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Short Communication

Electrolyte-balanced heparin in blood gas syringes can introduce a significant bias in the measurement of positively charged electrolytes

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Abstract

Background: Heparin binds positively charged electrolytes. In blood gas syringes, electrolyte-balanced heparin is used to prevent a negative bias in electrolyte concentrations. The potential pre-analytical errors introduced by blood gas syringes are largely unknown. Here, we evaluate electrolyte concentrations in non-anticoagulated blood compared with concentrations measured in electrolyte-balanced blood gas syringes.

Methods: Venous blood was collected into plain tubes. Ionized calcium, potassium, sodium and hydrogen ions were analyzed directly using a blood gas analyzer and the remaining blood was collected into different blood gas syringes in random order: Preset (Becton Dickinson), Monovette (Sarstedt) and Pico 50-2 (Radiometer).

Results: Ionized calcium and sodium concentrations were significantly lower in blood collected in Becton Dickinson and Sarstedt syringes compared to non-heparinized (NH) blood. The mean bias exceeded biological variation-based total allowable error, which in most cases leads to clinically misleading individual results. In contrast, ionized calcium concentrations in blood collected in Pico 50-2 syringes were identical to values obtained from NH blood. Sodium showed a minor, yet statistically significant, bias.

Conclusions: Despite the fact that blood gas syringes now contain electrolyte-balanced heparin, one should be aware of the fact that these syringes can introduce pre-analytical bias in electrolyte concentrations. The extent of the bias differs between syringes.

Keywords: blood gas syringes; electrolyte-balanced heparin; ionized calcium; potassium; pre-analytical errors; sodium.

Commercially available syringes for the analysis of blood gases and electrolytes in whole blood contain heparin as an anticoagulant. Heparin has the disadvantage of binding positively charged electrolytes in whole blood (1), and is especially notorious for the introduction of pre-analytical errors in the measurement of ionized calcium (2). To prevent a negative bias in reported ionized calcium concentrations, blood gas syringes containing electrolyte-balanced heparin are commonly used. The use of electrolyte-balanced blood gas syringes have been shown to reduce the negative bias on the measurement of ionized calcium and other positively charged electrolytes (3), although others still find heparin-induced pre-analytical errors (4). A discrepancy in results from electrolyte-balanced syringes is likely to originate from differences in the manufacturing process of balanced heparin. Therefore, we evaluated commercially available syringes for the bias on positively charged electrolytes in whole blood.

The effects of heparin-containing syringes on ionized calcium, sodium, potassium and pH were determined in whole blood collected from 16 healthy donors in non-heparinized (NH) tubes and in electrolyte-balanced dry heparin containing syringes from three different manufactures: Becton Dickinson (BD) preset (heparin 80 IU), Sarstedt (SS) monovette (heparin 50 IU) and Radiometer (RM) Pico 50-2 (heparin 80 IU). The order of blood collection into the different syringes showed no effect on the concentration of the measured electrolytes (data not shown). All data sets were normally distributed (Shapiro-Wilk test, data not shown) and data sets were analyzed using the two-tailed Student's t-test.

The absolute concentration of each electrolyte and pH, collected in the different syringes, was related pairwise to the respective concentration in NH blood. Figure 1A shows the mean percent deviation (and range of results) of the analyte concentration in syringes compared to the concentration in NH blood. Values in NH blood were set at 100%. Ionized calcium concentrations were significantly lower in blood collected in syringes from BD and SS compared to ionized calcium concentrations in NH blood ($p<0.0001$). Mean ionized calcium concentrations were –4.4% (BD) and –2.8% (SS) lower than those in NH blood (Figure 1A). The negative bias is clinically relevant for most subjects since it exceeds the total allowable error (TE) of 2.1% for ionized calcium (5). In contrast, ionized calcium measured in blood collected in RM syringes was identical to values obtained from NH blood ($p=1.0$). Ionized calcium concentrations were determined at the actual sample pH and could therefore be subject to bias introduced through differences in pH. The pH values were slightly influenced by the use of different syringes (mean of 7.37, 7.39, 7.38 and 7.38 for NH, BD, SS and RM, respec-

