Original Article

Study of Ethylenediaminetetraacetic acid (EDTA) - Dependent Pseudothrombocytopenia

Oza N*, Ingole N and Gangane N

Department of Pathology, Mahatma Gandhi Institute of Medical Sciences, Sevagram- 442 102, Wardha, Maharashtra, India.

*Corresponding Author
Dr. Oza N
Post-graduate student,
Department of Pathology,
Mahatma Gandhi Institute of Medical Sciences,
Sevagram- 442 102, Wardha, Maharashtra, India
E-mail: drnikitaaoza@gmail.com

Keywords:
Ethylenediaminetetraacetic acid (EDTA);
CPT (trisodium citrate, pyridoxal 5'-phosphate and Tris);
Pseudothrombocytopenia (PTCP);
Platelet count

Abstract

Objectives: To evaluate and compare the cases of EDTA-PTCP with actual platelet counts at different time intervals with new anticoagulant.

Methods: This cross-sectional study was carried out in rural tertiary centre in central India. Blood samples were collected in K3-EDTA and CPT vials separately and subjected for peripheral smear examination and manual platelet counts. Comparison of platelet counts obtained by automated cell counter at 30 minutes, 3-4 hours and at 24 hours of blood collection from both anticoagulants and with manual counts was done.

Results: The platelet counts in EDTA anticoagulated blood on suspicion of thrombocytopenia, as well as even after collection of fresh blood sample from same patient in EDTA and the counts at different time intervals showed significantly lower counts than that observed in CPT anticoagulated blood at the parallel time intervals and that in the manual platelet counts.

Conclusions: Unrecognized PTCP may result in unnecessary laboratory testing, bone marrow aspiration and unwarranted transfusions and will prevent needless evaluations of thrombocytopenia and related therapeutic decisions.

1. Introduction

Spurious thrombocytopenia, also called Pseudothrombocytopenia (PTCP), results from low platelet counts due to in vitro platelet clumping [1-7]. Platelet clumping in PTCP results in inaccurate platelet concentration, which leads to misdiagnosis of thrombocytopenia when analyzed with hematology analyzer [7-8].

Frequency of 0.3% and 1.2% respectively was reported by Mant et al.[9] and Manthorpe et al.[10], which referred however to a small case series. This type of alteration is now familiar to the clinical pathologist; and consequently it has been evaluated more accurately, and at present its frequency is considered to be within range of 0.09 – 0.11% [11]. Although PTCP is an infrequent condition, it accounts for a sizable fraction of all the cases of “thrombocytopenia” that are referred to for further evaluation [12].

Pseudothrombocytopenia (PTCP) is an immunologically mediated phenomenon caused by the presence of EDTA – dependent cold anti-platelet auto-antibodies in blood that cause in vitro platelet clumping as shown in figure 1 – A, B [5,7,13-15].

Fig.1 (A, B): Platelet clumps in EDTA-anticoagulated blood.
2. Methods

This cross sectional study was carried out in the Hematology division of the Department of Pathology, over a period of 2 years from May 2011 to May 2013, in a medical institute in central India.

Subjects:

Inclusion criteria:

All the cases in which the automated counter report showed thrombocytopenia with platelet counts less than 130 x 10^9/litre with peripheral blood film examination showing platelets in fair number, either diffusely distributed or in clumps or aggregates and appeared to be within normal limits; were considered as Pseudothrombocytopenia and included in the study.

Exclusion criteria:

1. The platelet counts between 130-150 x 10^9 /litre were excluded.
2. The cases with known cause for thrombocytopenia as obtained from history, clinical examination and medical records, were excluded.

Study methodology:

Blood samples were collected in K3-EDTA and CPT (trisodium citrate, pyridoxal 5'-phosphate and Tris) vials separately and examination of well prepared, air-dried, labeled peripheral smear stained with Leishman was done. Examination was done using light microscope under oil immersion (100x) with (10x objectives) for evaluation of platelet morphology, clumps, and counts. For EDTA-PTCP cases, the manual platelet count is considered 'gold standard' for this comparison as reported by Hyun-Sook Chi[15] and Lippi et al.[17]. This method was performed using improved Neubauer's chamber. Convenient procedure is to count five groups of 16 small squares in the central area (0.02µl).

Platelet count per litre = Number of cells counted x Dilution x 10^-6

Volume counted (µl)

To ensure a coefficient of variation of 8-10 %, the total number of platelet count should always exceed 200.

