OPINION REVIEW

4280  Role of monoclonal antibody drugs in the treatment of COVID-19

Ucciferri C, Vecchiet J, Falasca K

MINIREVIEWS

4286  Review of simulation model for education of point-of-care ultrasound using easy-to-make tools

Shin KC, Ha YR, Lee SJ, Ahn JH

4303  Liver injury in COVID-19: A minireview

Zhao JN, Fan Y, Wu SD

ORIGINAL ARTICLE

Case Control Study

4311  Transanal minimally invasive surgery vs endoscopic mucosal resection for rectal benign tumors and rectal carcinoids: A retrospective analysis

Shen JM, Zhao JY, Ye T, Gong LF, Wang HP, Chen WJ, Cai YK

4320  Impact of mTOR gene polymorphisms and gene-tea interaction on susceptibility to tuberculosis


Retrospective Cohort Study

4331  Establishment and validation of a nomogram to predict the risk of ovarian metastasis in gastric cancer: Based on a large cohort

Li SQ, Zhang KC, Li JY, Liang WQ, Gao YH, Qiao Z, Xi HQ, Chen L

Retrospective Study

4342  Predictive factors for early clinical response in community-onset Escherichia coli urinary tract infection and effects of initial antibiotic treatment on early clinical response

Kim YJ, Lee JM, Lee JH

4349  Managing acute appendicitis during the COVID-19 pandemic in Jiaxing, China

Zhou Y, Cen LS

4360  Clinical application of combined detection of SARS-CoV-2-specific antibody and nucleic acid

Meng QB, Peng JJ, Wei X, Yang JY, Li PC, Qu ZW, Xiong YF, Wu GJ, Hu ZM, Yu JC, Su W

4370  Prolonged prothrombin time at admission predicts poor clinical outcome in COVID-19 patients

## Contents

### Semimonthly Volume 8 Number 19 October 6, 2020

<table>
<thead>
<tr>
<th>Page</th>
<th>Title</th>
<th>Authors</th>
</tr>
</thead>
<tbody>
<tr>
<td>4380</td>
<td>Percutaneous radiofrequency ablation is superior to hepatic resection in patients with small hepatocellular carcinoma</td>
<td>Zhang YH, Su B, Sun P, Li RM, Peng XC, Cai J</td>
</tr>
<tr>
<td>4388</td>
<td>Clinical study on the surgical treatment of atypical Lisfranc joint complex injury</td>
<td>Li X, Jia LS, Li A, Xie X, Cui J, Li GL</td>
</tr>
<tr>
<td>4410</td>
<td>Optimal hang time of enteral formula at standard room temperature and high temperature</td>
<td>Lakananurak N, Nalinthassanai N, Suansawang W, Panarat P</td>
</tr>
<tr>
<td>4416</td>
<td>Meta-analysis reveals an association between acute pancreatitis and the risk of pancreatic cancer</td>
<td>Liu J, Wang Y, Yu Y</td>
</tr>
<tr>
<td>4431</td>
<td>Global analysis of daily new COVID-19 cases reveals many static-phase countries including the United States potentially with unstoppable epidemic</td>
<td>Long C, Fu XM, Fu ZF</td>
</tr>
<tr>
<td>4443</td>
<td>Left atrial appendage aneurysm: A case report</td>
<td>Belov DV, Moskalev VI, Garbuzenko DV, Arefyev NO</td>
</tr>
<tr>
<td>4466</td>
<td>Primary rhabdomyosarcoma: An extremely rare and aggressive variant of male breast cancer</td>
<td>Satală CB, Jung I, Bara TJ, Simu P, Simu I, Vlad M, Szodorai R, Gurzu S</td>
</tr>
<tr>
<td>4475</td>
<td>Bladder stones in a closed diverticulum caused by <em>Schistosoma mansoni</em>: A case report</td>
<td>Alkhamees MA</td>
</tr>
<tr>
<td>4481</td>
<td>Cutaneous ciliated cyst on the anterior neck in young women: A case report</td>
<td>Kim YH, Lee J</td>
</tr>
<tr>
<td>4488</td>
<td>Extremely rare case of successful treatment of metastatic ovarian undifferentiated carcinoma with high-dose combination cytotoxic chemotherapy: A case report</td>
<td>Kim HB, Lee HJ, Hong R, Park SG</td>
</tr>
</tbody>
</table>
Acute amnesia during pregnancy due to bilateral fornix infarction: A case report
Cho MJ, Shin DI, Han MK, Yum KS

