HbA1c NEST FS
The Future Of Diabetes Monitoring

The New Routine Reference Method
For Determination Of HbA1c.
The worldwide, rapid rise in diabetes is a challenge for treatment as well as for diagnosis and control. What we need are new diagnostic technologies providing precise, dependable and quick results. With HbA1c net FS, DiaSys has made available an enzymatic test whose high specificity and precision sets new standards for accuracy.

Innovative and easy:
a modern method for more efficiency
The new HbA1c net FS test is based on an enzymatic method that provides a number of advantages in addition to high-quality results. It simplifies operations and increases throughput because it requires less steps than other methods.

• With the enzymatic HbA1c test, cuvettes do not get contaminated by latex. Due to this, in comparison to latex-based tests, less cleaning effort is required, which saves costs and time.
• Calibration stability of four weeks ensures usage up to the last drop of reagent.
• The combination of liquid-stable, ready-to-use 2-component reagent, 2-level calibrator set and 2-level controls stands for a comfortable and easy handling.
• With on-board hemolysis of the HbA1c test the workflow will be optimized.

High specificity for highly accurate results
The HbA1c net FS mainly owes its great accuracy to the test’s high specificity because it determines fructosyl dipeptides at the N-terminus of the hemoglobin β-chain. The specificity is comparable to the IFCC reference measurement procedure for HbA1c determination which measures the N-terminal hexapeptide of the hemoglobin β-chain.

An extensive variety of hemoglobin variants (HbS, HbC, HbE, HbF etc.), acetylated hemoglobin, carbamylated hemoglobin and other interfering substances such as ascorbate, bilirubin, triglycerides and urea do not show significant interference.

Clinical significance of HbA1c assessment
Hemoglobin A1c (HbA1c) is metabolically produced by the reaction of glucose with the N-terminal valine residue of each β-chain of hemoglobin and the subsequent formation of a stable ketoamine. It is defined as “hemoglobin beta-chain (blood)-N-(1-deoxyfructos-1-y1) hemoglobin beta-chain, substance fraction”.[1]

The HbA1c test correlates to the average blood glucose level for approximately the last three months and is used for long-term glycemic control in patients with diabetes mellitus. Clinical studies have shown that the lowering of the HbA1c level can help to prevent or delay the incidence of diabetes complications.
The Crucial Advantage: Unsurpassed Accuracy.

Common HbA1c assays based on HPLC or immunoturbidimetry may be affected by hemoglobinopathies which are frequent in certain patient groups. The extraordinary specificity of DiaSys HbA1c net FS ensures reliable results for all which makes it the new standard in HbA1c determination.

Specific and precise: the principle behind the enzymatic determination

The test is based on a colorimetric, enzymatic method. The concentrations of HbA1c and total hemoglobin are determined separately. Calculation of the HbA1c ratio from total hemoglobin is done by the instrument automatically.

Consistently precise results: the assay procedure

**Pretreatment and hemoglobin measurement**

Whole blood samples are lysed with hemolyzing solution. Hemoglobin is released from the erythrocytes. The absorbance of hemoglobin is measured at 570 nm after the addition of reagent R1 and is proportional to the total hemoglobin concentration in the sample.

**HbA1c measurement**

After the addition of R2, fructosylated dipeptides are released by protease, including the N-terminal part of the hemoglobin β-chain. Hydrogen peroxide (H2O2) is released after the oxidative cleaving of the fructosylated dipeptides by FPOX (Fructosyl peptide oxidase). The generated H2O2 is determined colorimetrically by a reaction with a suitable chromogen and the enzyme peroxidase at 660 nm. The delta absorbance of the determined color is proportional to the HbA1c concentration.

Method comparison against HPLC assay

No significant interference by Hb variants with HbA1c net FS

<table>
<thead>
<tr>
<th>HbA1c</th>
<th>HbAD</th>
<th>HbAE</th>
<th>HbAS</th>
<th>HbCC</th>
<th>HbEE</th>
<th>HbSS</th>
<th>Elevated HbF</th>
<th>Acetylated Hb</th>
</tr>
</thead>
<tbody>
<tr>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
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</tr>
</tbody>
</table>

More Hb variants were tested and showed no significant interference.

DiaSys HbA1c net FS shows excellent precision over the entire measuring range.

<table>
<thead>
<tr>
<th>Intra-assay N = 20</th>
<th>Mean (mmol/mol)</th>
<th>SD (mmol/mol)</th>
<th>CV (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sample 1</td>
<td>37.3</td>
<td>0.218</td>
<td>0.58</td>
</tr>
<tr>
<td>Sample 2</td>
<td>59.5</td>
<td>0.470</td>
<td>0.79</td>
</tr>
<tr>
<td>Sample 3</td>
<td>115</td>
<td>0.375</td>
<td>0.33</td>
</tr>
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</tr>
</thead>
<tbody>
<tr>
<td>Sample 1</td>
<td>26.0</td>
<td>0.583</td>
<td>2.24</td>
</tr>
<tr>
<td>Sample 2</td>
<td>39.3</td>
<td>0.745</td>
<td>1.90</td>
</tr>
<tr>
<td>Sample 3</td>
<td>75.7</td>
<td>1.64</td>
<td>2.17</td>
</tr>
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</table>

Imprecision

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</thead>
<tbody>
<tr>
<td></td>
<td>Passing/Bablok</td>
<td>y = 1.015 X - 0.213 %</td>
<td>r = 0.9977</td>
</tr>
</tbody>
</table>

Test characteristics

- Liquid-stable, ready-to-use 2-component reagent, 2-level calibrator set and 2-level controls for easy handling
- On-board and calibration stability of 4 weeks
- Wide measuring range from 15 to 150 mmol/mol IFCC (3 – 16 % DCCT/NGSP) within a hemoglobin concentration range from 3.5 up to 29 g/dL
- Excellent intra and inter-assay precision
- No contamination of cuvettes by latex
- Standardized against the IFCC1 reference method and also traceable according to the DCCT/NGSP2 network

1 International Federation of Clinical Chemistry
2 Diabetes Control and Complications Trial/National Glycohemoglobin Standardization Program
### HbA1c @*@ F5

<table>
<thead>
<tr>
<th>Cat. No.</th>
<th>Kit size</th>
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<tr>
<td>13348 99 10 930</td>
<td>R1 3 x 18 mL + R2 3 x 6 mL</td>
</tr>
</tbody>
</table>

#### Hemolyzing Solution
14590 99 10 113
- 1 x 500 mL
- HbA1c net Hemolyzing Solution

#### Calibrator set – 2 different levels
13340 99 10 043
- 1 x 0.25 mL
- Level 1
- TruCal HbA1c net

#### Controls – 2 different levels
- **Level 1**
  - 5 9790 99 10 074
  - 4 x 0.25 mL
  - TruLab HbA1c liquid Level 1
  - 5 9800 99 10 060
  - 1 x 0.25 mL

- **Level 2**
  - 5 9790 99 10 074
  - 4 x 0.25 mL
  - TruLab HbA1c liquid Level 2
  - 5 9800 99 10 060
  - 1 x 0.25 mL