Using automated blood analyser, platelet counts were obtained at 30 minutes, 3-4 hours and at 24 hours of blood collection.

Thus, the platelet counts obtained by manual method; by automated counter at 30 minutes, 3-4 hours and at 24 hours of blood collection using two different anticoagulants were compared and these were also compared with the initial platelet counts on which pseudothrombocytopenia was suspected.

The data was recorded and findings were analyzed statistically using z-test and test statistics. The software used in the analysis is SPSS 17.0 version and graph pad prism 5.0. The p-value of less than 0.05 is considered as statistically significant.

3. Results

In the present study, we assessed the cases of suspected EDTA dependent Pseudothrombocytopenia (showing thrombocytopenia on initial automated platelet counts from EDTA anticoagulated blood with adequate platelet count and presence of platelet clumps in the peripheral blood smear) for accurate platelet count with manual method and also using EDTA and CPT as anticoagulants with automated platelet counts at different time intervals.

Study included 43 males and 60 females with M: F ratio of 1:1.3. The patient's age varied from 3-85 years with the mean age of 36.78 ± 20.33 years. No significant association was found with respect age or distribution of cases. EDTA-PTCP cases were found to be associated both in health and disease state.

The present study compared the mean platelet counts in EDTA anticoagulated blood on initial suspicion of pseudothrombocytopenia and at different time intervals after collection of fresh blood sample from same patients in EDTA anticoagulant with parallel platelet counts in CPT anticoagulated blood and also with manual platelet counts.

The study observed the mean initial platelet counts in EDTA (103.67 ± 25.34 x 10^9/l) to be much lower than the mean manual platelet count (222.63 ± 85.22 x 10^9/l) and the difference was statistically significant. The mean platelet count in EDTA anticoagulated blood at 0-30 minutes and at 3-4 hours (171.40 ± 78.10 x 10^9/l) and (171.63 ± 81.16), though it was in the normal range, was also significantly lower than mean manual platelet count and the parallel automated platelet count in CPT anticoagulated blood (226.63 ± 93.25 x 10^9/l) and (230.25 ± 97.57) at 0-30 minutes and 3-4 hours respectively (Table 1).

Table 1: Showing comparison of initial platelet count suspicious of pseudothrombocytopenia with manual platelet count and platelet count in EDTA and CPT anticoagulated blood at 0-30 minutes and at 3-4 hours.

<table>
<thead>
<tr>
<th>Number of cases</th>
<th>Mean initial platelet count (x 10^9/l)</th>
<th>Mean manual platelet count (x 10^9/l)</th>
<th>Mean automated platelet count (x 10^9/l) at 0-30 minutes</th>
<th>Mean automated platelet count (x 10^9/l) at 3-4 hours</th>
</tr>
</thead>
<tbody>
<tr>
<td>103</td>
<td>103.67 ± 25.34</td>
<td>222.63 ± 85.22</td>
<td>171.40 ± 78.10</td>
<td>230.25 ± 97.57</td>
</tr>
<tr>
<td>Initial Platelet Count</td>
<td>p-value</td>
<td>0.000, S, p&lt;0.05</td>
<td>0.000, S, p&lt;0.05</td>
<td>0.000, S, p&lt;0.05</td>
</tr>
<tr>
<td>Manual Platelet Count</td>
<td>p-value</td>
<td>0.000, S, p&lt;0.05</td>
<td>0.74, NS, p&gt;0.05</td>
<td>0.000, S, p&lt;0.05</td>
</tr>
<tr>
<td>Initial Vs Manual Platelet Count</td>
<td>p-value</td>
<td>0.000, S, p&lt;0.05</td>
<td>0.000 S, p&lt;0.05</td>
<td>0.000 S, p&lt;0.05</td>
</tr>
<tr>
<td>S: Significant; NS: Not significant</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

The present study also assessed the changes in the platelet counts in anticoagulated blood samples after preserving the blood samples at 3-4°C in a refrigerator for 24 hours, in total 74 cases. The mean platelet counts in EDTA and CPT anticoagulated blood were found to be 183.70 ± 100.21 x 10^9/l and 266.04 ± 103.51 x 10^9/l respectively. In the same cases, the mean initial platelet count was 100.51 ± 26.57 x 10^9/l and the manual platelet count was 240.68 ± 89.24 x 10^9/l, as shown in table 2.

Table 2: Showing comparison of initial platelet count suspicious of pseudothrombocytopenia with manual platelet count and platelet count in EDTA and CPT anticoagulated blood at 24 hours.

