enches between the results produced by the various immunoassay methods, which produce a variable bias compared with a selective technique, require careful scrutiny at sample time points other than the traditional predose (trough) measurement. As shown above, and from interlaboratory comparisons (11), there is ample evidence demonstrating differences between analytical techniques for trough CsA concentration measurements, but these differences need to be studied for samples collected in the period 1–6 h after ingestion of CsA. We will be investigating this issue more fully in a series of controlled studies to be performed in the near future.

In addition, we hope that the manufacturers of commercially available CsA analytical systems will respond to changes in CsA TDM practices. For this, they should address the need for an increase in the ranges of their assay calibrators, as well as ensuring that validated dilution protocols are available for their customers.

References

Interference by Glycolic Acid in the Beckman Synchron Method for Lactate: A Useful Clue for Unsuspected Ethylene Glycol Intoxication, William H. Porter,* Margaret Crettin, Philip W. Rutter, and Peter Oeltgen*1,2 (1 Department of Pathology and Laboratory Medicine, University of Kentucky Medical Center, Lexington, KY 40536; 2 Lexington VA Hospital, Lexington, KY 40536; *author for correspondence: fax 606-257-8932, e-mail wporter@pop.uky.edu)

In the differential evaluation of patients with high anion gap metabolic acidosis of unknown origin, lactate determinations are frequently performed. For patients who ingest ethylene glycol (present in antifreeze), the high anion gap metabolic acidosis is the result of the metabolism of ethylene glycol to glycolic acid (1, 2).

We encountered unusual lactate results, when measured on the Beckman LX 20 (Beckman Coulter), for two patients who had ingested ethylene glycol. Specifically, the lactate results were suppressed (i.e., no result) with an appended error message, “rate high”. When these specimens were diluted threefold, measurable lactate values were obtained.

The Beckman lactate method is based on a lactate oxidase/peroxidase coupled reaction with endpoint determination. The lactate concentration is determined from the absorbance (A) measurement taken after reaction equilibrium has been established. To ensure an equilibrium steady state, a rate measurement is made during the expected steady-state portion of the measurement period. A reaction rate ≥ 10 mA/min would indicate a nonequilibrium reaction condition and would lead to suppressed results and a “rate high” error flag.

We suspected, based on their structural similarities, that glycolate reacted as a poor substrate for lactate oxidase, generating a reaction rate > 10 mA/min during the expected steady-state portion of the lactate reaction and thus causing the “rate high” error flag. To investigate this possibility, aqueous solutions with glycolic acid concentrations of 0–26.3 mmol/L were assayed for lactate on the Beckman LX 20 and on the Vitros 950 (Johnson & Johnson).

When measured by LX 20, glycolate concentrations up to 11.8 mmol/L produced an apparent lactate value up to 0.4 mmol/L, whereas glycolate concentrations ≥ 13.2 mmol/L produced a suppressed lactate result and a “rate high” flag (Table 1). Specimens with a “rate high” flag indeed displayed reaction rates > 10 mA/min. Fig. 1 depicts the reaction course for lactate, glycolic acid, and combined lactate/glycolic acid. The equilibrium state for lactate after 100 s and the nonequilibrium reaction rate for

<table>
<thead>
<tr>
<th>Glycolic acid</th>
<th>Apparent lactate, mmol/L</th>
<th>Net absorbance, a mA</th>
<th>Absorbance rate, mA/min</th>
</tr>
</thead>
<tbody>
<tr>
<td>mmol/L</td>
<td>mg/dL</td>
<td></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>0</td>
<td>0</td>
<td>6.0</td>
</tr>
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<td>3.3</td>
<td>25</td>
<td>0.1</td>
<td>7.9</td>
</tr>
<tr>
<td>6.6</td>
<td>50</td>
<td>0.2</td>
<td>16.0</td>
</tr>
<tr>
<td>9.9</td>
<td>75</td>
<td>0.3</td>
<td>24.0</td>
</tr>
<tr>
<td>10.5</td>
<td>80</td>
<td>0.4</td>
<td>27.6</td>
</tr>
<tr>
<td>11.8</td>
<td>90</td>
<td>0.4</td>
<td>31.2</td>
</tr>
<tr>
<td>13.2</td>
<td>100</td>
<td>Supp; b rate high</td>
<td>32.7</td>
</tr>
<tr>
<td>19.7</td>
<td>150</td>
<td>Supp; rate high</td>
<td>50.3</td>
</tr>
<tr>
<td>26.3</td>
<td>200</td>
<td>Supp; rate high</td>
<td>66.7</td>
</tr>
</tbody>
</table>

a Final absorbance measurement minus reagent blank absorbance just prior to sample addition.

b Supp, results suppressed.
glycolic acid are clearly apparent. The vertical lines mark the time period during which the reactions rate measurements were made.

When measured by the Vitros lactate method, all specimens with glycolic acid concentrations of 0–26.3 mmol/L registered zero lactate values. The Vitros lactate method is also based on a lactate oxidase/peroxidase coupled reaction. However, reaction conditions are obviously sufficiently different to avoid glycolic acid interference.

Values of zero were obtained when aqueous solutions with ethylene glycol concentrations up to 80.7 mmol/L (500 mg/dL) were assayed for lactate by the Beckman LX 20 method.

In ethylene glycol poisoning, the concentration of glycolic acid in serum correlates more closely with clinical severity than does that for ethylene glycol (3). For this reason, our laboratory developed gas chromatography-flame ionization (4) and gas chromatography-mass spectrometry (5) methods for the simultaneous determination of ethylene glycol and glycolic acid in serum. A rapid, nonchromatographic measure of glycolic acid would be of value for those laboratories without gas chromatographic instruments. We thus explored the possibility of using the Beckman lactate oxidase-based reaction for a kinetic measure of glycolic acid concentration. Whereas the reaction rate for glycolic acid is a reasonably linear function of its concentration between 3.3 and 26.3 mmol/L (Table 1 and Fig. 1), it becomes curvilinear between 26.3 and 52.6 mmol/L (data not shown). Moreover, the slope of the reaction rate vs glycolic acid concentration response curve is inversely related to the lactate concentration. Thus, application of this kinetic method as a measure of glycolic acid concentration in known cases of ethylene glycol ingestion cannot be recommended. An enzymatic assay for glycolic acid, based on a glycolate oxidase/peroxidase coupled reaction, has been reported (6). Lactate interferes; therefore, the glycolic acid response must be corrected for the contribution of endogenous lactate.

We measured serum ethylene glycol and glycolic acid concentrations for a series of 35 cases of ethylene glycol ingestion. Initial values for ethylene glycol and glycolic acid were 0.97–130.6 mmol/L (6–810 mg/dL) and 0–38 mmol/L, respectively. As reported previously (3,5), ethylene glycol concentrations correlate poorly with those for glycolic acid in ethylene glycol intoxication. For 16 patients, the initial glycolic acid concentration was >13 mmol/L [with corresponding ethylene glycol concentrations of 0.97–121.9 mmol/L (6–756 mg/dL)] and, based on the data in Table 1, would have produced a “rate high” flag if their sera were also measured for lactate on the Beckman LX 20. Thus, laboratories using the Beckman Synchron lactate method should be alert to the possibility of glycolic acid as the cause of a “rate high” error flag. This information could be an important clue to the possibility of ethylene glycol ingestion as the cause of an otherwise unknown high anion gap metabolic acidosis. Indeed, this scenario occurred in our institution. These laboratories should also be aware of the modest overestimation of lactate caused by glycolic acid when present at concentrations <13 mmol/L.

References