Ascaris-mimicking common bile duct stone: A case report
Choi SY, Jo HE, Lee YN, Lee JE, Lee MH, Lim S, Yi BH

Eight-year follow-up of locally advanced lymphoepithelioma-like carcinoma at upper urinary tract: A case report
Yang CH, Weng WC, Lin YS, Huang LH, Lu CH, Hsu CY, Ou YC, Tung MC

Spontaneous resolution of idiopathic intestinal obstruction after pneumonia: A case report
Zhang BQ, Dai XY, Ye QY, Chang L, Wang ZW, Li XQ, Li YN

Successful pregnancy after protective hemodialysis for chronic kidney disease: A case report
Wang ML, He YD, Yang HX, Chen Q

Rapid remission of refractory synovitis, acne, pustulosis, hyperostosis, and osteitis syndrome in response to the Janus kinase inhibitor tofacitinib: A case report
Li B, Li GW, Xue L, Chen YY

Percutaneous fixation of neonatal humeral physeal fracture: A case report and review of the literature
Tan W, Wang FH, Yao JH, Wu WP, Li YB, Ji YL, Qian YP

Severe fundus lesions induced by ocular jellyfish stings: A case report
Zheng XY, Cheng DJ, Lian LH, Zhang RT, Yu XY

Application of ozonated water for treatment of gastro-thoracic fistula after comprehensive esophageal squamous cell carcinoma therapy: A case report
Wu DD, Hao KN, Chen XJ, Li XM, He XF

Germinomas of the basal ganglia and thalamus: Four case reports
Huang ZC, Dong Q, Song EP, Chen ZJ, Zhang JH, Hou B, Lu ZQ, Qin F

Gastrointestinal bleeding caused by jejunal angiosarcoma: A case report
Hui YY, Zhu LP, Yang B, Zhang ZY, Zhang YJ, Chen X, Wang BM

High expression of squamous cell carcinoma antigen in poorly differentiated adenocarcinoma of the stomach: A case report
Wang L, Huang L, Xi L, Zhang SC, Zhang JX

Therapy-related acute promyelocytic leukemia with FMS-like tyrosine kinase 3-internal tandem duplication mutation in solitary bone plasmacytoma: A case report
Hong LL, Sheng XF, Zhuang HF

Metastasis of esophageal squamous cell carcinoma to the thyroid gland with widespread nodal involvement: A case report
Zhang X, Gu X, Li JG, Hu XJ
Contents

Semimonthly Volume 8 Number 19 October 6, 2020

4595 Severe hyperlipemia-induced pseudoerythrocytosis - Implication for misdiagnosis and blood transfusion: A case report and literature review
Zhao XC, Ju B, Wei N, Ding J, Meng FJ, Zhao HG

4603 Novel brachytherapy drainage tube loaded with double 125I strands for hilar cholangiocarcinoma: A case report
Lei QY, Jiao DC, Han XW

4609 Resorption of upwardly displaced lumbar disk herniation after nonsurgical treatment: A case report
Wang Y, Liao SC, Dai GG, Jiang L

4615 Primary hepatic myelolipoma: A case report and review of the literature
Li KY, Wei AL, Li A

4624 Endoscopic palliative resection of a giant 26-cm esophageal tumor: A case report
Li Y, Guo LJ, Ma YC, Ye LS, Hu B

4633 Solitary hepatic lymphangioma mimicking liver malignancy: A case report and literature review
Long X, Zhang L, Cheng Q, Chen Q, Chen XP

4644 Intraspineous venous malformation of the maxilla after enucleation of a hemophilic pseudotumor: A case report
Cai X, Yu JJ, Tian H, Shan ZF, Liu XY, Jia J

4652 Intravesically instilled gemcitabine-induced lung injury in a patient with invasive urothelial carcinoma: A case report
Zhou XM, Wu C, Gu X