<table>
<thead>
<tr>
<th>Number of cases</th>
<th>Mean initial platelet count (x 10^9/l)</th>
<th>Mean manual platelet count (x 10^9/l)</th>
<th>Mean automated platelet count (x 10^9/l)</th>
</tr>
</thead>
<tbody>
<tr>
<td>74</td>
<td>100.51 ± 26.57</td>
<td>240.68 ± 89.24</td>
<td>183.70 ± 100.21</td>
</tr>
<tr>
<td>Initial Platelet Count</td>
<td>z-value</td>
<td>6.90</td>
<td>13.32</td>
</tr>
<tr>
<td>Manual Platelet Count</td>
<td>z-value</td>
<td>3.65</td>
<td>1.59</td>
</tr>
<tr>
<td>Initial Vs Manual Platelet Count</td>
<td>z-value</td>
<td>12.95</td>
<td></td>
</tr>
<tr>
<td>S: Significant; NS: Not significant</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

By using z-test, statistically significant difference was found between initial platelet count and platelet count in EDTA anticoagulated
blood at 24 hours ($z = 6.90; p = 0.000$), and between initial platelet count and platelet count in CPT anticoagulated blood at 24 hours ($z = 13.32; p=0.000$).

Similarly, statistically significant difference was found between manual platelet count and platelet count in EDTA anticoagulated blood at 24 hours ($z = 3.65; p = 0.000$); but no significant difference was found between manual platelet count and platelet count in CPT anticoagulated blood at 24 hours ($z = 1.59; p=0.11$) also.

Thus, the platelet counts in EDTA anticoagulated blood on suspicion of thrombocytopenia, as well as even after collection of fresh blood sample from same patient in EDTA and the counts at 0-30 minutes, 3-4 hours and after 24 hours showed significantly lower counts than that observed in CPT anticoagulated blood at the parallel time intervals and than that in the manual platelet counts. Thus, we found the difference of initial mean platelet count in EDTA with manual platelet count of 55% and the difference of mean platelet count in EDTA and CPT anticoagulated blood of about 25% with counts at 0-30 minutes and at 3-4 hours, and of about 31% at 24 hours. Thus, low platelet counts were probably because of the effect of EDTA anticoagulant on platelets.

The mean automated platelet counts in CPT anticoagulated blood at 0-30 minutes and at 3-4 hours as well as at 24 hours after blood collection were found to be comparable with the mean manual platelet counts. Thus, the CPT anticoagulant was found to be a better anticoagulant for getting correct platelet counts in cases of EDTA-dependent pseudothrombocytopenia.

The present study also compared the initial platelet count in EDTA (103.67 ± 25.34 x 10^9/l) with that at 0-30 minutes (171.40 ± 78.10 x 10^9/l) and at 3-4 hours (171.63 ± 81.16 x 10^9/l) in the same anticoagulant. The statistical analysis is shown in table 3:

**Table 3: Showing comparison of initial platelet count in EDTA anticoagulated blood with that at 0-30 minutes and 3-4 hours in same anticoagulant.**

**Descriptive Statistics:**

<table>
<thead>
<tr>
<th></th>
<th>EDTA</th>
<th>No. of cases</th>
<th>Mean platelet count x 10^9/l</th>
<th>Standard Deviation</th>
<th>Standard Error Mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>Initial</td>
<td>103</td>
<td>103.67</td>
<td>25.34</td>
<td>2.49</td>
<td></td>
</tr>
<tr>
<td>0-30 min</td>
<td>103</td>
<td>171.40</td>
<td>78.10</td>
<td>7.69</td>
<td></td>
</tr>
<tr>
<td>3-4 hrs</td>
<td>103</td>
<td>171.63</td>
<td>81.16</td>
<td>7.99</td>
<td></td>
</tr>
</tbody>
</table>