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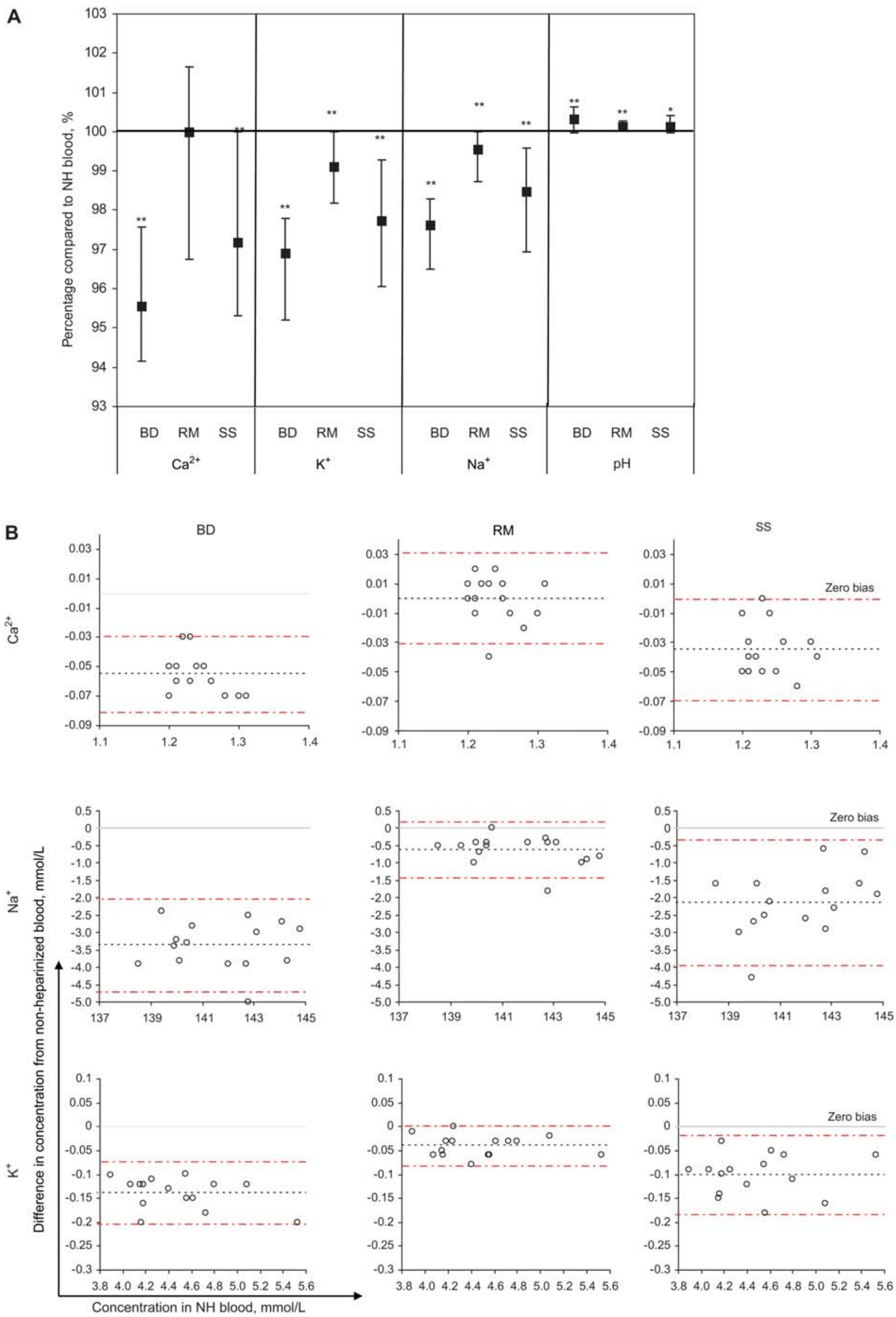
A
Percentage compared to NH blood, %
103
102
101
100
99
98
97
96
95
94
93
BD RM SS
Ca2+
K+
Na+
pH
**
*
B
BD
RM
SS
Ca2+
Na+
K+
Zero bias
Difference in concentration from non-heparinized blood, mmol/L
Concentration in NH blood, mmol/L

Figure 1 Introduction of a negative bias in positively charged electrolyte concentration by different blood gas syringes containing electrolyte-balanced heparin.
Venous blood (NH blood) was collected into a tube without additives and free of separator gel (Vacutainer ref. 368500, Becton Dickinson BV, Breda, The Netherlands) and measured immediately using a blood gas analyzer (ABL 735, Radiometer, Zoetermeer, The Netherlands). Within 1 min after venipuncture, 1 mL of NH blood was aspirated into three different blood gas syringes in random order: preset by Becton Dickinson (BD) (ref. 364390, Breda, The Netherlands), monovette by Sarstedt (SS) (ref. 051147020, Etten-Leur, The Netherlands) and Pico 50-2 by Radiometer (RM) (ref. 956-552VR10, Zoetermeer, The Netherlands). Blood was collected from 16 volunteers. Ionized calcium (Ca^{2+}), potassium (K^+), sodium (Na^+) and hydrogen ion (pH) concentrations were determined. All measurements were completed within 15 min after venipuncture. (A) The absolute concentration of each electrolyte and pH collected in the different syringes was related pairwise to the respective concentration in NH blood. The concentration in NH blood was set at 100%. The mean of the percentage deviation from values in NH blood (solid squares) and the range of the relative concentrations (bars) are shown. Significant differences are depicted (**$p<0.0001$, *$p=0.0003$). (B) Bland-Altman bias plots of the absolute electrolyte concentrations measured in blood from individual subjects collected in heparinized syringes compared to electrolyte concentration in NH blood. The x-axis shows the electrolyte concentration in NH blood. The y-axis shows the difference in either Ca^{2+}, Na^+ or K^+ concentration between NH blood and blood collected in BD, RM or SS syringes. The thin, dotted lines indicate zero bias, the evenly stippled lines the observed bias, and the unevenly stippled lines show the 95% limits of agreement. Bland-Altman analysis was performed using Analyse-it Version 1.72 (Analyse-it Software, Ltd, Leeds, UK).

