American Journal of
Health, Medicine and Nursing Practice
(AJHMN)

Common Clinical Manifestations and a Rare Diagnosis: A Case Report of Hemoglobin Köln in Saudi

Abeer A. Alzahrani, Afnan A. Malibari, Khalid A. Abdalla, Eman A. Khan, and Tariq G. Alasaad

AJP

American Journal of Health, Medicine and Nursing Practice
ISSN 2520-4017 (Online)
Vol.8, Issue 2, pp 19 – 26, 2023

www.ajpojournals.org

Common Clinical Manifestations and a Rare Diagnosis: A Case Report of Hemoglobin Köln in Saudi

Abeer A. Alzahrani[1], Afnan A. Malibari[1*], Khalid A. Abdalla[2], Eman A. Khan[3], and Tariq G. Alasaad[2]

[1]General Pediatric Resident, King Abdulaziz Medical City, WR, Ministry of National Guard, Jeddah, Saudi Arabia

[2]Consultant of Pediatric Hematology and oncology Prince Norah Oncology Center, Ministry of the National Guard Jeddah, Saudi Arabia

[3]Hematopathology Department, King Abdulaziz Medical City, WR, Ministry of National Guard, Jeddah, Saudi Arabia

[*]Corresponding Author's Email: afnan.a.malibari@gmail.com

Article history

Submitted 12.03.23; Revised Version Received 25.03.23; Accepted 04.04.23

Abstract

Background: Hemoglobin Köln is the most widely diagnosed among unstable hemoglobin. Patients with hemoglobin Köln often have moderate hemolytic anemia, reticulocytosis, splenomegaly, and high levels of lactate dehydrogenase and bilirubin in the blood. Hemoglobin Köln happened as a result of the substitution of an amino acid methionine for the usual valine at position 98 of the beta chain. Up to the time, there were no previous reports in Arabian Peninsula, hence, the patient involved in this study is considered to be the first reported case.

Case: A 13 years old Saudi male, product of consanguineous marriage, known to have chronic hemolytic anemia and jaundice since birth. Previously labeled as query Gilbert syndrome and treated with folic acid supplements by primary health care. However, due to the several emergency room admissions caused by severe left upper quadrant abdominal pain in association with dark urine, gallstones and hepatosplenomegaly, the patient was referred to pediatric hematology. Subsequent investigations identified that patient's vague and common complaints was a result of hemoglobin Köln disease. Those investigations showed mild macrocytic anemia, polychromasia, and bite cells in blood smear, while in hemoglobin electrophoresis, a small peak in zone (E) was observed. Hemoglobin Köln was confirmed by whole exome sequencing (WES) test that reported the presence of amino acid exchange (Val99Met).

Recommendations: Careful examination is necessary for the differential diagnosis of hemoglobinopathy variations, which can be challenging especially in pediatric cases. Unstable hemoglobin should be investigated in cases mimicking a β-thalassemia trait, but associated with abnormally rapid hemolysis and reticulocytosis, or in cases that cannot be explained with common causes.

Keywords: *Unstable hemoglobin, Hb Köln, Saudi Arabia*

1.0 INTRODUCTION

In many countries, hemoglobinopathies are regarded as a serious public health issue. This is understandable given their prevalence, genetic variety, and clinical significance. When the amino acid valine is substituted for methionine at position 98 of hemoglobin's Beta chain, Hgb Köln is produce. An autosomal dominant inheritance pattern characterizes Hgb Köln. Some case report described Hgb Köln disease as a de novo or fresh mutations, implying there is no proof that the parents or other family members have unstable hemoglobin.[1] Patients with Hgb Köln typically have reticulocytosis, splenomegaly, bilirubin, jaundice, and prolonged dark urine excretion in addition to moderate hemolytic anemia.[2] Prolonged thrombocytopenia or priapism were also reported in certain cases.[3,4] Hemoglobin Köln reports are more common among several regions of the Orient, and throughout Europe. Recently, more cases form India, South Africa, Brazil, Spain and Turkey were reported as well. [5,7] Yet, no cases were reported among Arabic countries or Gulf region. Hence, our case is considered to be the first case of hemoglobin Köln reported from Saudi Arabia.

2.0 CASE PRESENTATION

The patient is a 13 years old Saudi male, known to have chronic hemolytic anemia and jaundice. He presented to the emergency room, complaining of left upper quadrant abdominal pain that has been on and off for a while but remained constant for the past 3 days. The pain was colicky in nature, accompanied by two episodes of vomiting and subjective fever, dizziness and dark urine. There was no history of anemia symptoms, or bleeding from the gums or nasal mucosa or stool. This was not the first time the patient present with this complaint, as he had previous similar episodes.