**Wilcoxon Signed Rank Test:**

<table>
<thead>
<tr>
<th>Paired Differences</th>
<th>Mean</th>
<th>Std. Deviation</th>
<th>Std. Error Mean</th>
<th>95% Confidence Interval of the Difference</th>
<th>z</th>
<th>df</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Initial &amp; 0-30 min</td>
<td>67.72</td>
<td>80.45</td>
<td>7.92</td>
<td>52.00; 83.45</td>
<td>8.54</td>
<td>102</td>
<td>0.000, S, p&lt;0.05</td>
</tr>
<tr>
<td>Initial &amp; 3-4 hrs</td>
<td>67.95</td>
<td>82.37</td>
<td>8.11</td>
<td>51.85; 84.05</td>
<td>8.37</td>
<td>102</td>
<td>0.000, S, p&lt;0.05</td>
</tr>
<tr>
<td>0-30 min &amp; 3-4 hrs</td>
<td>0.22</td>
<td>28.45</td>
<td>2.80</td>
<td>5.33; 5.78</td>
<td>0.08</td>
<td>102</td>
<td>0.937, NS, p&gt;0.05</td>
</tr>
</tbody>
</table>

By using Wilcoxon signed rank test, significant difference was found between initial platelet count and that at 0-30 minutes ($z = 8.54; p = 0.000$); and between initial platelet count and that at 3-4 hours ($z = 8.37; p = 0.000$); but, no statistical change was found in platelet count from 0-30 minutes to 3-4 hours ($z = 0.08; p = 0.937$).

Similarly, the initial platelet count in EDTA was compared with the platelet count in CPT anticoagulated blood at 0-30 minutes and at 3-4 hours. The statistical analysis is shown in table 4:

**Table 4: Showing comparison of initial platelet count in EDTA with platelet counts in CPT anticoagulated blood at 0-30 minutes and at 3-4 hours.**

**Descriptive Statistics:**

<table>
<thead>
<tr>
<th></th>
<th>CPT</th>
<th>No. of cases</th>
<th>Mean platelet count x 10^9/l</th>
<th>Std. Deviation</th>
<th>Std. Error Mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>Initial</td>
<td>103</td>
<td>103.67</td>
<td>25.34</td>
<td>2.49</td>
<td></td>
</tr>
<tr>
<td>0-30 min</td>
<td>103</td>
<td>226.63</td>
<td>93.25</td>
<td>9.18</td>
<td></td>
</tr>
<tr>
<td>3-4 hrs</td>
<td>103</td>
<td>230.25</td>
<td>97.57</td>
<td>9.61</td>
<td></td>
</tr>
</tbody>
</table>

**Wilcoxon Signed Rank Test:**

<table>
<thead>
<tr>
<th>Paired Differences</th>
<th>Mean</th>
<th>Std. Deviation</th>
<th>Std. Error Mean</th>
<th>95% Confidence Interval of the Difference</th>
<th>z</th>
<th>df</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Initial &amp; 0-30 min</td>
<td>122.95</td>
<td>96.16</td>
<td>9.47</td>
<td>104.15; 141.74</td>
<td>12.97</td>
<td>102</td>
<td>0.000, S, p&lt;0.05</td>
</tr>
<tr>
<td>Initial &amp; 3-4 hrs</td>
<td>126.57</td>
<td>100.48</td>
<td>9.90</td>
<td>106.93; 146.21</td>
<td>12.78</td>
<td>102</td>
<td>0.000, S, p&lt;0.05</td>
</tr>
<tr>
<td>0-30 min &amp; 3-4 hrs</td>
<td>3.62</td>
<td>18.82</td>
<td>1.85</td>
<td>0.05; 7.29</td>
<td>1.95</td>
<td>102</td>
<td>0.054, NS, p&gt;0.05</td>
</tr>
</tbody>
</table>

By using Wilcoxon signed rank test, statistically significant difference was found between initial platelet count in EDTA anticoagulated blood with platelet count in CPT anticoagulated blood at 0-30 minutes ($z = 12.97; p = 0.000$) and that at 3-4 hours ($z = 12.78; p = 0.000$); but no statistical change was found in platelet count from 0-30 minutes to 3-4 hours ($z = 1.95; p = 0.054$) in CPT anticoagulated blood.

4. Discussion

The present study entitled, "Ethylenediaminetetraacetic acid (EDTA) - Dependent Pseudothrombocytopenia", was carried out in the Department of Pathology in a medical institution in central India. Total of 103 cases of suspected EDTA-dependent Pseudothrombocytopenia (EDTA-PTCP) were assessed for its correctness using manual platelet count as the gold standard. The platelet counts in two different anticoagulants (EDTA and CPT) were compared at different times from the time of collection of blood sample.