4660 Bochdalek hernia masquerading as severe acute pancreatitis during the third trimester of pregnancy: A case report
Zou YZ, Yang JP, Zhou XJ, Li K, Li XM, Song CH

4667 Localized primary gastric amyloidosis: Three case reports
Liu XM, Di LJ, Zhu JX, Wu XL, Li HP, Wu HC, Tuo BG

4676 Displacement of peritoneal end of a shunt tube to pleural cavity: A case report
Liu J, Guo M

4681 Parathyroid adenoma combined with a rib tumor as the primary disease: A case report
Han L, Zhu XF
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Peer-reviewer of *World Journal of Clinical Cases*, Prof. Adrián Ángel Inchauspe, obtained his MD in 1986 from La Plata National University (Argentina), where he remained as Professor of Surgery. Study abroad, at the Aachen and Tubingen Universities in Germany in 1991, led to his certification in laparoscopic surgery, and at the Louis Pasteur University in Strasbourg France, led to his being awarded the Argentine National Invention Award in 1998 for his graduate work in tele-surgery. He currently serves as teacher in the Argentine Acupuncture Society, as Invited Foreigner Professor at the China National Academy of Sciences and Hainan Medical University, and as editorial member and reviewer for many internationally renowned journals. (L-Editor: Filipodia)

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<table>
<thead>
<tr>
<th>NAME OF JOURNAL</th>
<th>World Journal of Clinical Cases</th>
</tr>
</thead>
<tbody>
<tr>
<td>ISSN</td>
<td>ISSN 2307-8960 (online)</td>
</tr>
<tr>
<td>LAUNCH DATE</td>
<td>April 16, 2013</td>
</tr>
<tr>
<td>FREQUENCY</td>
<td>Semimonthly</td>
</tr>
<tr>
<td>EDITORS-IN-CHIEF</td>
<td>Dennis A Bloomfield, Sandro Vento, Bao-Gan Peng</td>
</tr>
<tr>
<td>EDITORIAL BOARD MEMBERS</td>
<td><a href="https://www.wjgnet.com/2307-8960/editorialboard.htm">https://www.wjgnet.com/2307-8960/editorialboard.htm</a></td>
</tr>
<tr>
<td>PUBLICATION DATE</td>
<td>October 6, 2020</td>
</tr>
<tr>
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</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>INSTRUCTIONS TO AUTHORS</th>
<th><a href="https://www.wjgnet.com/bpg/gerinfo/204">https://www.wjgnet.com/bpg/gerinfo/204</a></th>
</tr>
</thead>
<tbody>
<tr>
<td>GUIDELINES FOR ETHICS DOCUMENTS</td>
<td><a href="https://www.wjgnet.com/bpg/GerInfo/287">https://www.wjgnet.com/bpg/GerInfo/287</a></td>
</tr>
<tr>
<td>GUIDELINES FOR NON-NATIVE SPEAKERS OF ENGLISH</td>
<td><a href="https://www.wjgnet.com/bpg/gerinfo/240">https://www.wjgnet.com/bpg/gerinfo/240</a></td>
</tr>
<tr>
<td>PUBLICATION MISCONDUCT</td>
<td><a href="https://www.wjgnet.com/bpg/gerinfo/208">https://www.wjgnet.com/bpg/gerinfo/208</a></td>
</tr>
<tr>
<td>ARTICLE PROCESSING CHARGE</td>
<td><a href="https://www.wjgnet.com/bpg/GerInfo/242">https://www.wjgnet.com/bpg/GerInfo/242</a></td>
</tr>
<tr>
<td>STEPS FOR SUBMITTING MANUSCRIPTS</td>
<td><a href="https://www.wjgnet.com/bpg/GerInfo/239">https://www.wjgnet.com/bpg/GerInfo/239</a></td>
</tr>
<tr>
<td>ONLINE SUBMISSION</td>
<td><a href="https://www.f6publishing.com">https://www.f6publishing.com</a></td>
</tr>
</tbody>
</table>

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Severe hyperlipemia-induced pseudoerythrocytosis - Implication for misdiagnosis and blood transfusion: A case report and literature review

