Intended Use
The Glycmark test provides quantitative measurement of 1,5-anhydroglucitol (1,5-AG) in serum or plasma. The test is for professional use, and is indicated for in vitro diagnostic use only. The test should not be used for therapeutic decision making.

General Information

Materials Required But Not Supplied
- Glycmark Calibration Control (06-320)
- Glycmark Controls (RO1-038.32)
- Saline rinse blank (50 µL NaCl in deionized water)
- Automated chemistry analyzer: Roche/Hitachi 717 (Roche Diagnostics Corporation, Indianapolis, IN), or other appropriate open systems

Procedure
The assay is based on a kinetic determination principle. All reagents are used without dilution or other preparation. The reaction is performed at 37°C on the Hitachi 717 automated analyzer. At time zero, 4 µL of standard, control, reagent blank, or sample is added to 120 µL of Reagent 1. Following completion of the kinetic change in absorbance at 700 nm, the containers should be discarded in accordance with the rules of the facility, and in accordance with local, State, and Federal regulations.

1. Analytical Method/Materials point (point end value) (2005/05/20)
2. Wavelength (sub/main) (700/546)
3. Sample volume (standard) (4.0)
4. Sample volume (decrease) (2.0)
5. Sample volume (increase) (0.0)
6. Dilute (water/diluent)
7. Reagent volume (R1) (12.0mL/10.0mL)
8. Reagent volume (R2) (30.0mL/10.0mL)
9. Reagent volume (R3) (15.0mL/10.0mL)
10. Reagent volume (R4) (25.0mL/20.0mL)
11. Time point (2 point end) (2 point end changeover)
12. Auto-calibration
13. Calibration point (linear)
14. Weight factor (0)
15. Span point (2)
16. Purity value

For the quantitative measurement of 1,5-anhydroglucitol (1,5-AG) in serum or plasma.

For in vitro diagnostic use.
Results

Results are expressed in μg/mL, 1,5-AG.

Specimen Collection and Storage
Gco3M test provides equivalent results with serum or plasma samples. Studies have shown that serum samples are stable at 2-8°C for up to one week. For longer storage, freezing of the serum sample is recommended. Absence of frozen samples may be thawed and re-frozen for up to three cycles.

Kit Storage and Stability
Gco3M test is stable at refrigerated temperature (2-8°C) until the expiration date noted on the outer box. Once the refrigerated box have been opened, they are stable for 30 days at 2-8°C.

Limitations
- Gco3M test is to be used with serum or EDDS plasma; performance in other matrices has not been evaluated.
- Positively primary urinary glucose levels, or uric aciduria after administration, may result in a false 1,5-AG value. Low values have also been observed in pregnancy, terminal stage renal failure, dialysis patients, advanced cirrhosis, and prolonged incapability of oral ingestion of food. Abnormal values have also been noted in individuals with abnormally glomerular filtration rates. 5,6
- For some patients with severe hyperglycemia, the internal post of 1,5-AG may tend to remain depleted as a result of persistent hyperglycemia. In these cases, measurements of 1,5-AG may be less indicative of initial recovery following initiation of antidiabetic therapy.
- 1,5-AG values may be increased when some Chinese medicines, such as Polypsis delavayi and Sargaem variegatus, are administered. Urine values may also be increased during intravenous hyperalimentation. 1,5-AG values may be lower in patients undergoing therapy with high protein diet 10 mg/dL, 20 mg/dL.
- 1,5-AG blood levels are falsely lowered by the diabetes drug INVOKANA® which prevents reabsorption 1,5-AG in the kidneys. INVOKANA® is a trademark of Janssen Pharmaceutical, Inc.
- Some alpha-glucosidase inhibitors, such as Acarbose, may potentially reduce 1,5-AG levels due to interference with intestinal absorption of 1,5-AG.
- As with all diagnostic tests, Gco3M test results should be interpreted along with clinical findings and results from other diagnostic methods.

Quality Controls
Good laboratory practice includes the assigning of controls at regular intervals. It is recommended to use the two-level 1,5-AG Controls (Low” and “High”), and this control set is available from Glycocard, Inc. Users should also follow applicable federal, state, and local requirements for quality control testing.