The patient is a product of uncomplicated spontaneous vaginal delivery. As a neonate, the patient developed jaundice requiring 3 days of phototherapy. He was breastfed only for 4 months and his development was similar to his siblings except for noticeable poor weight gain. At the age of seven, he started to have obvious jaundice in the skin again, along with episodes of the symptoms mentioned earlier. Consequently, he was labelled as query Gilbert syndrome by primary care and was started on folic acid tablets. The patient had no known history of any other chronic illnesses. Moreover, there was no history of previous surgeries, and he never needed a blood transfusion or an intensive care unit admission. The patient is fully immunized, does not have allergies, and was not on medications at that time.

When family history was acquired, it was found out that the patient is a product of 1st degree consanguinity. The father was diagnosed with B-thalassemia and has a history of recurrent malignancies. He had undergone splenectomy and received blood transfusion during it. All four patient's sisters had jaundice since birth, only two of them were diagnosed with B-thalassemia. Multiple relatives on the paternal side -including the grandmother and uncles- also had a positive history of chronic jaundice and some were diagnosed with B-thalassemia**.** These relatives also had splenectomy and were treated with folic acid supplement.

On examination, the patient was vitally stable, mild dehydration and scleral jaundice were noted, and abdomen palpating showed both the epigastric and left upper quadrant regions tenderness. Hepatosplenomegaly was found; the spleen measured 8 cm below costal margin, while the liver 1.5 cm below costal margin with a span of 14 cm.

Initial investigations were conducted in emergency department included CBC and bilirubin levels as shown in Table 1. Coagulation profile, liver function test, renal function, viral serology, and chest x-ray all yield unremarkable results. Urine analysis was negative along with urine and blood cultures that showed no growth of any organisms. Abdominal ultrasound concluded that the liver was 15.1 cm, Spleen was 17.3cm. The gallbladder was distended with multiple mobile sludge and stones; however, no sonographic evidence of cholecystitis. Therefore, the patient was managed with pain control and hydration, and then was referred to pediatric hematology for further investigation and management.

Table 1: Patient's initial investigations

Test	Result	Normal range
WBC	8.2	4.5 to 11.0 × 10^9/L
Mono	0.55	0.2 – 0.8 × 10^9/L
Neut	6.78	2 – 7.50 × 10^9/L
Lymph	0.81	1.5 – 4 × 10^9/L
RBC	4.4	4.5 – 6.5 × 10^{12}/L
Hgb	12.1	13 – 18 g/dL
Hct	43.2	30 – 44 %
MCV	98.0	75 – 96 fL
MCH	27.4	27 – 32 pg
MCHC	28.0	32 – 36 g/dL
RDW	18.0	11.5 – 14.5%
Platelet	176	150 – 450 × 10^9/L
MPV	8.15	8-12 fL
Retic percent	9.47	0.20 – 2 %
Absolut Retic	416.68	20 – 100 × 10^9/L
Direct Bilirubin	10.6	1.7 – 6.7 umol/L
Total Bilirubin	115.4	1.7 – 11.9 umol/L

LDH	546	130 – 250 U/L
Pyruvate kinase	34.8	5.3 – 17.3 U/g Hb
Direct Coombs test	Negative	

WBC: white blood cell, Mono: monocytes, Neut: neutrophils, Lymph: lymphocytes, RBC: red blood cells, Hgb: hemoglobin, Hct: Hematocrit, MCV: mean corpuscular volume, MCH: mean corpuscular hemoglobin, MCHC: mean corpuscular hemoglobin concentration, MPV: mean platelet volume, Retic: reticulocyte, LDH: lactate dehydrogenase.

Under pediatric hematology, further work up included blood smear, and hemoglobin electrophoresis (Figure 1, Figure 2). Blood smear showed mild macrocytic anemia with polychromasia, echinocytes, bite cells, blisters cells and occasional spherocytes, which might indicate the presence of underlying enzymopathy, unstable hemoglobin or ongoing oxidative hemolysis. Blood film Testing for G6PD, Malaria, as well as direct coombs test were all negative. Pyruvate kinase level was normal (34.8); however, haptoglobin was found to be very low (<.058). Both ferritin and vitamin B12 were normal while folate level was 11.9 with the normal range being ≥ 27 nmol/L. Hemoglobin electrophoresis was conducted twice and six months apart. On both studies, a small peak was found in zone (E), which usually represents hemoglobin E variants. Rarely that peak can also represent another condition which is hemoglobin Köln. The patient was thought to possibly have Hgb E trait; however, it was excluded as a differential diagnosis in this case due to low levels of Hgb E variant with no history of recent blood transfusion.