Xi-Chen Zhao, Bo Ju, Na Wei, Jian Ding, Fan-Jun Meng, Hong-Guo Zhao

Abstract

BACKGROUND
Severe hyperlipemia (SHLE) has an impact on the results of many kinds of laboratory tests. Complete blood count (CBC) examination by automated blood cell counter (ABCC) is a quick and convenient measurement for screening abnormalities of blood cells that are triggered by various pathogenic insults in disease diagnosis and for monitoring changes in the treatment of existing hematological conditions. However, CBC results are frequently affected by many intrinsic and extrinsic factors from blood samples, such as in the setting of hypergammaglobulinemia and certain anticoagulants. SHLE could also affect CBC results.

CASE SUMMARY
A 33-year-old Chinese male presented with painful foot numbness and abdominal pain. He was initially misdiagnosed as having a myeloproliferative neoplasm (MPN) because of the marked abnormalities in CBC examination by the ABCC. Morphological evaluation of the bone marrow smears and biopsy showed no evidence of MPN. Gene mutations in Breakpoint cluster regions-Abelson murine leukemia viral oncogene homologue 1 (BCR-ABL1), Janus kinase 2 (JAK2), calreticulin (CALR), myeloproliferative leukemia virus (MPL), and colony-stimulating factor 3 receptor (CSF3R) were negative. Having noticed the thick chylomicron layer on blood samples and the dramatically fluctuating CBC results, we speculated that the fat droplets formed by shaking the blood samples in the setting of SHLE were
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mistakenly identified as blood cells due to the limited parameters of ABCC. Therefore, we removed a large part of the chylomicron layer and then reexamined the CBC, and the CBC results, as we expected, differed significantly from that of the sample before the chylomicron layer was removed. These significant differences had been validated by the subsequently repeated laboratory tests by measuring dual blood samples that the chylomicron layer was removed in one sample and was not in another, and comparing the CBC results. Computerized tomography reexamination of the upper abdomen revealed an exudative lesion surrounding his pancreas. After intensive consultation, definitive diagnosis was made as recurrent pancreatitis, hyperlipemia and pseudoerythrocytosis.

CONCLUSION

SHLE may become a potential cause of misdiagnosis of hyperlipemia-related diseases as MPNs and the resultant mistreatment. It may also lead to the misinterpretation of transfusion indications in patients with hematological disorders who critically need blood transfusion for supportive treatment.

Key Words: Case report; Hyperlipemia; Fat droplet; Pancreatitis; Pseudoerythrocytosis; Blood transfusion indication

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Core Tip: Severe hyperlipemia could affect the results of complete blood cell examination by automated blood cell counter. Here, we report a patient with severe hyperlipemia who was at first misdiagnosed as a myeloproliferative neoplasm because of the marked abnormalities in complete blood cell examination. Repeated laboratory tests, by measuring dual blood samples that the chylomicron layer was removed in one sample and was not in another, confirmed that the marked abnormalities was caused by the mistaken readings on automated blood cell counter. This phenomenon may lead to the misjudgment of many laboratory tests and the misinterpretation of blood transfusion indications.

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INTRODUCTION

Severe hyperlipemia (SHLE) has a significant impact on the results of many kinds of laboratory tests. Complete blood count (CBC) examination by automated blood cell counter (ABCC) is an important measurement for screening abnormalities of blood cells that are triggered by various pathogenic factors in disease diagnosis and for monitoring changes in the treatment of existing hematological conditions. However, CBC results are frequently affected by many intrinsic and extrinsic factors in blood samples, such as in the setting of hypergammaglobulinemia and certain anticoagulants. SHLE can also affect the CBC readings. Here, we report a recurrent pancreatitis patient who was initially misdiagnosed with myeloproliferative neoplasm (MPN) due to mistaken readings by the ABCC, but he was finally diagnosed with SHLE-induced pseudoerythrocytosis. It is speculated that the fat droplets in blood samples of individuals with SHLE are mistakenly identified as blood cells due to the limited parameters of ABCC. This phenomenon may become a potential cause of misdiagnosis of hyperlipemia-related diseases as MPNs and the resultant mistreatment, as reported in this paper. It may also lead to the misinterpretation of indications for blood transfusion in patients with hematological disorders who critically need blood transfusion as supportive treatment, such as in retinoic acid treatment for patients with acute promyelocytic leukemia and in L-asparaginase treatment for patients with acute lymphoblastic leukemia.
CASE PRESENTATION

Chief complaints
Painful foot numbness for 2 mo and abdominal pain for 2 d.

