L-Glutamate Determination

INTRODUCTION
L-Glutamate 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 L-glutamate 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 glutamate diffuses into the membrane containing glutamate oxidase. The glutamate is immediately oxidized to hydrogen peroxide, \( \alpha \)-ketoglutarate, and ammonia. 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 glutamate concentration.

I. MATERIALS & SETUP
A. YSI 2900 Series Biochemistry Analyzer - equipped with a 2754 Glutamate Membrane and 2357 Buffer.
B. L-Glutamate standards (5.00 mmol/L, 10.0 mmol/L).
C. Connect the 2900 Series instrument to a suitable power source.
D. Perform the instrument and membrane daily checks described in the Operations Manual.
E. Connect the 2900 Series instrument to a suitable power source.
F. Volumetric glassware (Class A recommended).
G. The following instrument setup is recommended. Sample size:25 \( \mu \)L

Probe A Parameters

<table>
<thead>
<tr>
<th>Chemistry</th>
<th>Glutamate</th>
</tr>
</thead>
<tbody>
<tr>
<td>Unit</td>
<td>mmol/L</td>
</tr>
<tr>
<td>Calibrator</td>
<td>5.00 mmol/L</td>
</tr>
<tr>
<td>End Point</td>
<td>30 Sec</td>
</tr>
</tbody>
</table>

Autocal Parameters

<table>
<thead>
<tr>
<th>Temperature</th>
<th>1°C</th>
</tr>
</thead>
<tbody>
<tr>
<td>Time</td>
<td>30 Min</td>
</tr>
<tr>
<td>Sample</td>
<td>2 Sam</td>
</tr>
<tr>
<td>Cal Shift</td>
<td>2%</td>
</tr>
</tbody>
</table>

II. METHOD
A. Total glutamate concentration should not exceed 10.0 mmol/L, as determined in Part D below; otherwise the sample will require further dilution. Use volumetric glassware for all dilutions.
B. Calibrate the 2900 Series instrument with a 5.00 mmol/L L-Glutamate Calibration Standard.
C. Check the linearity of the membrane at least once a day by injection of a glutamate linearity check solution (10.0 mmol/L). Refer to the Operators Manual for specifications.
D. Assay the sample by aspiration into the 2900 Series. The linear range of the system is 0.1 to 10.0 mmol/L glutamate. If the value reported exceeds this, further dilution is required.
E. Calibrate frequently as described in the Operations Manual.

III. CALCULATIONS
To calculate % glutamate, multiply the reported value by the appropriate dilution factor.

For the examples, glutamate concentrations are expressed as monosodium glutamate. The molecular weight of monosodium glutamate (MSG) is 187.13 g/mole or 0.18713 g/mmol.

Example: The contents of a can of soup were blended in a blender on medium speed for about 3 minutes. 10.09 g of blended soup was diluted to 100 mL in a Class A volumetric flask with water. When assayed, the value reported was 1.42 mmol/L glutamate. continued
Example: 2.50 grams of a dry powder seasoning mix was diluted to 100 mL in a Class A volumetric flask with water. The mixture was stirred for 5 minutes. When assayed, the value reported was 6.85 mmol/L glutamate.

\[
\begin{array}{c|c}
\text{% Glutamate:} \\
1.42 \text{ mmol/L} \times \\
0.100L/10.09g \times \\
0.18713g/mmol MSG & = 0.0027 \text{ g MSG/g soup} \\
& = 0.27\% \text{ (w/w)}
\end{array}
\]

\[
\begin{array}{c|c}
6.85 \text{ mmol/L} \times \\
0.100L/2.50g \times \\
0.18713g/mmol MSG & = 0.0512 \text{ g MSG/g seasoning mix} \\
& = 5.12\% \text{ (w/w)}
\end{array}
\]

**ORDERING INFORMATION**

YSI Part Numbers:
- 2900  Biochemistry Analyzer
- 2754  Glutamate Oxidase Membrane Kit
- 2755  L-Glutamate Standard Solution (5.00 mmol/L)
- 2756  L-Glutamate Standard Solution (10.0 mmol/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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