Platelet Phagocytosis by Neutrophils, Platelets Lack of Granules and Giant Platelets Result in Pseudothrombocytopenia

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Case Report

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**Abstract**

Pseudothrombocytopenia (PTCP) is a condition in which the decreased platelet count does not agree with the clinical status of the patient, can lead to misdiagnose, unnecessary tests, unnecessary treatment. The present case describes a 73-year-old man suffered with pulmonary tuberculosis, treated with anti-tuberculosis therapy (isoniazid, rifampicin, pyrazinamide, ethambutol, 2HRZE/4HR). One month later, the patient had a significant decrease in platelets (101 to 56 x10^9/L). Peripheral blood smear showed that 28% platelets were phagocytosis by neutrophils, 26% platelets were lack of granules and 6% platelets’ volume increased significantly. When the anticoagulant was changed from EDTA to sodium citrate, there was no change in the above phenomenon. By manual count, the value of platelets was 113 x 10^9/L. After the completion of anti-tuberculosis therapy, platelet morphology gradually returned to normal. HRZE treatment may cause platelet morphology abnormal, resulting in PTCP. In such cases, we should regularly review the peripheral blood smear to ensure the accuracy of the results and avoid unnecessary examination and treatment. The emergence of PTCP may does not mean the presence of specific disorders.

**Background**

Thrombocytopenia can have several causes, including platelet clumping or formation of platelet rosettes around neutrophils, the use of certain drugs. When the decreased platelet count does not agree with the clinical status of the patient, incorrect measurements of platelet may lead PTCP, then cause the mistake of diagnostic and treatment. The knowledge of PTCP is important for the accuracy of a clinical assessment and for avoiding unnecessary treatment [1]. PTCP is usually caused by improper blood collection, thylenediaminetetraacetic acid dependent pseudothrombocytopenia (EDTA-PTCP), platelet volume increases or decreases [2, 3]. The mechanism is not clearly defined so far. Aggregation is the most common phenomenon while platelet phagocytosis is the rarest one in PTCP [4, 5].

This little known in vitro phenomenon still under recognized may lead to misdiagnosis of thrombocytopenia, overtreatment, and further, even invasive, unnecessary testing. The present case describes a case platelet phagocytosis by neutrophils, platelet lack of granules and giant platelet result in PTCP. We regularly examined the peripheral blood cell morphology of our patient, there was no platelet satellite and EDTA-dependent platelet phenomena. Anti-tuberculosis treatment may cause platelet morphology abnormal, resulting in PTCP. In such cases, we should regularly review the peripheral blood smear to ensure the accuracy of the results and avoid unnecessary examination and treatment.

**Case Presentation**

A 73-year-old man presented to our hospital with chest discomfort, cough, no fever. The patient has a 30-year history of chronic hepatitis B, his liver function was normally. He has no history of hypertension, diabetes or heart disease, drug allergy, respiratory diseases, Corona Virus Disease 2019 (COVID-19). The patient had a long history of smoking and drinking.
The patient's platelet was $101 \times 10^9$/L on admission, peripheral blood smear showed no obvious change in morphology. Routine laboratory tests showed that there was no obvious abnormality in white blood cell (WBC), liver and kidney function, or coagulation function. Chest computed tomography (CT) showed bilateral lung infection, emphysema, pulmonary tuberculosis and cavitation were considered. On physical examination, breathing sounds in both lungs were rough, and a few moist crackles could be heard in the left upper lung. Mycobacterium tuberculosis rifampicin resistance gene test and serum tuberculosis antibody test were positive. Combined with the patient's symptoms, signs, CT scan and laboratory tests, he was diagnosed with tuberculosis. Subsequently, he accepted anti-tuberculosis therapy (isoniazid, rifampicin, pyrazinamide, ethambutol, 2HRZE/4HR) on the daily dose recommended by the World Health Organization tuberculosis guidelines.

One month later, laboratory tests showed that there was no obvious abnormality in WBC, liver and kidney function, or coagulation function, but there was a significant decrease in platelets ($56 \times 10^9$/L reference range: $125–350\times10^9$/L) (Table 1). Peripheral blood smear showed that 28% platelets were phagocytosis by neutrophils, 26% platelets were lack of granules and 6% platelets' volume increased significantly, no platelet satellite was observed, other cells presented no morphological abnormalities (Table 1 and Fig. 1). When the anticoagulant was changed from EDTA to sodium citrate, there was no change in the above phenomenon. By manual count, the value of platelets was $136 \times 10^9$/L. The patient did not have any bleeding tendency. Therefore, we believe that the patient developed PTCP and did not need to undergo bone marrow puncture and platelet enhancement therapy. In subsequent testing, platelet phagocytosis, platelet lack of granules and giant platelet, all these situation existed all the time (Table 1). Three months after the completion of antituberculous therapy, platelet morphology gradually returned to normal (Table 1).