tively) (Figure 1A and data not shown), and thus made a minor contribution to the differences found in ionized calcium concentrations between the syringes. For sodium, a statistically significant negative bias was introduced by using BD (–2.4%, $p<0.0001$) and SS syringes (–1.5%, $p<0.0001$), exceeding the TE for sodium of 0.9%. Sodium measured in blood collected in RM syringes caused only minimal, though statistically significant bias (–0.4%, $p<0.0001$). Potassium concentrations in whole blood collected in RM, BD and SS syringes were repeatedly and significantly lower compared to those found in NH blood (mean difference of –0.9%, –3.1% and –2.4%, respectively, all $p<0.0001$). The syringe introduced bias on potassium concentrations did not exceed the TE for potassium (5.8%). Therefore, the clinical significance is limited.

The biases in absolute electrolyte concentrations between NH and the heparinized syringes were assessed using Bland-Altman analysis (6). BD syringes introduced the most pronounced bias in concentrations of all electrolytes, while the biases introduced by RM syringes were minimal (Figure 1B). Similarly, the 95% limits of agreement were the narrowest and always encompassed zero bias with RM syringes.

Taken together, two out of the three syringes tested here introduced a clinically significant negative bias for many subjects in both ionized calcium concentrations and sodium concentrations, despite the fact that all syringes contain electrolyte balanced heparin. These findings were not caused by inadequate filling volumes since the negative bias persisted after filling the syringes to more than the minimally necessary volume (according to manufactures instruction, data not shown). The mean bias exceeded the TE for ionized calcium and sodium, which are based on intra-individual biological variation. Therefore, a clinically significant difference in results can be introduced by using the syringes from BD or SS for analysis of these electrolytes. Electrolyte values in blood collected into syringes manufactured by RM were most comparable to values measured in NH blood.

ABL blood gas analyzers do not use an algorithm to correct for a potential heparin bias caused by heparin in RM syringes (personal communication with H. Froklage, Radiometer). Therefore, the extent of interference found for the ABL blood gas analyzer used in this study is expected to be reproduced on an analyzer from a different manufacturer. Differences in the process of balancing heparin with electrolytes between manufacturers most likely explain the findings described here.

In conclusion, one should be aware of the bias that can be introduced in electrolyte concentrations by the use of different blood gas syringes, despite the fact that blood gas syringes now contain electrolyte balanced heparin. These results emphasize the need for improving the manufacturing process of balanced heparin. Furthermore, a single type of electrolyte-balanced heparin should be used in one laboratory only in order to minimize the pre-analytical effects on variation of sodium, potassium and ionized calcium concentrations.

The experimental procedures were in accordance with the Declaration of Helsinki, Dutch law and the standards of the medical Ethical Commission of our institution. Informed consent was obtained from each subject before blood sampling.

Conflict of interest statement

Authors' conflict of interest disclosure: The authors stated that there are no conflicts of interest regarding the publication of this article.
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References

1. Dimeski G, Badrick T, St John A. Ion selective electrodes (ISEs) and interferences – a review. Clin Chim Acta 2010;411:309–17.
2. Sachs C, Rabouine P, Chaneac M, Kindermans C, Dechaux M,

Falch-Christiansen T. Preanalytical errors in ionized calcium measurements induced by the use of liquid heparin. Ann Clin Biochem 1991;28:167–73.
3. Toffaletti J, Ernst P, Hunt P, Abrams B. Dry electrolyte-balanced heparinized syringes evaluated for determining ionized calcium and other electrolytes in whole blood. Clin Chem 1991;37:1730–3.
4. Swanson JR, Heeter C, Limbocker M, Sullivan M. Bias of ionized calcium results from blood gas syringes. Clin Chem 1994;40:669–70.
5. Ricos C, Alvarez V, Cava F, Garcia-Lario JV, Hernandez A, Jimenez CV, et al. ‘‘Current databases on biologic variation: pros, cons and progress.’’ Scand J Clin Lab Invest 1999;59:491–500.
6. Bland JM, Altman DG. Statistical methods for assessing agreement between two methods of clinical measurement. Lancet 1986;i:307–10.