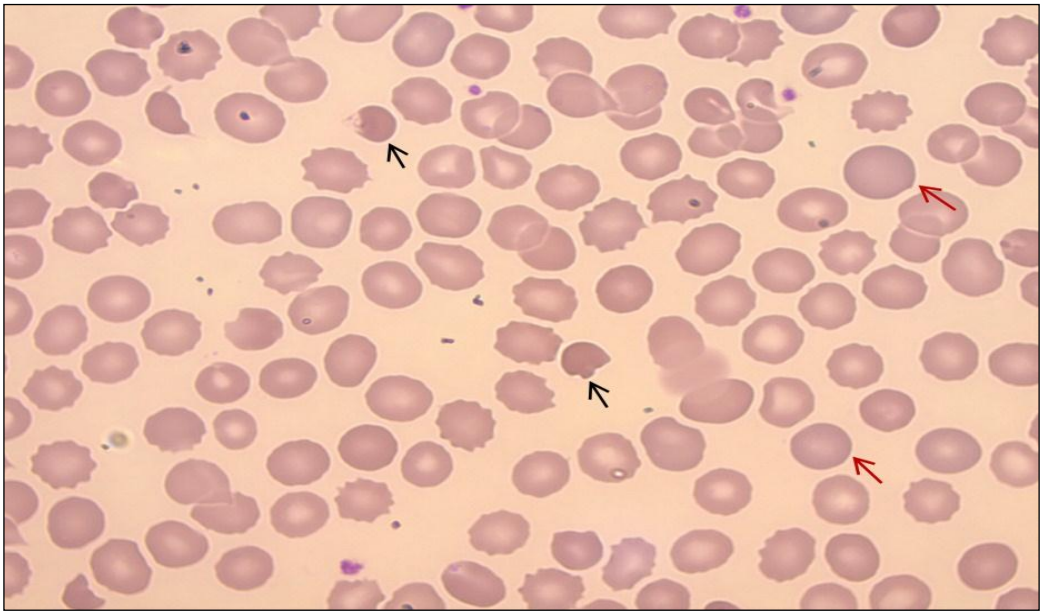
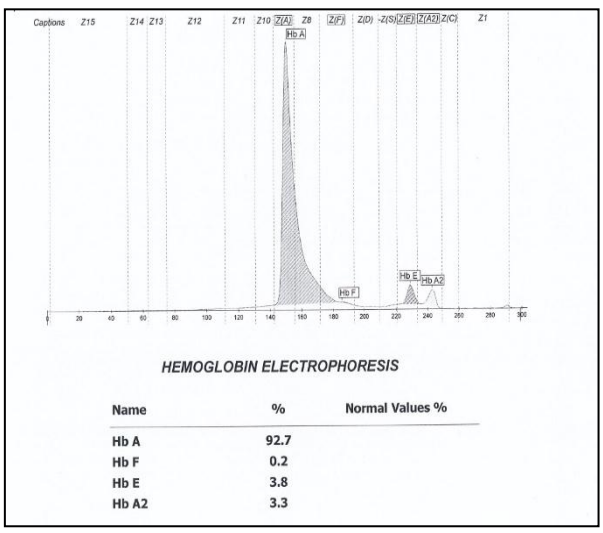
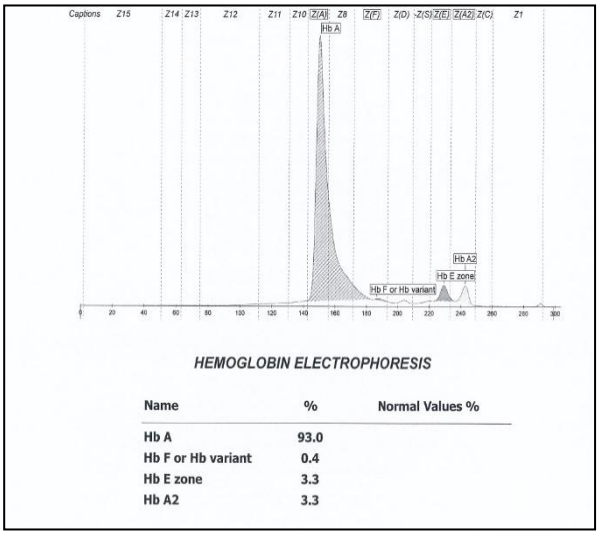


Figure 1: Blood smear shows polychromasia (red arrows) with frequent irregularly contracted cells and keratocytes or bite cells (black arrows).