Table 1

| Laboratory tests results of the patient | platelet (PLT), reference range: $125–350\times10^9$/L. |
| PLT | Jan-27 | Mar-2 | Mar-10 | May-19 | Jun-14 | Sep-30 |
|-------------------------------|---------|--------|--------|--------|--------|--------|
| Instrument count ($\times10^9$/L) | 89 | 56 | 58 | 74 | 46 | 132 |
| Manual count ($\times10^9$/L) | 101 | 136 | 121 | 109 | 93 | 136 |
| Platelet phagocytosis by neutrophils (%) | 0 | 28 | 22 | 20 | 20 | 0 |
| Platelet lack of granules (%) | 0 | 26 | 20 | 20 | 18 | 0 |
| Giant platelet (%) | 0 | 6 | 4 | 6 | 11 | 1 |
| Total rate of abnormal platelets (%) | 0 | 60 | 46 | 46 | 49 | 1 |

*: none. Detecting instrument: Japan, Sysmex, XN9000.
Discussion

Platelets play an important role in the body, such as regulators of hemostasis and thrombosis. In pathological conditions, platelets are essential for formation of occlusive thrombus formation. Platelets have also been shown to play an important role in innate immunity as well as regulation of tumor growth and extravasations in the vessel[6]. Once the number or function of platelets is abnormal, further testing or treatment is required. PTCP was identified as being in vitro phenomenon which can be caused by following situation: EDTA-dependent platelet phenomena[7], platelet agglutination due to improper specimen collection, cold agglutination[8], platelet satellite phenomenon[9], large/ giant platelets[10], drug therapy[11]. The prevalence of PTCP in blood and platelet apheresis donors, with frequency ranging from 0.01–0.2%[12, 13]. PTCP was significantly higher in males aged 50 years or older[14]. PTCP caused by platelet satellitism and phagocytosis by neutrophils only occurs in approximately 0.1% in clinical[15, 16]. Here, we present an extremely rare case of PTCP results from platelet phagocytosis by neutrophils, platelet lack of granules and giant platelet. EDTA-dependent autoantibodies triggers several signaling pathways that activate platelets, leading to their aggregation, satellitism or phagocytosis[15, 16]. Platelet aggregation is the most common situation while platelet phagocytosis is the rarest one. Some studies suggest that platelet phagocytosis is related with EDTA-dependent autoantibodies, it usually occurs after platelet satellite phenomenon and is an extension of platelet satellite[4, 17]. In our case, we find platelet phagocytosis by neutrophils, platelet lack of granules and giant platelet in the patient's peripheral blood smear(Fig. 1). When the anticoagulant was changed from EDTA to sodium citrate, these phenomena still exist. At present, there are no reported cases of the simultaneous occurrence of these three abnormal forms. The optical platelet-counting method identifies platelets with laser light scatter technique shown as a reliable method for accurate platelet counting in thrombocytopenic patients, this technique combines scattered light and side fluorescence detectors, a fluorescent dye (oxazine) is used beforehand to stain platelets and reticulocytes[18–20]. However, this technique also fails to detect engulfed platelets and platelets without granules In multiple tests, the number of platelets counted by manual was significantly higher than that counted by instrument (Table 1). This also indicate that instrumentation is not a complete substitute for manual testing, and peripheral blood smears must be performed in cases of PTCP. The PTCP may have only minor pathophysiologic significance. However, this situation must be distinguished from true in vivo platelet clumps detected by chance during a blood test. As the in vitro phenomenon of PTCP could be misdiagnosed with thrombocytopenia, it does affect diagnostic, management, and therapeutic decisions.

Although PTCP has been previously reported during therapy with abciximab, the incidence and significance of this occurrence are unknown, PTCP is a benign laboratory condition that does not increase bleeding, stroke, transfusion requirements or the need for repeat revascularization[21]. As one of the most effective chemotherapy medicines for tuberculosis (TB), rifampicin is widely used in China as there is high incidence of this disease. The common adverse effects of rifampicin are gastrointestinal disorders, skin rash, hepatotoxicity, etc, rifampicin is the most likely drug to cause thrombocytopenia in the course of antituberculous therapy[22, 23], it can even cause disseminated intravascular coagulation (DIC) in patients[24]. But, PTCP caused by antituberculous therapy has not been reported so far. In our
case, before receiving anti-tuberculosis treatment, the patient showed no significant abnormality in the number \((101 \times 10^9/L)\) and morphology of platelets. One month after antituberculous treatment \((2HRZE/4HR)\), platelets count was significantly reduced by instrument detection \((56 \times 10^9/L)\). However, through manual counting of peripheral blood smear, we found no significant change in the number of platelets in the patient \((136 \times 10^9/L)\). During the course of treatment, this phenomenon persists (Table1), the patient did not show a tendency of bleeding or thrombosis, so we deduced that the patient developed PTCP. Three months after the end of treatment, no abnormal platelets were observed in the peripheral blood, so we speculated that the patient’s PTCP was caused by anti-tuberculosis treatment. Of course, since we are reporting a single case, we have less evidence and didn't do any further research, our opinion is not conclusive considering that the mechanisms of drug-induced PTCP are extremely complex.

Here, we report it for the first time that an extremely rare case of PTCP results from platelet phagocytosis by neutrophils, platelet lack of granules and giant platelet. HRZE treatment may cause platelet morphology abnormal, resulting in PTCP. In such cases, we should regularly review the peripheral blood smear to ensure the accuracy of the results and avoid unnecessary examination and treatment. The emergence of PTCP may does not mean the presence of specific disorders.

**Declarations**

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**Competing interests**

The authors declare that they have no competing interests.

**Data availability**

All data are included in this article

**Code availability**

Not applicable

**Ethics approval**

Not applicable

**Author contributions**

YZ and JZ contribute to thesis selection and design, data collection; SL participate in data analysis and interpretation; FG contributes to critical review of the intellectual content of an article; YD and MZ contribute to the manuscript writing.
Consent to participate

The study is supported by the patient’s wife and she has signed informed consent.

Consent for publication

Not applicable

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**Figures**
Figure 1

Wright-Giemsa–stained peripheral blood smear of the patient. Platelet phagocytosis by neutrophils, platelets lack of microparticle and giant platelets can be observed in the above pictures.