History of present illness
A 33-year-old Chinese male was referred to the emergency department for complaints of painful foot numbness for 2 mo and abdominal pain for 2 d, with the absence of fever, chills, headache, dizziness, cough, expectoration, vomiting, diarrhea, and abnormalities in the urine and feces. Upon physical examination, except for moderate tenderness in his left upper abdomen, no other abnormalities were found. His CBC showed the following results: white blood cell (WBC) count, 11.65 × 10^9/L; absolute neutrophil count (ANC), 8.85 × 10^9/L; red blood cell (RBC) count, 4.59 × 10^12/L; hemoglobin (Hb) value, 225 g/L; and platelet (Plt) count, 229 × 10^12/L. Serum amylose level was 33 U/L. Computerized tomography (CT) examination of his abdomen revealed the presence of adiposis hepatica, without other abnormalities. Ultrasonic inspection of his viscera revealed no obvious abnormalities in the liver, gallbladder, bile duct, pancreas, spleen, kidneys, ureter, or bladder. He was prescribed an antibiotic treatment with etimicin, and his abdominal pain was not relieved. The next day, reexamination of his CBC and serum amylase showed the similar results. He was thought to have erythrocytosis, and his abdominal pain was thought to result from a thrombotic event in the mesenteric vessels, which was probably due to the increased blood viscosity and enhanced Plt activities. So he was admitted to the hematology department.

History of past illness
He had a history of acute pancreatitis 2 years previously.

Physical examination
His height was at 176 cm; body weight 76.0 kg. His body temperature was at 37.2 °C; breathing rate 22 bp per min; heart rate 86 bp per min; blood pressure 126/78 mmHg. Upon physical examination, except for the bruise-looking appearance and moderate tenderness in his left upper abdomen, no other abnormalities were recorded. Conspicuous mucocutaneous plethora, hemorrhage, jaundice and exanthemata were not presented. No significant signs of nervous system, respiratory system, cardiovascular system, urogenital system and skeletal musculature system were found.

Laboratory examinations
On admission, the CBC showed as follows: WBC, 8.97 × 10^9/L; ANC, 6.66 × 10^9/L; hematocrit (Hct), 37.6%; RBC, 4.15 × 10^12/L; Hb, 204 g/L; mean corpuscular volume (MCV), 90.5 fL; mean corpuscular hemoglobin (MCH), 41.9 pg; mean corpuscular hemoglobin concentration (MCHC), 463 g/L; and Plt, 218 × 10^9/L. Urinalysis showed ketone bodies 3+, blood 1+, protein 3+, and glucose 3+. Examination of the coagulation profile and biochemical test could not be performed due to SHLE. The tests for hepatitis virus A, B, C, and E and a series of tumor markers were negative. Serum testosterone level was 175 ng/L. Morphological evaluation of the bone marrow (BM) smears and biopsy showed no evidence of MPN. Cytogenetic analysis of BM culture reported a normal karyotype of 46, XY[25]. Gene mutations in Breakpoint cluster regions-Abelson murine leukemia viral oncogene homologue 1 (BCR-ABL1), Janus kinase 2 (JAK2), calreticulin (CALR), myeloproliferative leukemia virus (MPL), colony-stimulating factor 3 receptor (CSF3R) were negative. These laboratory tests did not meet criteria for the diagnosis of MPN.

