Pneumatic tube system transport and false hyperkalemia related to leukocytosis: a retrospective analysis

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Abstract. Extreme leukocytosis may lead to false hyperkalemia when blood samples are conveyed by pneumatic tube system (PTS). The aim of this study was to define whether even moderate leukocytosis and also non malignancy cells like neutrophils may influence potassium values after PTS transportation. Materials and methods. Uncentrifuged blood samples are sent to the local laboratory through PTS. Data were retrospectively collected from routine testing carried out on all specimens arrived in the laboratory between September 2017 and March 2018. Clinical chemistry testing is routinely performed using lithium-heparin tubes. When false hyperkalemia induced by leukocytosis is suspected, potassium measurement is then performed in serum (clotting activator tubes) or whole blood samples. The analysis was focused on samples with both leukocytosis (i.e., \( >15 \times 10^9/L \)) and plasma potassium \( >5.0 \) mmol/L, before any corrective therapeutic measure to lower potassium levels was established. Results. A total number of 18 samples were included in our analysis, 9 drawn from patients with hematologic malignancies and 9 without. In the 9 patients without hematologic malignancies (median leukocyte count, \( 20.4 \times 10^9/L \)), the median potassium value was 5.4 mmol/L in plasma and 4.5 mmol/L in serum or whole blood. In the 9 patients with hematologic malignancies (median leukocyte count, \( 151.9 \times 10^9/L \); \( p <0.001 \)), the median potassium value was 7.7 mmol/L in plasma and 4.3 mmol/L in serum or whole blood (median difference, 2.9 mmol/L; \( p <0.001 \)). Conclusion. The results of our study suggest that even modest leukocytosis (i.e., around \( 15 \times 10^9/L \)), which can be frequently encountered in clinical practice, may be associated with a significant variation of plasma potassium. This would lead us to conclude that plasma samples transportation by PTS should be avoided in patients with even mild leukocytosis.

Key words: potassium, pneumatic tube system, variability, reference range
à des échantillons présentant à la fois une hyperleucocytose (> 15 × 10⁹/L) et un potassium plasmatique > 5 mmol/L avant toute thérapeutique visant à réduire la kaliémie. Résultats. Un total de 18 échantillons ont été inclus dans notre analyse, 9 provenant de patients avec hémopathie et 9 sans hémopathie.

Chez les 9 patients sans pathologies hémato logicques (valeur médiane de leucocytose à 20,4 × 10⁹/L), la valeur médiane de potassium était de 5,4 mmol/L sur plasma et 4,5 mmol/L sur sérum ou sang total. Chez les 9 patients avec pathologies hémato logicques (valeur médiane de leucocytes à 151,9 × 10⁹/L), la valeur médiane de potassium était de 7,7 mmol/L sur plasma et 4,3 mmol/L sur sérum ou sang total. Conclusion. Les résultats de notre étude suggèrent que même une leucocytose modérée à partir de 15 x 10⁹/L, fréquemment rencontrée en pratique clinique, peut être associée à une variation significative du potassium plasmatique. Ceci nous amène à une vigilance particulière sur les résultats de biochimie issus d’échantillons plasmatiques en cas de leucocytose modérée après transport par pneumatique.

Mots clés : potassium, transport pneumatique, variabilité, valeurs de référence

Potassium is a common but essential parameter in clinical chemistry testing. Severe or sudden hyperkalemia is a diagnostic and therapeutic, emergency, since it can trigger life-threatening cardiac rhythm disturbances. Albeit pneumatic tube system (PTS) is a frequent method for transporting blood samples transportation in many hospitals, it may be the cause of some preanalytical issues such as hemolysis and blood cell lysis, which would finally lead to spuriously increased values of some biochemistry parameters displaying intracellular concentration higher than in blood [1-3].

Pseudo or spurious hyperkalemia is conventionally defined as spurious elevation of measured potassium whilst in vivo potassium concentration is normal. Many causes can induce spurious hyperkalemia, the most frequent of which is hemolysis and leakage of potassium from blood cells [4]. Pseudo-hyperkalemia caused by uncentrifuged samples transportation by PTS in patients with extreme leukocytosis has been well documented over the past decades [5, 6]. This is mainly due to the fact that leukocytes may be injured and intracellular components (including potassium and several enzymes) are released in blood, especially in patients with fragile leukemic cells, which display the highest fragility [2-4].

