Case Reports

Transfusion Associated Peak in Hb HPLC Chromatogram – a Case Report

Sonal Jain, Jasmita Dass and Hara Prasad Pati

Department of Hematology, All India Institute of Medical Sciences, New Delhi

Correspondence to: Dr. Jasmita Dass, M.D. Department of Hematology, All India Institute of Medical Sciences, New Delhi. Tel: 9999347544, Fax: 01126588663. E-mail: drjasmita@gmail.com

Competing interests: The authors have declared that no competing interests exist.

Abstract. High performance liquid chromatography (HPLC) and electrophoresis are commonly used to diagnose various hemoglobinopathies. However, insufficient information about the transfusion history can lead to unexpected and confusing results. We are reporting a case of Juvenile myelomonocytic leukemia (JMML) in which HbHPLC was done to quantify fetal hemoglobin (HbF). The chromatogram showed elevated HbF along with a peak in the HbD window. A transfusion acquired peak was suspected based on the unexpectedly low percentage of HbD and was subsequently confirmed using parental HbHPLC.

Introduction. It is well known that incomplete history on test request forms sent to laboratories and inappropriate patient samples can lead to wrong diagnosis and hazardous consequences. Hemoglobin electrophoresis and High Performance Liquid Chromatography (HPLC) are routinely done to diagnose and classify hemoglobinopathies. Acquired and inherited conditions in which abnormal HPLC result can be seen include high fetal hemoglobin in Juvenile Myelomonocytic Leukemia (JMML), Diamond Blackfan Anemia (DBA) and Fanconi Anemia. Blood transfusion from donors with hemoglobinopathies which are clinically silent (e.g. HbE, HbD, HbS) may lead to abnormal peaks or altered percentages of abnormal hemoglobins.

Case. A 7 year old female presented to the outpatient department with complaints of hepatosplenomegaly and lymphadenopathy for 3 months. Hemogram showed a hemoglobin of 9.5 gm/dl, total leukocyte count of 57.24 X10⁹/L and platelet count of 16 X10⁹/L. Peripheral smear showed 35% monocytes with an absolute monocyte count of 20 X10⁹/L. Diagnosis of Juvenile myelomonocytic leukemia was suggested and Hb HPLC was advised to look for increased fetal haemoglobin. High Performance Liquid Chromatography (HPLC) was done using BioRad Variant II instrument with beta thalassemia short program. Hb HPLC chromatogram showed a raised HbF (11%) with HbA₀ 69.2%, HbA₂ 1.8% and a peak in D-window of 10.2% with a retention time of 4.06 minutes (Figure 1). The expected percentage of HbD in heterozygotes is ~40% but in our case it was significantly low. Hence, a possibility of transfusion acquired HbD was considered and Hb HPLC was done in both the parents. Both mother and father showed normal Hb HPLC (Figure 2A and 2B). The transfusion history was taken and it was discovered that the patient had received 3 units of PRBC one week prior to HPLC. Thus a final diagnosis of transfusion
associated peak in D window was made.

There are only a few reports of abnormal

References:

1. Teo JT, Klaassen R, Fernandez CV, Yanofsky R, Wu J, Champagne J et al. Clinical and genetic analysis of unclassifiable inherited bone marrow failure syndromes. Pediatrics 2008;122;e139-e148. [http://dx.doi.org/10.1542/peds.2007 -3415 PMid:18595958

2. Lippi G, Mercandanti M, Alberta C, Franchini M. An unusual case of a spurious, transfusion-acquired haemoglobin S. Blood Transfus 2010;8:199-202. PMid:20671882 PMcid:2906190

3. Suarez AA, Polski JM, Grossan BJ, Johnston MF M. Blood Transfusion–Acquired Hemoglobin C: A Case Report and Review of the Literature. Arch Pathol Lab Med. 1999;123:642–643. PMid:10388925

4. 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. 2006;130:1830–1833. PMid:17149958

5. Wong P, Tapprom A, Jermnim N, Charoenporn, Kanthiyawong S. A blood transfusion leading to misdiagnosis of beta-thalassaemia carrier status. Blood Transfus 2010; 8:69-70. PMid:20104282 PMcid:2809515

Figure 1. Hb HPLC of patient showing high HbF along with a peak in D window (RT – 4.06 minutes).

Figure 2. Hb HPLC of father (2A) and mother (2B) showing normal chromatogram.