Performance Characteristics
The following performance parameters were established on the Roche Hitachi 917 analyzer:

- Analytical sensitivity
  - Determined to be 0.2 μg/mL, and this is defined as the mean 1,5-AG concentration plus one standard deviation.
- Expected Values
  - A study was done with a presumptively normal population in order to determine Gco3M reference ranges. The study population was 306 subjects, of which 168 were males and 138 were females, ages 18 to 39, and 30 males and 30 females of age 40 or greater, for a total of 224 individuals. Ethnic backgrounds included African Americans, Caucasians, Asians, and Hispanics. The data did not demonstrate differences in ages, but there were gender differences. The following table provides the male and female ranges, based on the reference population.

<table>
<thead>
<tr>
<th>Gco3M Reference Ranges</th>
<th>Mean (μg/mL) 1,5-AG</th>
<th>Reference interval (μg/mL) 1,5-AG</th>
</tr>
</thead>
<tbody>
<tr>
<td>Males</td>
<td>22.5 (5.8)</td>
<td>10.7 to 32.0</td>
</tr>
<tr>
<td>Females</td>
<td>17.7 (3.2)</td>
<td>6.8 to 29.3</td>
</tr>
</tbody>
</table>

Each laboratory should establish their own reference ranges.

Linearity
Linearity was evaluated in a series of experiments using spiked samples. The concentrations of 1,5-AG in the samples ranged from 0 µg/mL to at least 50 µg/mL. The results were also unaffected by the following substances at their noted concentrations: triglycerides up to 1153 mg/dL, and bilirubin up to 40 mg/dL. Gco3M results were also unaffected by ascorbic acid- 25 mg/dL, uric acid- 20 mg/dL; glucose- 1000 mg/dL; maltose- 500 mg/dL; ascorbic acid- 25 mg/dL; uric acid- 20 mg/dL; L-arginine- 10 mg/dL; L-lysine- 10 mg/dL; L-tryptophan- 20 mg/dL; L-tyrosine- 5 mg/dL; L-phenylalanine- 10 mg/dL; L-leucine- 10 mg/dL; L-lysine- 20 mg/dL. The following antigens were not interfaced with the Gco3M test, at their noted concentrations: EDTA- 100 units/mL; sodium fluoride- 0.2%; sodium citrate- 2.0%.

Precision
Precision studies were performed to evaluate both within-assay and between-assay precision. The summarized results are described below.

Within-Assay Variance (20 replicates of the Gco3M controls were assayed according to standard procedure. Mean, standard deviation, and percent coefficient of variation (CV)) were calculated for each control solution. The within-assay precision ranged from 1.3 to 3.8%.

Clinical Use
A prospective, longitudinal study was performed with 77 patients with diabetes (both type 1 and type 2). The patients exhibited suboptimal glycolytic control (A1C level greater than or equal to 7%) at study entry, and these patients were monitored for eight weeks following initiation or modification of antihyperglycemic treatment used for the monitoring of glycemic control. There was an overall improvement of less than an association between glucose and the other three markers, as glucose can only provide a "snapshot" of glucose monitoring at the time of sampling.

Assessment of the impact of glycemic control on determination of A1C was determined. "Concordance" was defined as either increases in 1,5-AG values with corresponding increases in A1C values, or, conversely decreases in 1,5-AG values with corresponding increases in A1C values. 80.6% of the patients (80/97) displayed concordance in changes in 1,5-AG and A1C standard values with time.

References


8. F-19854 FU-100371

9. October 2013

Japan

Imported by Toyota Tissue America, Inc.
Distributor: Toyota Tissue America, Inc.
BDS., Tissue America, Inc.

Tissue America, Inc.

F-19854 FU-100371

For a complete listing of references, please visit the manufacturer's website.
Intended Use
The GlycoMark Controls are to be used with the GlycoMark test.

The GlycoMark test provides quantitative measurement of 1,5-anhydroglucitol (1,5-AG) in serum or plasma. The test is for professional use, and is indicated for the intermediate term monitoring of glycemic control in people with diabetes. Please see the GlycoMark Kit Reagents package insert for full product description.

Warnings and Precautions
1. Directions in the GlycoMark Kit Reagents package insert must be followed for optimal results.
2. The GlycoMark Controls contain 1.0 mg/mL sodium azide as a preservative. Avoid contact by mouth, with the skin, or any mucous membranes. In the case of contact with the standard, immediately wash the affected areas with large amounts of water. Sodium azide reacts with lead and copper pipes to generate an explosive metal azide compound. When disposing leftover reagents down a sink, large amounts of water should be used to flush the pipes.
3. The controls should not be used if flocculation or discoloration occur.
4. Do not use the controls past their expiration dating.
5. The controls are to be stored at refrigerated temperatures (2-8°C). Open vials should also be stored at refrigerated temperatures (2-8°C).
6. Do not dilute the controls.
7. After GlycoMark analysis, the containers should be discarded in accordance with rules of the facility, and in accordance with local, State, and Federal regulations.