In patients with leukemia, spurious hyperkalemia may be very dangerous, since it may trigger inappropriate therapeutic actions aimed at lowering an otherwise normal in vivo concentration of this essential electrolyte. Previous reports showed that this effect in higher in plasma than in serum, since the blood cells entrapped into the clot are less vulnerable to transportation injury [7]. The centrifugation of anticoagulated samples could then enhance propensity of blood cell lysis after PTS, so that potassium measurement in serum or whole blood (i.e. with direct potentiometry) may be effective to overcome the risk of blood cell injury-related hyperkalemia.

Spurious hyperkalemia after PTS transportation of uncentrifuged blood samples has not been described in patients without hematologic malignancies to the best of our knowledge. Some occasional cases of in vitro plasma hyperkalemia have been observed in patients with non-leukemic leukocytosis, even in blood samples with modestly elevated to high leukocyte counts (i.e., > 15 × 10⁹/L) and no other obvious causes of pseudo hyperkalemia. Although this event is basically rare in routine laboratory practice, it shall not be overlooked because the establishment of unjustified potassium lowering therapies may then trigger adverse consequences on patients’ health [8].

Therefore, aim of this study was to define whether even moderate leukocytosis and also non malignancy cells like neutrophils may influence plasma potassium values after uncentrifuged blood samples transportation by PTS.

Methods

Patient selection and data collection

Data extraction was retrospective performed using the laboratory information management system (LIMS) MOLIS 4.3 software (Compugroup, Nevele, Belgium). The study population consisted of all patients undergoing routine laboratory testing at Lille University Hospital, between September 2017 and March 2018. Patients received detailed information that their clinical data and/or residual blood samples after routine testing had been completed could be used for research purposes. All data were retrieved from the human biological database previously authorized by the
False hyperkalemia related to leukocytosis

French Ministry of Research (No. DC-2008-642). Therefore, no written informed consent was needed for this study. Exclusion criteria were spurious hyperkalemia due to hemolysis (i.e., hemolysis index >100 μmol/L). The hemolysis index is assessed qualitatively in the local laboratory; no hemolysis defined in samples with <20 μmol/L of cell-free hemoglobin, whilst light hemolysis is defined in those with cell-free hemoglobin concentration comprised between 20-50 μmol/L. We also have excluded EDTA contamination, as defined by abnormally low calcium values associated with high potassium concentration, blood drawn from (or above) an intravenous infusion site (i.e. displaying abnormally low or high concentration of analytes biased by infusion, such as glucose and sodium), >2 hours delay between sample collection and centrifugation as identified from the local laboratory information system [9]. Patients with platelet count >400×10⁹/L were also excluded, since thrombocytosis is another well-known cause of spurious hyperkalemia [10].

The analysis was hence limited to patient samples simultaneously drawn in a lithium-heparin tube (4 mL BD Vacutainer™ Heparinized Tube [HT]; Becton Dickinson, Franklin Lakes, NJ, United States), a clotting activator blood tubes (4 mL BD Vacutainer™ Clot Activator Tubes [CAT]; Becton Dickinson, Franklin Lakes, NJ, United States) or whole-blood heparin syringes, displaying both leukocytosis (i.e., >15×10⁹/L) and high plasma concentration (i.e., >5.0 mmol/L), with potassium measured before corrective actions to lower hyperkalemia were established [7].

Control potassium
Plasma potassium measured in lithium-heparin blood tubes was compared with potassium values measured in two additional samples matrices (i.e., “control” potassium). The first possible control measurement was carried out in serum (i.e., CAT tubes) [7], whilst the second possible control measurement was performed using direct potentiometry in whole-blood heparin syringes (figure 1).

Laboratory measurements
All blood tubes were conveyed to the central laboratory by PTS (Swisslog, Switzerland), within 30 min from collection. The tubes are introduced into a first disposable plastic bag, which is then placed in a second bag propelled in a one-way air-flow system, with speed limited at 6 m/s and softlanding capabilities. To avoid bias related to centrifugation conditions, plasma and serum samples were immediately centrifuged both at 4,000 g for 5 min. All samples were kept at room temperature throughout the study period.