Further investigation of the abnormalities in CBC examination
Having noticed the thick chylomicron layer and dramatically fluctuating CBC results, we speculated that the fat droplets formed by shaking the blood samples in the setting of SHLE were mistakenly identified as blood cells due to the limited parameters of ABCC. Therefore, we removed a large part of the chylomicron layer and then reexamined the CBC and biochemical tests. The CBC showed: WBC, 8.34 × 10^9/L; ANC, 6.43 × 10^9/L; RBC, 4.15 × 10^12/L; Hb, 204 g/L; and Plt, 164 × 10^9/L. Biochemical tests showed: triglycerides (TGs), 3.96 mmol/L; total cholesterol (TC), 9.74 mmol/L; and low density lipoprotein (LDL), 5.67 mmol/L.
To determine whether the erythrocytosis was truly caused by SHLE, we drew dual blood samples, measured them (the chylomicron layer was removed in one sample
and was not in another) at the same time, and then compared the CBC results. As we expected, there was a significant difference in the CBC results. While the CBC readings in the sample without removing the layer showed WBC, 10.94 × 10^9/L; ANC, 8.86 × 10^9/L; RBC, 4.26 × 10^12/L; Hb, 208 g/L; and Plt, 287 × 10^9/L; the readings in the sample in which a large part of the layer was removed showed WBC, 4.34 × 10^9/L; ANC, 2.88 × 10^9/L; RBC, 3.69 × 10^12/L; Hb, 114 g/L; and Plt, 215 × 10^9/L. Subsequently, the repeated tests using this method yielded the similar results (listed in Table 1), confirming the contribution of SHLE to the formation of pseudoerythrocytosis.

**Imaging examinations**

On 5th day of hospitalization, CT reexamination revealed an exudative lesion surrounding the pancreas, in accordance with the diagnosis of pancreatitis.

**FINAL DIAGNOSIS**

After intensive consultation with specialists in gastroenterology, ultrasonography and radiology, a definitive diagnosis was made as recurrent pancreatitis, hyperlipemia and pseudoerythrocytosis.

**TREATMENT**

After the definitive diagnosis of recurrent pancreatitis, hyperlipemia and pseudoerythrocytosis was made, he was transferred to the gastroenterology department and was treated for his pancreatitis according to guidelines for diagnosis and treatment of chronic pancreatitis (Nanjing, 2018).

**OUTCOME AND FOLLOW-UP**

This patient was treated in the gastroenterology department for his pancreatitis, and the serum level of TGs, TC, and LDL gradually decreased. Along with the decreasing serum level of TGs, TC, and LDL, the results of CBC examination were normalized with the exception of a mild anemia.

**DISCUSSION**

MPNs, including chronic myelogenous leukemia, 8P11 myeloproliferative syndrome, chronic eosinophilic leukemia, polycythemia vera (PV), essential thrombocythemia, primary myelofibrosis, chronic neutrophilic leukemia, systemic mastocytosis, and chronic basophilic leukemia, are clonal diseases resulting from the uncontrolled proliferation of hematopoietic stem and progenitor cells without obvious differential arrest and dysplasia, leading to the hypercellular BM and the increased periphery blood (PB) cell counts. Diagnosis and classification of MPNs are primarily based on the morphological examination of the BM and PB cells. These seemingly normal-appearance blood cells are caused by gain of function reciprocal translocations and mutations in genes encoding receptor tyrosine kinases or their c-Jun N-terminal kinase-signal transducer and activator of transcription (JNK-STAT)-associated signal pathway components. Constitutive activation of the JNK-STAT signaling pathway is responsible for excessive and autonomous blood cell production\(^1\)-\(^3\). MPN-associated fusion genes caused by the recurrent rearrangements frequently involved the genes ABL1, platelet-derived growth factor receptor-α (PDGFR-α), PDGFR-β, and fibroblast growth factor receptor-1, whereas MPN-associated gene mutations frequently involved the genes JAK2, CALR, MPL, CSF3R, and the stem cell growth factor receptor gene CD117\(^1\)-\(^7\).