American Journal of Health, Medicine and Nursing Practice
ISSN 2520-4017 (Online)
Vol.8, Issue 2, pp 19 – 26, 2023
www.ajpojournals.org



A



B

Figure 2: Two capillary electrophoresis were done 6 months apart, both revealed the same findings of a very small peak appears in zone (E) which is the zone where Hemoglobin E variant usually runs. Other less common hemoglobin variants can appear in the same zone including Hgb Köln. Due to its instability, Hgb Köln is usually found in a very low percentage and typically seen as a tiny peak running in zone E as demonstrated in the figure above. After excluding recent history of blood transfusion, the very low percentage of Hgb variant in this case helped us exclude HgbE trait which usually exhibits higher percentage.

Investigations were also done for the family including: CBC, hemoglobin electrophoresis, and blood smear. The mother's electrophoresis was normal. However, for both sisters the electrophoresis showed the following Hgb A 92.1, Hgb A2 Qua 3.6, Hgb F Qua 0.8, Hgb E 3.8. This demonstrated a similar pattern to our indexed case, however further genetic testing for the family awaits. Finally, genetic testing was a necessity to reach the diagnosis. Thus, whole exome sequencing (WES) test was sent. The result identified the heterozygous variant c.295 G>A, p.(Val99Met) in HBB (OMIM:141900) which leads to an amino acid exchange. 9 out of 9 bioinformatic in silico programs predict a pathogenic effect for this variant. Considering the heterozygous pathogenic variant in BB and the supportive phenotype of the patient a genetic diagnosis of Heinz body anemia (Hgb Köln) is most likely. Regarding the management, the patient was largely on supportive measurements i.e. folic acid supplements. Later on, with the progression of the disease, he required cholecystectomy and splenectomy.

3.0 DISCUSSION

Disorders that affect the formation of the hemoglobin chain are known as hemoglobinopathies. They can be classified as either qualitative (structural Hb variants) or quantitative diseases (thalassemia syndromes). A change in the amino acid sequence in the alpha (α)- or beta (β)-chains causes structural variations in the hemoglobin, which impact its properties including

stability, solubility, and O2 affinity.

Even though there are more than two hundred unstable variations discovered, less than half of them result in clinical disease, making unstable hemoglobin disease (UHD) a rare and hardly spotted condition with an unknown prevalence. The most common of them is hemoglobin Köln (β98 [FG5]Val > Met), one of the first unstable hemoglobins to be described, which has been reported in thirty four cases so far since the original report in 1961.[8] The methylated CpG dinucleotide regions of the b globin gene can operate as "hotspots" for mutation through the deamination of the methylcytosine nucleotide to create thymine, which is the hypothesized reason for the numerous unrelated occurrences of this unstable hemoglobin.[9] Genetic testing is therefore the most conclusive test for making the diagnosis and describing the variation.

Hemoglobin Köln can be detected at a variety of ages. Many with moderate, compensated hemolysis will not seek medical treatment until later in life or if a precipitating event occurs such a severe infections or exposure to oxidizing drugs. While in some cases including ours, the diagnosis is made during childhood.[10] The distinguishing manifestations of hemoglobin Köln include congenital nonspherocytic hemolytic anemia, splenomegaly, pigmented (bilirubin) gallstones, and in severe cases, pigmenturia. As supported by our case findings, the affected individuals typically have low haptoglobin, high lactate dehydrogenase, and negative direct antiglobulin (Coombs) test findings since there is no immunological component. White blood cell and platelet counts are usually normal but can occasionally be high. The RBCs in the peripheral blood smear might include bite cells, spherocytes, or other abnormal RBC morphologies, and they could be hypochromic, giving the sample a thalassemia-like appearance. Due to the existence of small spherocytes and huge reticulocytes that are out of proportion to the hemoglobin level, a normocytic anemia with an enlarged red cell distribution width can also be noticed.[11]

Hemolytic anemia can result in the production of Heinz bodies, however this is not a requirement for the diagnosis of unstable hemoglobin. It's important to note that these abnormalities found on the peripheral blood smear can occur with a number of different hemolysis-related disorders in addition to unstable hemoglobins.[8] Due to the functional characteristics of hemoglobin Köln, such as its strong affinity for oxygen in the absence of compensatory erythrocytosis and ease of heme dissociation, abnormally low hemoglobin oxygen saturation by pulse oximetry has been reported.[13] For hemoglobin Köln, homozygosity has never been described. Rarely have compound heterozygotes been reported. There has only been one case of a patient who had both beta thalassemia and Hb Köln, and this patient only had mild hemolytic anemia.[14] The clinical severity of the condition is influenced by the co-inheritance of another hemoglobin variant, most frequently the b-thalssemia trait.