The concentration of potassium in lithium-heparin plasma and serum was measured with Roche Cobs 8000® PLCs (Roche Diagnostics, Risch-Rotkreuz, Switzerland), whilst whole-blood potassium was measured using ABL800 FLEX (Radiometer®). The local reference ranges of potassium are 3.4-4.5 mmol/L for plasma and 3.5-5.1 mmol/L for serum. Creatinine was measured on Roche Cobs 8000® module c701/702 (Roche Diagnostics, Bâle, Suisse) using enzymatic assays. White blood cell count (WBC) neutrophils, lymphocytes, and platelets count were measured in ethylenediaminetetraacetic (EDTA)-anticoagulated blood (BD Vacutainer™; Becton Dickinson, Franklin Lakes, NJ, United States), using a Sysmex XN (Sysmex Corporation, Kobe, Japan). All samples were measured using the same analyzers throughout the study period.

Statistical analysis
Data analysis was performed with paired Wilcoxon ranked test using R software (www.R-project.com) and Graph-Pad Prism software v6.0. Statistical significance was set at p<0.05.

Results
A total number of 23 samples were initially identified according to our inclusion and exclusion criteria. Three of these specimens ought to be excluded due to hemolysis and 2 additional samples were excluded because of suspected sample contamination by intravenous infusions. Therefore, a total number of 18 samples were finally included in our analysis. Biological data are shown in tables 1 and 2. Nine patients had leukocytosis associated with inflammatory diseases (neutrophil count was also available for 3 of these). The other 9 patients all had hematologic malignancies, including chronic lymphoid leukemia (CLL) (n=8) and acute lymphoid leukemia (ALL) (n=1).

In the 9 patients without hematologic malignancies, the median potassium value was 5.4 mmol/L in plasma and 4.5 mmol/L in serum or whole blood (i.e., “control” matrix), with a median difference of 0.5 mmol/L. The median WBC count of these patients was 20.4×10⁹/L (figure 2A). In the 9 patients with CLL or ALL, the median potassium value was 7.7 mmol/L in plasma potassium and 4.3 mmol/L in serum or whole blood (median difference, 2.9 mmol/L). The median WBC count of these patients was 151.9×10⁹/L (figure 2B and table 3). A highly significant difference was observed between potassium measurement in plasma and in the other two control matrices, in both patients with or without hematologic malignancies (both p<0.001) (figure 2).
Original article

Plasma potassium level >5 mmol/L

Known etiology of high potassium levels according to clinic (renal failure, cardiac troubles…)

yes

Validation of plasma potassium measurement

Known causes of spurious high potassium level:
- Hemolysis
- >2 hours delay between sample collection and centrifugation
- EDTA contamination
- Blood drawn from infusion

yes

No validation of plasma potassium measurement, new sample required

Leukocytes levels >15×10^9/L

no

Validation of plasma potassium measurement, with controlled sample required according to potassium levels

Different controlled sample according to available sample on laboratory (or new controlled sample required)

Serum available: potassium measurement on serum

no

Concordance of results

yes

Validation of plasma potassium measurement

Blood gas analysis performed: potassium measurement on whole blood

no

No validation of plasma potassium measurement

Validation of controlled potassium measurement (serum or whole blood) and new sample required

Figure 1. Flow chart of the retrospective selection of patients with exclusion criteria.

Discussion

Spuriously, PTS-induced hyperkalemia has been occasionally described in leukemic patients with consistent leukocytosis (i.e., >100×10^9/L), but no definitive information has been published in blood samples with modestly increased WBC count to the best of our knowledge. The results of this study suggest that plasma potassium values may be influenced by the WBC count, even in those with modest leukocytosis. This important bias was found not only in samples drawn from patients with hematological malignancies, but also in those without leukemias. Our study has a limitation that we do not made comparison with human courier transport, however our recent published case report seems to show that tube effect is most important than transport mode [11]. These results are of pivotal significance for the managed care of patients with leukocytosis. In PTS-induced plasma potassium increase not only can trigger unjustified potassium-lowering therapies in patients with hyperkalemia, but may also lead to misdiagnosing a real hypokalemia, which could be falsely interpreted as normokalemia due to the spuriously increased potassium values. In this specific case, the low potassium value may remain unnoticed, thus leaving the patients without appropriate therapeutic management. This second aspect is especially important because normal potassium values are not typically verified in routine clinical and laboratory
Table 1. Biological data of patients without hematological disorder. Plasma and control potassium in mmol/L after transport by pneumatic tube system. A negative hemolysis is less than 20 μmol/L whereas a light hemolysis is between 20 and 50 μmol/L.