In this paper, we described a pancreatitis patient who was at first misdiagnosed as erythrocytosis (presumptive diagnosis of PV) because of the markedly elevated Hb level at presentation due to the mistaken readings of the ABCC in the evaluation of complete blood cells, but he was eventually diagnosed with SHLE-induced pseudoerythrocytosis. The purpose of reporting this patient and the diagnostic process is for the doctors to call attention to the comprehensive and objective evaluation of the
Table 1 Complete blood count results before and after removing the chylomicron layer

<table>
<thead>
<tr>
<th>Times No.</th>
<th>WBCs</th>
<th>ANC</th>
<th>RBCs</th>
<th>Hb</th>
<th>Hct</th>
<th>Plts</th>
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<tr>
<td>1</td>
<td>BRL</td>
<td>10.94</td>
<td>8.86</td>
<td>4.26</td>
<td>208</td>
<td>36.20</td>
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<tr>
<td>ARL</td>
<td>4.83</td>
<td>2.88</td>
<td>3.69</td>
<td>114</td>
<td>33.70</td>
<td>215</td>
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<tr>
<td>2</td>
<td>BRL</td>
<td>9.85</td>
<td>7.08</td>
<td>4.63</td>
<td>221</td>
<td>40.30</td>
</tr>
<tr>
<td>ARL</td>
<td>4.49</td>
<td>2.10</td>
<td>4.37</td>
<td>132</td>
<td>39.50</td>
<td>177</td>
</tr>
<tr>
<td>3</td>
<td>BRL</td>
<td>10.72</td>
<td>8.76</td>
<td>4.23</td>
<td>188</td>
<td>36.30</td>
</tr>
<tr>
<td>ARL</td>
<td>4.98</td>
<td>2.93</td>
<td>3.76</td>
<td>115</td>
<td>34.10</td>
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WBC: White blood cell, × 10⁹/L; ANC: Absolute neutrophil count, × 10⁹/L; RBC: Red blood cell, × 10¹²/L; Hb: Hemoglobin, g/L; Hct: Hematocrit, %; Plt: Platelet, × 10⁹/L; BRL: Before removing the chylomicron layer; ARL: After removing the chylomicron layer.

CBC results on ABCC, especially in the setting of SHLE, a very common laboratory finding in clinical practice.

The World Health Organization diagnostic criteria for PV is based on the elevated Hb (> 16.5 g/dL in men or > 16.0 g/dL in women) or Hct (> 49% in men or > 48% in women) levels, with the prominent erythroid, granulocytic and megakaryocytic proliferation in BM biopsy, incorporating the presence of JAK2V617F or JAK2 exon 12 mutation[2, 3]. In clinical practice, the elevated Hb level is the most commonly used parameter for the diagnosis and grading of anemia and erythrocytosis. In this patient, the markedly elevated Hb level at presentation (up to 225 g/L = 22.5 g/dL) strongly indicated the possibility of the presence of PV. However, the following laboratory investigations, including morphological examination, cytogenetic and molecular analysis, did not meet criteria for the diagnosis of any kind of MPN.

The CBC profile in this patient revealed a noteworthy feature: WBCs, ANCs, Hb, MCH, and MCHC were all increased to some degree, with the presence of markedly elevated Hb and MCHC level, but the absence of a paralleled increase in RBCs and Hct was the most prominent feature. The dramatically fluctuating and obviously discrepant CBC results among Hct, RBCs, Hb, MCV, MCH, and MCHC were easily identified. In addition, there was a conspicuous chylomicron layer floating on the surface of the blood samples. Having noticed these, we removed a large part of the chylomicron layer and then evaluated the CBC readings. After removing a large part of the chylomicron layer, WBCs, ANC, Hb, MCH, MCHC, and Plt were all decreased to varying degrees. We presumed that the erythrocytosis was the result of fatty droplets that are mistakenly identified as blood cells due to the limited parameters of ABCC. So a laboratory investigation was performed by drawing dual blood samples, measuring them (the chylomicron layer was removed in one sample and was not in another) at the same time, and comparing the CBC results. As we expected, there was a significant difference in the CBC results. This laboratory investigation provided strong evidence to conform the contribution of SHLE to the markedly elevated Hb and MCHC levels in CBC examination in this patient.

