sangue. juill 2010;8(3):199-202.