<table>
<thead>
<tr>
<th>Patient n°</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
<th>7</th>
<th>8</th>
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<td>no</td>
<td>light</td>
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<td>no</td>
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<tr>
<td>Plasma Potassium (mmol/L)</td>
<td>5.1</td>
<td>5.4</td>
<td>6</td>
<td>7.2</td>
<td>6</td>
<td>5.8</td>
<td>5.1</td>
<td>5</td>
<td>5</td>
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<tr>
<td>Control Potassium (mmol/L)</td>
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<td>4.5</td>
<td>5.6</td>
<td>6.3</td>
<td>5.5</td>
<td>4.5</td>
<td>4.5</td>
<td>4.5</td>
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<tr>
<td>∆ potassium (mmol/L)</td>
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<td>0.9</td>
<td>0.4</td>
<td>0.9</td>
<td>0.5</td>
<td>1.3</td>
<td>0.6</td>
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<td>0.5</td>
</tr>
<tr>
<td>Type of potassium control</td>
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<td>WB</td>
<td>WB</td>
<td>WB</td>
<td>CAT</td>
<td>WB</td>
<td>WB</td>
<td>WB</td>
<td>WB</td>
</tr>
<tr>
<td>WBC count (×10⁹/L)</td>
<td>35.3</td>
<td>20.4</td>
<td>26.6</td>
<td>20.9</td>
<td>14.7</td>
<td>25.4</td>
<td>18.1</td>
<td>16.2</td>
<td>16.2</td>
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<td>Neutrophils (×10⁹/L)</td>
<td>17.1</td>
<td>11.5</td>
<td>11.5</td>
<td>11.5</td>
<td>11.5</td>
<td>11.5</td>
<td>11.5</td>
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<td>11.5</td>
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<tr>
<td>Creatinine (μmol/L)</td>
<td>176</td>
<td>44</td>
<td>132</td>
<td>79</td>
<td>79</td>
<td>79</td>
<td>79</td>
<td>79</td>
<td>79</td>
</tr>
<tr>
<td>Platelet count (×10⁹/L)</td>
<td>325</td>
<td>380</td>
<td>89</td>
<td>14</td>
<td>276</td>
<td>300</td>
<td>121</td>
<td>294</td>
<td>208</td>
</tr>
</tbody>
</table>

WBC: white blood cells, WB: whole blood, CAT: clot activator tube.

Table 2. Biological data of patients with hematological disorder. Plasma potassium in mmol/L and control potassium in mmol/L after transport by pneumatic tube system. A negative hemolysis is less than 20 μmol/L whereas a light hemolysis is between 20 and 50 μmol/L.

<table>
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<th>Patient n°</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
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<td>CLL</td>
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<td>CLL</td>
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<td>CLL</td>
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<td>CLL</td>
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<td>light</td>
<td>light</td>
<td>light</td>
<td>light</td>
<td>no</td>
<td>no</td>
<td>light</td>
<td>no</td>
</tr>
<tr>
<td>Plasma Potassium (mmol/L)</td>
<td>6.9</td>
<td>6.5</td>
<td>7.7</td>
<td>9</td>
<td>8</td>
<td>5.1</td>
<td>9.9</td>
<td>7.7</td>
<td>5.4</td>
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<tr>
<td>Control Potassium (mmol/L)</td>
<td>4.4</td>
<td>3.8</td>
<td>4.3</td>
<td>4.2</td>
<td>5.1</td>
<td>4.2</td>
<td>4.9</td>
<td>4.3</td>
<td>4.8</td>
</tr>
<tr>
<td>∆ potassium (mmol/L)</td>
<td>2.5</td>
<td>2.7</td>
<td>3.4</td>
<td>4.8</td>
<td>2.9</td>
<td>0.9</td>
<td>5</td>
<td>3.4</td>
<td>0.6</td>
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<tr>
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<td>CAT</td>
<td>CAT</td>
<td>CAT</td>
<td>CAT</td>
<td>CAT</td>
<td>WB</td>
<td>WB</td>
</tr>
<tr>
<td>WBC count (×10⁹/L)</td>
<td>144.8</td>
<td>104.6</td>
<td>151.9</td>
<td>261</td>
<td>238.4</td>
<td>186.3</td>
<td>487.7</td>
<td>81.8</td>
<td>26.7</td>
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<tr>
<td>Lymphocytes (×10⁹/L)</td>
<td>139</td>
<td>90.9</td>
<td>93</td>
<td>245</td>
<td>209.7</td>
<td>180.9</td>
<td>77.7</td>
<td>12.1</td>
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<tr>
<td>Creatinine (μmol/L)</td>
<td>79</td>
<td>70</td>
<td>70</td>
<td>53</td>
<td>79</td>
<td>62</td>
<td>62</td>
<td>88</td>
<td>70</td>
</tr>
<tr>
<td>Platelet count (×10⁹/L)</td>
<td>118</td>
<td>83</td>
<td>148</td>
<td>152</td>
<td>220</td>
<td>210</td>
<td>126</td>
<td>37</td>
<td>178</td>
</tr>
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</table>