Hyperlipemia, one of the essential compartments of metabolic syndrome, combination of genetic background and environmental factors in its pathogenesis, is a common laboratory finding in biochemical tests. SHLE could be the complication of various diseases mainly involving diseases in the cardiovascular system, endocrine system, liver, pancreas and kidneys, in which hyperlipemia may play an essential role in the pathogenesis of targeted tissue damages[8-21]. In the genetically predisposed individuals, systemic inflammatory conditions and dietary regimen may be the major environmental factors to influence the lipid metabolism and insulin resistance[22-27]. SHLE sometimes occurs in the natural history or in the treatment of certain hematological diseases, such as in tyrosine kinase inhibitor treatment for patients with MPNs, retinoic acid treatment for patients with acute promyelocytic leukemia and L-asparaginase treatment for patients with acute lymphoblastic leukemia[28-32]. It is well known that SHLE have an unexpected impact on many kinds of laboratory tests, making the diagnostic process more complicated and perplexed. SHLE may also affect the CBC results on the ABCC.

To date, there are too few reports that have documented the erythrocytosis to be complicated by hyperlipemia in that erythrocytosis was in association with the high frequency of cardiovascular events and the incitation of pancreatitis[33-38]. However, it has never been recognized that SHLE can lead to the induction of
pseudoerythrocytosis. The higher frequency of erythrocytosis in the context of SHLE strongly indicates that some patients may be misdiagnosed as MPNs due to mistaken readings by the ABCCs as reported in the present case. The high frequency of cardiovascular events and the induction of pancreatitis in this setting may lie in the hyperlipemia-related diseases themselves which may result from the dysregulated metabolic activities and the co-existing inflammatory conditions.

The impact of SHLE on the CBC results may be overlooked because hyperlipemia patients seldom see the hematologists, and thus, this phenomenon has not been rigorously investigated. Differential diagnosis between true erythrocytosis and pseudoerythrocytosis in the setting of hyperlipemia may be simply by intellectual and comprehensive analysis of the reasonableness of each CBC results (especially by the observation whether the Hb level is in parallel to the Hct value or not) at first and by comparing the CBC results between blood samples with and without removal of the chylomicron layer subsequently by a means employed in this paper. In addition, the elevated even normal serum levels of total, LDL and high density lipoprotein cholesterol are biased to the diagnosis of hyperlipemia-induced pseudoerythrocytosis or have an co-existence of MPNs and hyperlipemia-related diseases. However, this investigation is limited by the changes in the value of blood sample, and these changes may make it difficult to interpret the true Hb concentration in patient’s blood. Another question is what concentration and components of blood lipids are able to significantly affect the CBC readings on the ABCC. So, this investigation merely provides an information that SHLE could affect the CBC results measured by ABCC, and how to correctly measure the true Hb concentration in patient’s blood warrants further investigations.

Recognizing this phenomenon helps better understand the CBC results on ABCC in the context of hyperlipemia, especially in individuals with SHLE. MPNs are clonal diseases characterized by the constitutive activation of genes in growth factor receptors or their signal pathway components, resulting in the uncontrolled proliferation of hematopoietic cells. Treatment of MPNs differ completely from that of hyperlipemia-related diseases. Therefore, the differential diagnosis between these diseases is very important as demonstrated in this case. In addition, SHLE sometimes occurs in the treatment of hematological malignancies, and this may lead to the misinterpretation of transfusion indications, which may result in serious consequences such as severe ischemic episodes and severe bleeding events.

**CONCLUSION**

SHLE could significantly affect the CBC results on ABCC. This phenomenon may lead to the misdiagnosis of hyperlipemia-related diseases as MPNs and the resultant mistreatment. It may also lead to the misinterpretation of transfusion indications in the treatment of hematopoietic diseases.

When a patient presents with a high level of Hb and makes a presumptive diagnosis of MPNs, it is very necessary to carefully examine the blood samples, to correctly evaluate the CBC results and to look up the biochemical tests. The appearance of a chylomicron layer in blood samples, the discrepant results in CBC examination and the elevated serum levels of TGs, TC, and LDL strongly indicate the possible diagnosis of pseudoerythrocytosis. In this setting, investigations are warranted as showed in our paper, and the underlying diseases must be diagnosed as soon as possible so as to let the patient receive prompt and proper treatments. In patients with hematological diseases in the setting of hyperlipemia, the blood transfusion indications must be adjusted according to the patient’s symptoms rather than are dependent merely on the Hb levels or the Plt counts.

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