INTRODUCTION

Galactin-3 is one of several known beta-galactoside-binding lectins involved in the regulatory function of inflammation, immune response, and cell-cell interaction (1). Studies suggest that galactin-3 participates in a variety of functions, including cell-cell interaction, immune response, and cell-cell contact (2). Therefore, galectin-3 is a potential biomarker for disease state (3). The ELISA is the preferred method for measuring galectin-3 concentrations in serum (4). Serum samples are collected after fasting for 12 hours, centrifuged, and the supernatant is stored at -80 °C. The test kit manufacturer’s recommended storage temperature is -20 °C for 22 days (5). The analyte stability claim for galectin-3 by the test kit manufacturer was 22 days for both ambient and refrigerated temperatures. However, our initial studies showed these time intervals were too short. Therefore, galectin-3 concentrations of 17.9–25.9 ng/mL are to be interpreted with caution.

RESULTS

The results in Figure 1 demonstrate acceptable linearity throughout the ELISA’s clinical analytical measurement range (AMR) of 1.4–94.8 ng/mL with slope = 0.981 (95% confidence interval CI = 0.940 – 1.023), intercept = -1.028 (95% CI = -3.140 – 1.354) and r² = 0.992. (Linear regression, n = 11).<ref>Figure 1. Galectin-3 ELISA Linearity.</ref>

As shown in Figure 2, the lot-to-lot variability was acceptable producing a slope = 1.1 (95% CI = 0.940 – 1.203), intercept = -1.905 (95% CI = -2.380 – -1.430) and r² = 0.991. (Linear regression, n = 11).<ref>Figure 2. Galectin-3 ELISA Linearity by Batch.</ref>

Table 1. Calibration Data Points

<table>
<thead>
<tr>
<th>Concentration (ng/mL)</th>
<th>Absorbance (OD)</th>
<th>Slope (95% CI)</th>
<th>Intercept (95% CI)</th>
<th>R²</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.4</td>
<td>0.145</td>
<td>0.981 (0.940 – 1.023)</td>
<td>-1.028 (95% CI = -3.140 – 1.354)</td>
<td>0.992</td>
</tr>
<tr>
<td>3.8</td>
<td>0.284</td>
<td>0.981 (0.940 – 1.023)</td>
<td>-1.028 (95% CI = -3.140 – 1.354)</td>
<td>0.992</td>
</tr>
<tr>
<td>7.8</td>
<td>0.423</td>
<td>0.981 (0.940 – 1.023)</td>
<td>-1.028 (95% CI = -3.140 – 1.354)</td>
<td>0.992</td>
</tr>
<tr>
<td>13.9</td>
<td>0.562</td>
<td>0.981 (0.940 – 1.023)</td>
<td>-1.028 (95% CI = -3.140 – 1.354)</td>
<td>0.992</td>
</tr>
<tr>
<td>23.9</td>
<td>0.701</td>
<td>0.981 (0.940 – 1.023)</td>
<td>-1.028 (95% CI = -3.140 – 1.354)</td>
<td>0.992</td>
</tr>
<tr>
<td>52.3</td>
<td>1.110</td>
<td>0.981 (0.940 – 1.023)</td>
<td>-1.028 (95% CI = -3.140 – 1.354)</td>
<td>0.992</td>
</tr>
</tbody>
</table>

The CVs were 7.8, 3.6 and 6.0% at the same respective concentrations. Accuracy was evaluated using 21 serum samples with known concentrations. The within-run accuracy for all galectin-3 concentrations was within ± 10% of the true value (n = 20 per concentration). The upper limit of detection was calculated to be 1.1 ng/mL. The calculated limit of detection was 1.1 ng/mL requiring the minimum absorbance for detection to be 0.0448. A minimum absorbance of 0.0448 corresponded to the measured absorbance of 0.0968 at a concentration of 1.4 ng/mL (95% confidence interval CI = 0.940 – 1.023), intercept = -1.028 (95% CI = -3.140 – 1.354) and r² = 0.992. (Linear regression, n = 11).<ref>Figure 3. Galectin-3 ELISA Linearity.</ref>

CONCLUSIONS

The BGM Galectin-3™ ELISA demonstrates acceptable linearity, accuracy, precision, analytical sensitivity and lot-to-lot variability for measuring galectin-3 protein in serum.

An upper reference interval limit of 26.2 ng/mL was verified in serum for the Galectin-3 ELISA. Analyte stability claims by the test kit manufacturer are overestimated compared to this study's results. Nevertheless, galectin-3 stability remains acceptable for clinical applications.

ACKNOWLEDGEMENTS

We would like to thank BG Medicine, Inc. for supplying the BG Galectin-3™ ELISA kits and instruments.

The ARUP Institute for Clinical and Experimental Pathology provided financial support.

REFERENCES