Figure 2. A. Median and interquartile ranges (IQR) of plasma or control potassium values in patients with hematological disorder after transport by pneumatic tube system (n=9). B. Median and interquartile ranges (IQR) of plasma or control potassium values in patients without hematological disorder after transport by pneumatic tube system (n=9). Control potassium by serum or whole blood measurement. Difference were analyzed with paired Wilcoxon test, **p<0.01, ***p<0.001.

practice, neither when potassium is measured in samples of patients with leukocytosis, particularly in those without hematologic disorders. In agreement with available literature data [7], we also found that the PTS-induced potassium bias was lower in serum or whole blood compared to the plasma.

Albeit the retrospective nature of this study may be seen as a limitation, our results suggest that even modest leukocyto-
Table 3. Potassium values in plasma and control (whole blood or serum) after transport by pneumatic tube system, and potassium differences (Δ potassium) and white blood cells count (WBC) in patients without hematological disorder compared to patient with hematological disorder. Control potassium by serum or whole blood measurement.

<table>
<thead>
<tr>
<th></th>
<th>Median</th>
<th>Min</th>
<th>Max</th>
<th>IQR 25</th>
<th>IQR 75</th>
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<tbody>
<tr>
<td>Patients with hematological disorder (n=9)</td>
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<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Plasma Potassium (mmol/L)</td>
<td>7.7</td>
<td>5.1</td>
<td>9.9</td>
<td>6.5</td>
<td>8</td>
</tr>
<tr>
<td>Control Potassium (mmol/L)</td>
<td>4.3</td>
<td>3.8</td>
<td>5.1</td>
<td>4.2</td>
<td>4.8</td>
</tr>
<tr>
<td>Δ potassium (mmol/L)</td>
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<td>0.6</td>
<td>5</td>
<td>2.5</td>
<td>3.4</td>
</tr>
<tr>
<td>WBC count (× 10^9/L)</td>
<td>151.9</td>
<td>26.7</td>
<td>487.7</td>
<td>104.6</td>
<td>238.4</td>
</tr>
<tr>
<td>p</td>
<td>0.002</td>
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<td>Plasma Potassium (mmol/L)</td>
<td>5.4</td>
<td>5</td>
<td>7.2</td>
<td>5.1</td>
<td>6</td>
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<tr>
<td>Control Potassium (mmol/L)</td>
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<td>4.5</td>
<td>6.3</td>
<td>4.5</td>
<td>5.5</td>
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<tr>
<td>Δ potassium (mmol/L)</td>
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<td>0.9</td>
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<td>WBC count (× 10^9/L)</td>
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<td>15</td>
<td>35.3</td>
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<tr>
<td>p</td>
<td>0.002</td>
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</table>

Median and interquartile are calculated using GraphPad Prism software. p-values are calculated with Wilcoxon paired signed rank test.

sis (i.e., around 15x10^9/L), which is frequently encountered in clinical and laboratory practice, may be associated with PTS-induced spurious elevation of plasma potassium. This would lead us to conclude that plasma samples transportation by PTS should be avoided in patients with even modest leukocytosis. Moreover, we also put forward the hypothesis that test results of plasma potassium shall be accompanied by the leukocyte count, so that alertness can be raised that plasma potassium may be falsely elevated in patients with an increased WBC count.

Conflict of interest: None of the authors has any conflict of interest to disclosure.

References


