Food & Beverage Series

CHOLINE DETERMINATION

Application Note 203LS
YSI Life Sciences
INTRODUCTION
Choline concentrations in complex matrices can be measured directly and quickly using the YSI 2900 Series Biochemistry Analyzer. YSI’s unique enzyme technology provides for specific choline measurement. Measurements are virtually unaffected by color, turbidity, density, pH, or the presence of reducing substances.

When a sample is injected into the sample chamber, the choline diffuses into the membrane containing choline oxidase. The choline is immediately oxidized to hydrogen peroxide and betaine. The hydrogen peroxide is detected amperometrically at the platinum electrode surface. The current flow at the electrode is directly proportional to the hydrogen peroxide concentration, and hence to choline concentration.

I. MATERIALS & SETUP

A. YSI 2900 Series Biochemistry Analyzer - equipped with a 2771 Choline Membrane and 2357 Buffer.

B. Choline standards (175 mg/L, 450 mg/L).

C. Connect the 2900 Series instrument to a suitable power source.


E. Volumetric glassware (Class A recommended).

F. The following instrument setup is recommended. Sample size: 25 μL.

<table>
<thead>
<tr>
<th>Probe A Parameters</th>
<th>Probe B Parameters</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chemistry</td>
<td>B Chemistry</td>
</tr>
<tr>
<td>Unit</td>
<td>None</td>
</tr>
<tr>
<td>Calibrator</td>
<td>175 mg/L</td>
</tr>
<tr>
<td>End Point</td>
<td>30 Sec</td>
</tr>
</tbody>
</table>

II. METHOD

A. Total choline concentration should not exceed 450 mg/L, as determined on Part D below; otherwise the sample will require further dilution. Use volumetric glassware for all dilutions. Dilute with either water or 2357 buffer.

B. Calibrate the 2900 series instrument with a 175 mg/L Calibration Standard.

C. Check the linearity of the membrane at least once a day by injection of a choline linearity check solution (450 mg/L). Refer to the User’s Manual (Section 5) for specifications.

D. Assay the sample by aspiration into the 2900 series instrument. The linear range of the system is 5 to 450 mg/L choline. If the value reported exceeds this, further dilution is required.

E. Calibrate frequently as described in the User’s Manual.

III. CALCULATIONS

To calculate % choline, multiply the reported value by the appropriate dilution factor.

Example: 5.0 grams of pet food and 100 mL of water were mixed in a blender for 5 minutes. The supernatant was analyzed for choline. The value reported was 77.0 mg/L choline.

$$\% \text{ Choline: } 77.0 \text{ mg/L} \times 0.100 \frac{\text{L}}{5000 \text{ mg}} = 0.0015 \text{ g choline/g pet food} = 0.15\% \text{ (w/w)}$$

continued
Example: Infant formula was aspirated into the 2900 Series (no dilution). The choline content was as follows:

<table>
<thead>
<tr>
<th>Sample</th>
<th>mg/L Choline</th>
</tr>
</thead>
<tbody>
<tr>
<td>Infant Formula</td>
<td>A232</td>
</tr>
<tr>
<td>Infant Formula</td>
<td>B138</td>
</tr>
<tr>
<td>Medical Nutritional Formula A</td>
<td>398</td>
</tr>
<tr>
<td>Medical Nutritional Formula B</td>
<td>386</td>
</tr>
</tbody>
</table>

**ORDERING INFORMATION**

YSI Part Numbers:
- 2900  Biochemistry Analyzer
- 2771  Choline Membrane Kit
- 2772  Choline Standard Solution (175 mg/L)
- 2773  Choline Standard Solution (450 mg/L)
- 2357  Buffer Kit
- 2363  Potassium Ferrocyanide Test Solution
- 2392  NaCl Solution (for membrane installation)

For further information, please contact:
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YSI Life Sciences develops and manufactures scientific instruments, sensors and systems that serve a variety of scientific and industrial markets worldwide. YSI has a long history in the life sciences and bioanalytical markets, most notably with our introduction of the world’s first commercial whole blood glucose analyzer in 1975. Today there are over 10,000 YSI instruments installed around the world, trusted in critical situations to provide the most accurate data in the shortest time.

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