Package Components (Materials Supplied)
Low Control – 3 vials, 2 mL each
• 1,5-AG: ~4.0 to 5.5 µg/mL
• SeraSub®
• Sodium azide: 1.0 mg/mL

High Control – 3 vials, 2 mL each
• 1,5-AG: ~13.0 to 16.0 µg/mL
• SeraSub®
• Sodium azide: 1.0 mg/mL

The control ranges are indicated for every lot of controls – see bottle label. The controls are prepared in a surrogate serum matrix to eliminate the risk of using biohazardous materials.

Materials Required But Not Supplied
• GlycoMark Kit Reagents – REF (Small) NK-8300
• GlycoMark Calibration Standard – REF NK-8320
• Saline reagent blank
• Automated chemistry analyzer, Roche/Hitachi 917 (Roche Diagnostics Corporation, Indianapolis, IN), or other appropriate open systems

Procedure
Bring the controls to room temperature and mix gently to avoid foaming. Controls should be assayed in the same manner as clinical samples. See the GlycoMark Kit Reagents package insert for procedural instructions.

Controls Storage and Stability
The GlycoMark Controls are stable at refrigerated temperatures (2-8°C) until the expiration date noted on the outer box. Opened vials are stable for one month if stored at refrigerated temperatures (2-8°C).

Limitations
• Instrument settings and parameters may change from instrument to instrument. Please contact GlycoMark, Inc. for assistance with a particular instrument. Incorrect results may be obtained if incorrect sample volumes or reagent volumes are used.
• As with all diagnostic tests, GlycoMark results should be interpreted along with clinical findings and results from other diagnostic methods.

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**Intended Use**

The GlycoMARK Calibration Standard is to be used with the GlycoMARK Test.

The GlycoMARK Test provides quantitative measurement of 1,5-anhydroglucitol (1,5-AG) in serum or plasma. The test is for professional use, and is indicated for the intermediate term monitoring of glycemic control in people with diabetes. Please see the GlycoMARK Kit Reagents package insert for full product description.

**Warnings and Precautions**

1. Directions in the GlycoMARK Kit Reagents package insert must be followed for optimal results.
2. The GlycoMARK Calibration Standard contains 1.0 mg/mL sodium azide as a preservative. Avoid contact by mouth, with the skin, or any mucous membranes. In the case of contact with the standard, immediately wash the affected areas with large amounts of water. Sodium azide reacts with lead and copper pipes to generate an explosive metal azide compound. When disposing leftover reagents down a sink, large amounts of water should be used to flush the pipes.
3. The standard should not be used if flocculation occurs.
4. Do not use the standard past its expiration dating.
5. The standard is to be stored at refrigerated temperatures (2-8°C). Open vials should also be stored at refrigerated temperatures (2-8°C).
6. Do not dilute the standard.
7. After GlycoMARK analysis, the containers should be discarded in accordance with rules of the facility, and in accordance with local, State, and Federal regulations.

**Package Components (Materials Supplied)**

Calibration Standard – 3 vials, 5 mL each
- 1,5-AG: 50 µg/mL
- Sodium azide: 1.0 mg/mL
- Water

**Materials Required But Not Supplied**

- GlycoMARK Kit Reagents – REF (Small) NK-8300
- GlycoMARK Controls – REF NK-8330
- Saline reagent blank (0.9% physiological saline)
- Automated chemistry analyzer, Roche/Hitachi 917 (Roche Diagnostics Corporation, Indianapolis, IN), or other appropriate open systems

**Procedure**

Refer to the GlycoMARK Kit Reagents package insert for details.

**Calibration Standard Storage and Stability**

The GlycoMARK Calibration Standard is stable at refrigerated temperatures (2-8°C) until the expiration date noted on the outer box. Opened vials are stable for one month if stored at refrigerated temperatures (2-8°C).

**Limitations**

- Instrument settings and parameters may change from instrument to instrument. Please contact GlycoMark, Inc. for assistance with a particular instrument. Incorrect results may be obtained if incorrect sample volumes or reagent volumes are used.
- As with all diagnostic tests, GlycoMARK results should be interpreted along with clinical findings and results from other diagnostic methods.

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