Prevention of platelet aggregation in EDTA-PTCP cases using CPT anticoagulant (Citric acid tri-sodium salt dehydrate, Pyridoxal 5'-phosphate, Tris/ hydroxymethyl/ aminomethane) is opined by Lippi et al [17], Lam et al [20]; Paparo et al [21] and Lippi and Facchinetti [22]. Most of the studies have tried different other anticoagulants to get correct platelet count in cases of EDTA-PTCP and have found CPT anticoagulant as a better alternative for EDTA in cases of EDTA-PTCP [15,17,20] similar to the present findings. The present study used manual platelet count to be the gold standard for platelet count in EDTA-PTCP cases [15, 17].

The study included 103 cases, of which 9 cases were found to be incorrect. Incidence of thrombocytopenia in 1.5% cases was noted and of total haemograms and 4.9% of total thrombocytopenia was observed. The study was comparable with incidence of 0.03-1.9% as reported by Sakurai et al [23].

The study compared the mean platelet count obtained in EDTA and from CPT anticoagulated blood at different time intervals with that of manual platelet count and with initial platelet count on which suspicion of PTCP was based.

The mean initial platelet counts in EDTA (103.67 ± 25.34 x 10^9/l) were much lower than the mean manual platelet count (222.63 ± 85.22 x 10^9/l) and the difference was statistically significant. The mean platelet count in EDTA anticoagulated blood at 0-30 minutes and at 3-4 hours (171.40 ± 78.10 x 10^9/l) and (171.63 ± 81.16 x 10^9/l) respectively,
though it was in the normal range, was also significantly lower than mean manual platelet count and the parallel automated platelet count in CPT anticoagulated blood (226.63 ± 93.25 x 10^3/l) and (230.25 ± 97.57 x 10^3/l) at 0-30 minutes and at 3-4 hours respectively (Table 1).

In 74 cases, the platelet counts were compared after 24 hours of preservation of blood sample. Even for this time, the mean platelet count in EDTA anticoagulated blood was significantly lower than that in CPT anticoagulated blood (Table 2). The present study observed the difference of mean initial platelet count in EDTA with manual platelet count of 55% and the difference of mean platelet count in EDTA and CPT anticoagulated blood of about 25% at 0-30 minutes and at 3-4 hours, and of 31% at 24 hours.

None of the studies reviewed have shown the comparison of the mean platelet counts in EDTA with manual platelet counts and with that in other anticoagulants, except for the study of Lippi et al [17] where they compared the automated platelet counts in EDTA and CPT anticoagulated blood in 4 different make cell counters with the manual mean platelet counts. The mean automated platelet counts in STKS counter counter in EDTA and CPT anticoagulated blood were found to be 181.75 ± 49.22 x 10^3/l and 256.25 ± 80.08 x 10^3/l respectively, whereas, the mean manual platelet count was 255.31 ± 82.82 x 10^3/l. These findings well correlate with that of the present study.

Different workers explained this phenomenon in different ways. The basic fact is there is in vitro binding of antibodies in the blood with some antigenic determinant on the platelet membrane in presence of EDTA which results in formation of platelet aggregates. The size of these platelet aggregates are beyond the upper limit of discrimination of platelet width of the automated cell counters and hence they are counted in the WBC channel and omitted from the platelet channel resulting in low platelet counts shown by automated cell counters.

The target antigen on platelet is a cryptic epitope that is normally hidden in platelet membrane glycoprotein; the glycoprotein being GP Ibb/Illa [24]. Although EDTA-PTCP have been reported in variety of diseases (autoimmune, neoplastic, liver, cardiovascular, viral etc., a documented trigger for the production of antiplatelet antibodies is unknown [25]. Lelie et al [26] showed that the binding of antiplatelet antibodies detected in patients with sepsis and normal platelets is completely or partially EDTA-dependent. They suggested, the damaged platelets in patients with sepsis could expose cryptic antigens and induce the synthesis of antiplatelet antibodies.

The antibodies are autoantibodies of all the major classes. But, IgG antibodies are much more frequently involved than IgM antibodies, and IgA antibodies are rarely involved [14, 27]. These autoantibodies are naturally occurring antibodies with antiplatelet activity, devoid of pathologic significance and are capable of recognizing cryptic antigens expressed by aged or damaged platelets to remove these from pathologic significance and are capable of recognizing cryptic antigens expressed by aged or damaged platelets.

Role of EDTA: The chelating effect of EDTA is in some way responsible for agglutination of platelets. The GP Ibb/Illa glycoprotein complex in platelet membrane requires the presence of calcium ions to maintain its heterodimeric structure. EDTA because of its chelating effect expresses by aged or damaged platelets to remove these from pathologic significance and are capable of recognizing cryptic antigens.

