

Anticoagulant induced leukoagglutination

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ABSTRACT

نقص خلايا الدم البيضاء بسبب تكتلها، والناج عن المادة المانعة لتجلط داخل الأنبوبة الخزامية (EDTA)، يحدث في الأمراض الحميدة والأمراض الخبيثة. هذه الحالة لمريض يبلغ من العمر 71 عاماً، تم تنويمه في المستشفى بسبب التهاب حاد في الصدر، وخلال فترة التنويم تم فحص المريض وعمل التحاليل اللازمة ومنها: صورة الدم لعينة دم تم سحبها في الأنبوبة الخزامية (EDTA) وتحليلها بجهاز سيسميكس (SE/9500) الذي أظهر أن عدد خلايا الدم البيضاء $8.2 \times 10^9/L$ ، وعدد خلايا الدم المتعادلة هو $4.8 \times 10^9/L$ ، وفحص شريحة الدم تحت الميكروسكوب أوضح وجود تكتل لخلايا الدم البيضاء المتعادلة. عند فحص عينة دم، جمعت في الأنبوبة المحتوية على مادة السيتريت المانعة لتجلط بنفس الجهاز، كان عدد خلايا الدم البيضاء $11.8 \times 10^9/L$ ، وهذا يعني أن نقص خلايا الدم البيضاء بسبب تكتلها وتجمعها الناتج عن المادة المانعة لتجلط في الأنبوبة الخزامية.

Low leukocyte count secondary to leukocyte aggregation caused by an ethylene diamine tetra acetic acid (EDTA) occur in both benign and malignant disorders. We report a 71-year-old male patient who was admitted to the hospital with acute chest infection. Complete blood count (CBC) collected in EDTA tube and analyzed by sysmex instrument (SE/9500) revealed low hemoglobin level of 9.4g/dl, white blood cell (WBC) count of $8.2 \times 10^9/L$ and neutrophils of $4.8 \times 10^9/L$. Peripheral blood smear review shows multiple leukocytes aggregation (one clump in each field). When we ask for another blood sample in citrate anticoagulant, the CBC showed WBC count of $11.8 \times 10^9/L$ and neutrophils of $6.26 \times 10^9/L$. This is a case of low leukocyte count secondary to leukocyte aggregation induced by EDTA.

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In vitro leukocyte aggregation is very rare. Ethylene diamine tetra acetic acid (EDTA) induced clumping of lymphoid cells, both benign and malignant, in peripheral blood samples has been reported only rarely. A case of low-grade B-cell splenic lymphoma is present with lymphocyte clumping in smear made from EDTA anticoagulated peripheral blood.¹ Two patients are described with spurious leukopenia secondary to in vitro aggregation of neutrophils. In one of this patient, the hematologist discovered multiple white blood cell (WBC) aggregates at the edges of a smear made from an EDTA-anticoagulated blood specimen.² The WBC count may be spuriously decreased (pseudoleukopenia) resulting from leukoagglutination and has been reported cirrhotic patient with liver failure and also in uremic and immunosuppressed patients whose blood was analyzed on coulter instruments.³ Although the condition seems to be very rare but very important to be detected as if such artifact occurred in case of chronic myeloid leukemia, or chronic lymphocytic leukemia, on treatment, the doubling time, and monitoring the response to chemotherapy could be seriously affected.

Case Report. A 71-year-old Saudi male patient presented in March 2007 with 2 days' history of fever, productive cough, and chest pain associated with rigor and sweating. His past medical history is unremarkable. On examination, the patient looked sick, and pyrexial. There was no lymphadenopathy or organomegaly. X-ray revealed white patches in both lungs. Sputum culture was negative. Coagulation profile, renal function, and liver function were normal. Complete blood count revealed a hemoglobin of 9.4 g/dl (normal 13.0-17.8), WBC count of $8.2 \times 10^9/L$ (4.0-11.0), platelet count of $232 \times 10^9/L$, neutrophils $4.86 \times 10^9/L$, and lymphocytes $2.03 \times 10^9/L$. Blood smear made from an EDTA anticoagulated blood specimen revealed normocytic normochromic anemia and multiple neutrophil aggregates (Figure 1). A blood sample collected in citrate anticoagulant was requested and showed WBC count of $11.8 \times 10^9/L$, and neutrophils of $6.26 \times 10^9/L$. Patient improved with intravenous antibiotic and discharged home from the hospital.

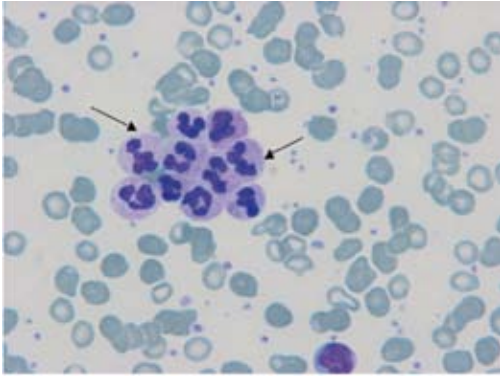


Figure 1 - Peripheral blood smear shows neutrophil aggregation induced by ethylenediamine tetraacetic acid (original magnification x1000).

Discussion. Ethylenediamine tetraacetic acid associated agglutination in peripheral blood is not uncommon for platelets but has also been reported in neutrophils, benign, and malignant lymphocytes.¹ The in vitro leukocyte aggregation secondary to EDTA is a rare phenomenon. The exact mechanism underlying this phenomenon has not been determined. Cold agglutinins (IgM) have been implicated. The IgM antibody is directed

specifically to a granulocyte antigen and depends on the presence of EDTA.⁴ Report describes a case of spurious neutropenia caused by EDTA-dependent in vitro agglutination of neutrophils, after raising the temperature of the sample to 37°C the agglutination was irreversible, but it resolved completely after addition of kanamycin.⁵ This reported case supports the interaction of the IgM antibody with leukocytes in the presence of EDTA as the WBC count performed in citrated anticoagulant increased from $8.2 \times 10^9/L$ to $11.8 \times 10^9/L$ and neutrophils count increased from $4.86 \times 10^9/L$ to $6.2 \times 10^9/L$.

References

1. Shelton JB Jr, Frank IN. Splenic B cell lymphoma with lymphocyte clusters in peripheral blood smears. *J Clin Pathol* 2000; 53: 228-230.
2. Epstein HD, Kruskal MS. Spurious leukopenia due to in vitro granulocyte aggregation. *Am J Clin Pathol* 1988; 89: 652-655.
3. Savage RA. Analytic inaccuracy resulting from hematology specimen characteristics. Three cases of clinically misleading artifacts affecting white blood cell and platelet counts. *Am J Clin Pathol* 1989; 92: 295-299.
4. Salim H, Khalil. EDTA-dependent leukoagglutination. *Clin Lab Haematol* 2002; 24: 67-69.
5. Hoffmann JJ. EDTA-induced pseudo-neutropenia resolved with kanamycin. *Clin Lab Haematol* 2001; 23: 193-196.

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