The patient's management is mostly supportive and includes avoiding oxidizing medications wherever feasible, preventing infections, treating them immediately, and prescribing folic acid supplements.[15] Splenectomy plays a role in patients with severe hemolysis as it will reduce the

frequency of transfusions required. However, it poses a possible risk of thrombo-embolism in these patients.[16]

4.0 CONCLUSION AND RECOMMENDATIONS

Given the fact that the initial symptoms overlaps with that of common hemolytic anemia syndromes, diagnosing hemoglobin Köln can be challenging for clinicians. This in turn renders many cases undiagnosed. As seen in our case, the patient was unnoticed and undiagnosed for years despite his recurrent complaints. Afterwards he was seen and investigated, yet mislabeled as Gilbert syndrome and hemoglobin E trait before the actual diagnosis of unstable hemoglobin was considered and reached. This sheds the light on the importance of in-depth comprehension of the variations in symptoms and analysis in cases with congenital nonspherocytic hemolytic anemia that cannot be explained by more common causes such as thalassemia, sickle cell disease, red blood cell (RBC) enzymopathies, or immune-related hemolysis. In such cases, unstable hemoglobin should be kept in mind and investigated. Furthermore, investigating Hb Köln in the patient and other family members is crucial to avoid the consequences of severe hemolytic anemia later in life including the need for splenectomy.

REFERENCES

1. Miller DR, Weed RI, Stamatoyannopoulos G, Yoshida A. Hemoglobin Köln disease occurring as a fresh mutation: erythrocyte metabolism and survival. *Blood.* 1971;38(6):715–729.

2. Pscheidt S, Pereima IML, Roberge MB, Rotolo MAS, Genovez G, Zunino JN. Köln's unstable hemoglobin: case report and literature review Hemoglobina instável de Köln: relato de caso e revisão de literatura. *J Bras Patol e Med Lab.* 2003;39(1):37–40.

3. Andrieu V, Dumonceau O, Grange MJ. Priapism in a patient with unstable hemoglobin: Hemoglobin Köln. *Am J Hematol.* 2003;74(1):73–74.

4. Cheung VYT, Silverman JA. Hemoglobin Köln and pregnancy. *J Obstet Gynaecol Canada*. 2008;30(10):907–909. doi:10.1016/S1701-2163(16)32971-1

5. Rios JO, Pereira LR, Bonini-Domingos CR. New report of Köln hemoglobin from countryside/interior of São Paulo. *Hematol Transfus Cell Ther*. 2021;43:S25. doi:10.1016/j.htct.2021.10.043

6. Warang P, Nair S, Nadkarni A, et al. Hb Köln [β98(FG5) [GTG → ATG, Val → Met]: The first report from India. Hematology [Internet]. 2014;19(4):199–201. doi:10.1179/1607845413Y.0000000116

7. Bird AR, Karabus CD, Hartley PS, Lehmann H. Haemoglobin Köln in Cape Town. *South African Med J.* 1987;72(2):154–156.

8. Lomas J, Anderson GM, Domnick-Pierre K, Vayda E, Enkin MW. Postsplenectomy erythrocytosis in Hemoglobin Köln disease. *N Engl J Med.* 1989;321(19):1306–1311.

9. Williamson D. The unstable haemoglobins. *Blood Rev.* 1993;7(3):146–163.

10. Zur B. Hemoglobin variants-pathomechanism, symptoms and diagnosis. *LaboratoriumsMedizin.* 2016;39(5).

11. Zinkham WH, Winslow RM. Unstable hemoglobins: influence of environment on phenotypic expression of a genetic disorder. *Medicine (Baltimore).* 1989 Sep;68(5):309–320.

12. Milner PF, Wrightstone RN. The unstable hemoglobins: a review. *Prog Clin Biol Res.* 1981;51:197–222.

13. de Furia FG, Miller DR. Oxygen affinity in Hemoglobin Köln disease. *Blood.* 1972;39(3):398–406.

14. Mittal N, Garg A, Kashyap R, Agarwal S. A case of compound heterozygous hemoglobin Köln/hemoglobin E in an Indian family. *Pediatr Hematol Oncol*. 2019;36(6):394–398. doi:10.1080/08880018.2019.1648620

15. White JM. The clinical features of unstable haemoglobin disease. J Clin Pathol Suppl (R Coll Pathol). 1974;8:19–22.

16. Crary SE, Buchanan GR. Vascular complications after splenectomy for hematologic disorders. *Blood.* 2009;114(14):2861–2868.