The proper collection of blood sample may cause thrombin release and a falsely low platelet count due to aggregation [22].

In the present study, the mean automated platelet counts in CPT anticoagulated blood at 0-30 minutes and at 3-4 hours as well as at 24 hours after blood collection were found to be comparable with the mean manual platelet counts (Table: 1, 2). Thus, the CPT anticoagulant was found to be a better anticoagulant for getting correct platelet counts in cases of EDTA-dependent pseudothrombocytopenia. These findings are consistent with that reported by Lippi et al [17].

Tri-sodium citrate do not alter cell counting and sizing after sampling in CPT mixture. Pyridoxal 5’-phosphate prevents platelet aggregation as it exhibits remarkable anti-aggregant and dis-aggregant effect in vitro. The pH is brought to neutrality by adding Tris to the CPT mixture.

Thus, inhibition of both platelet reaction and aggregation is prevented in CPT anticoagulant [17, 20]. Therefore, in routine hematological practice, CPT can be an alternative anticoagulant to K3EDTA, most suitable for automated complete blood count and useful in avoiding EDTA-induced platelet clumping.

PTCP is time – dependent phenomenon, gradually developing in 0-2 hours of venepuncture [30-31]. Platelet agglutination is detectable within minutes and maximum after 60-90 min. The magnitude of agglutination and the rate at which the clumping proceeded were strongly affected by the platelet concentration in the mixture. In most, the agglutination persisted without disaggregation for more than 24 hours [30]. The size of the aggregates approximates to that of the lymphocytes; often giving rise to suspect flag "platelet clumping" and/or flagging of the platelet parameters [31].

In the present study, comparison of platelet count in EDTA anticoagulated blood at different time intervals showed significant difference between initial platelet count and that at 0-30 minutes and between initial platelet count and that at 3-4 hours. But, no statistical change was found in platelet count from 0-30 minutes to 3-4 hours in EDTA anticoagulated blood (Table 3).

Similarly, the comparison of initial platelet count in EDTA with platelet counts in CPT anticoagulated blood at different time intervals showed significant difference between initial platelet count in EDTA anticoagulated blood with platelet count in CPT anticoagulated blood at 0-30 minutes and that at 3-4 hours. But no statistical change was found in platelet count from 0-30 minutes to 3-4 hours in CPT anticoagulated blood (Table 4).

The lower mean platelet count in initial EDTA blood sample on which EDTA-PTCP was suspected is probably because these samples were different and collected by the clinical residents and at different time. The samples which we personally collected and used for cell counts at 0-30 minutes and 3-4 hours, did not show significant difference of cell counts with lapse of time over upto 24 hours. However, the counts in EDTA anticoagulated blood were still significantly lower than that in manual counts and with CPT anticoagulated blood.

Most of the studies have tried different other anticoagulants to get correct platelet count in cases of EDTA-PTCP and have found CPT anticoagulant as a better alternative for EDTA in cases of EDTA-PTCP [15, 17, 20] similar to the present findings.

Pseudothrombocytopenia can complicate an accurate determination of platelet count even with an underlying thrombocytopenic disorder. Therefore, the presence of apparently obvious cause of thrombocytopenia should not be considered to rule out the diagnosis of EDTA-PTCP, which is confirmed by identifying the platelet clumping in EDTA anti-coagulated blood[6]. It is thus important to be able to distinguish between reduced platelet counts due to technique related variables or due to patient’s related medical condition [29].

5: Conclusions
Examination of well drawn peripheral blood smear for every case of thrombocytopenia is mandatory to rule out platelet clumping (PTCP). The new CPT mixture is an effective anticoagulant suitable for routine haematology and can be used as better alternative to EDTA in EDTA-PTCP cases. To get correct platelet count in these cases, the manual platelet count is the ‘gold standard’. Unrecognized PTCP may result in unnecessary laboratory testing, bone marrow aspirations and unwarranted transfusions and will prevent needless evaluations of thrombocytopenia and related therapeutic decisions.

Acknowledgement
There is no conflict of interest and no funding has been obtained for this research purpose.

References


Wu Wei, GUO Ye, ZHANG Lin, Cui Wei, Li Wei and Zhang Shuo. Clinical utility of automated platelet clump count in the screening for ethylene diamine tetraacetic acid-dependent pseudothrombocytopenia. Chinese Medical Journal 2011; 124(20) 3353-3357.


