Technique to Remove Interference Caused by Radio-Opaque Agents in Clinical Capillary Zone Electrophoresis

To the Editor:
We read with great interest the report “Interference of Radio-opaque Agents in Clinical Capillary Zone Electrophoresis” (1). A laboratory sent two similar patient samples for our evaluation. Both patients had received intravenous sodium meglumine ioxitalamate (Telebrix), a radio-opaque contrast material that absorbs at 214 nm. The interfering substance was not visible on agarose gels (Paragon® SPE), whereas the capillary zone electrophoresis (CZE) results (Paragon CZE® 2000) showed a peak in the alpha-2 zone, which may be mistaken for a monoclonal protein (see Fig. 1A). The anomalous peak was not removed by reaction with antisera to any of the immunoglobulins.

The samples were then desalted (D-Salt Dextran plastic desalting columns, 5-kDa cutoff; Pierce). Several similar products are available. After capillary electrophoresis, the desalted samples (see Fig. 1B) appeared normal with the questionable peak entirely removed, thus confirming that the interfering substance was of low molecular weight and, therefore, not a paraprotein. The desalting procedure was fast and simple. Clinical laboratories that use CZE could easily use this method to evaluate questionable samples.

This information was presented previously as part of a technical poster, “Detection of Non-Protein Peaks in Serum by Capillary Zone Electrophoresis”, by C. Blessum, M. Andres, and N. Khatter, at the 12th International Symposium on Capillary Electrophoresis and Related Microscale Techniques, January 23–28, 1999, in Palm Springs, CA.

References

Fig. 1. CZE electropherograms of a patient serum sample before (A) and after (B) desalting. (A), the peak for the radio-opaque agent is indicated by the arrow.

Comment on the Overestimation of Methemoglobin Concentrations in Neonatal Samples with the Chiron 800 System CO-Oximeter Module

To the Editor:
We wish to comment on the Technical Brief of Lynch et al. (1), who reported overestimation of methemoglobin concentrations in neonatal samples containing fetal hemoglobin and analyzed with the Chiron 800 system CO-oximeter. Overestimation of methemoglobin in the presence of fetal hemoglobin may have serious implications, with the possibility of premature termination of nitric oxide treatment. In our hospital, >1000 methemoglobin analyses are performed annually on request of our neonatal intensive care unit. These measurements have been performed with a CIBA Corning 270 CO-oximeter and, at present, are also performed with a Chiron 865 blood gas analyzer equipped with a CO-oximetry module (Chiron Diagnostics NV).

We decided to investigate this alarming report and used both analyzers to measure methemoglobin concentrations in three sets of samples. The first set consisted of 10 samples from our neonatology department. In these samples, the concentration of fetal hemoglobin was 65% or higher. The second and third sets of samples consisted of adult blood and umbilical cord blood, respectively, collected in heparin-containing tubes and divided into two portions. In one portion of each sample, 100% methemoglobin was obtained by adding potassium nitrite...
The other portion was not treated (<2% methemoglobin). All portions were washed with saline several times and subsequently used to produce samples with nominal percentages of methemoglobin of 2–25%. Samples were measured within 2 min on both blood gas analyzers. The results are given in Table 1.

In the samples from neonates, no statistical differences between the methemoglobin percentages measured with the Corning 270 and the Chiron 865 analyzer were observed (paired t-test, P < 0.05). The results obtained for adult blood and umbilical cord blood revealed that measurement of 5% methemoglobin or less was not markedly influenced by the presence of fetal hemoglobin on both analyzers. Above 10% methemoglobin, the Corning 270 blood gas analyzer has a tendency to underestimate the amount of methemoglobin compared with the Chiron 865 analyzer. This, however, was not influenced by the presence or absence of fetal hemoglobin because the discrepancy was present in adult blood as well as umbilical cord blood. These findings were also reported by Lynch et al. (1).

The divergence between our results and those of Lynch et al. in the low methemoglobin range (<5%) may be explained by the different software versions that were used. The analyzer used by Lynch et al. was equipped with software that uses 10 wavelengths to calculate the various hemoglobin fractions. The new software program (Ver. 4.4c) implemented in our Chiron 865 blood gas analyzer uses 40 wavelengths, including wavelengths in the optimal region for the measurement of methemoglobin.

We conclude that the Chiron 865 analyzer equipped with the 40-wavelength software (Ver. 4.4c) is suitable for measuring methemoglobin concentrations in the clinically relevant range, even in samples with high concentrations of fetal hemoglobin.

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References

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