Introduction
Serum antinuclear antibodies (ANA) are the most used autoimmune markers in clinical practice. These markers include anti-dsDNA, anti-His, anti-Jo-1, anti-nRNP/Sm, anti-P0, anti-Scl-70, anti-Sm, anti-SSA, anti-SSB/La, anti-Nuc, anti-PCNA and anti-PM-Scl et al. ANAs are found in patients with a number of different autoimmune diseases, such as the Systemic Lupus Erythematosus (SLE), Rheumatoid Arthritis (RA), Sjogren's Syndrome (SS), Progressive systemic sclerosis (PSS), Glomerulonephritis (GN) and Autoimmune Hepatitis (AIH).

Objective
The objective of this study is to develop and evaluate the clinical performances of a innovative HOB 4G Autoimmune ANA Panel on the Automated Immunoassay BIOCLIA®-1200 Analyzer.

Methods and results
The BioCLIA® 1200 assay utilizes an enzyme-enhanced chemiluminescence immunoassay (Figure 2) for the quantification of autoantibodies. Streptavidin-coated magnetic nanoparticle (M), biotinylated autoantigen (R1), and serum samples are incubated at 37°C for 15 mins. After washing, the alkaline phosphatase-labeled mouse anti-human IgG (R2) is added and incubated for another 15 mins. After washing, the reacted reagents are measured with the AMPPD chemiluminescence substrate for quantitative determination of autoantibodies in serum.

Materials and methods
A total of 200 clinical samples were tested in the multiprofiling study. ANAs were found in patients with a number of different autoimmune diseases, such as the Systemic Lupus Erythematosus (SLE), Rheumatoid Arthritis (RA), Sjogren's Syndrome (SS), Progressive systemic sclerosis (PSS), Glomerulonephritis (GN) and Autoimmune Hepatitis (AIH).

Table 1. Within-run and between-run

<table>
<thead>
<tr>
<th>Antigen</th>
<th>Within-run CV (%)</th>
<th>Between-run CV (%)</th>
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<tbody>
<tr>
<td>nRNP/Sm</td>
<td>3.51 ± 0.47</td>
<td>4.83 ± 0.47</td>
</tr>
<tr>
<td>Sm</td>
<td>2.94 ± 0.24</td>
<td>4.43 ± 0.28</td>
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<tr>
<td>SSA</td>
<td>3.59 ± 3.34</td>
<td>5.37 ± 3.24</td>
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<tr>
<td>SCL-70</td>
<td>3.65 ± 3.35</td>
<td>5.30 ± 3.08</td>
</tr>
<tr>
<td>PM-Scl</td>
<td>3.15 ± 2.61</td>
<td>4.96 ± 4.46</td>
</tr>
<tr>
<td>Jo-1</td>
<td>3.59 ± 3.16</td>
<td>4.76 ± 4.29</td>
</tr>
<tr>
<td>PCNA</td>
<td>3.42 ± 3.36</td>
<td>5.19 ± 5.02</td>
</tr>
<tr>
<td>dScl1A</td>
<td>2.97 ± 2.90</td>
<td>4.60 ± 4.23</td>
</tr>
<tr>
<td>P0</td>
<td>3.62 ± 3.60</td>
<td>5.40 ± 4.61</td>
</tr>
<tr>
<td>Nuc</td>
<td>3.67 ± 3.78</td>
<td>5.14 ± 4.49</td>
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Linearity:

The dilution series of the calibrator at 400 RU/L was evaluated on triplicate testing of a dilutional linearity: was evaluated on triplicate testing of a dilutional linearity:

- nRNP/Sm: LOD (RU/ml) = 0.027, Slope: 3.51, Coefficient of Determination (R²): 0.9991
- Sm: LOD (RU/ml) = 0.025, Slope: 2.94, Coefficient of Determination (R²): 0.9993
- SSA: LOD (RU/ml) = 0.036, Slope: 3.59, Coefficient of Determination (R²): 0.9996
- SCL-70: LOD (RU/ml) = 0.036, Slope: 3.65, Coefficient of Determination (R²): 0.9996
- PM-Scl: LOD (RU/ml) = 0.036, Slope: 3.15, Coefficient of Determination (R²): 0.9996
- Jo-1: LOD (RU/ml) = 0.036, Slope: 3.59, Coefficient of Determination (R²): 0.9996
- PCNA: LOD (RU/ml) = 0.036, Slope: 3.42, Coefficient of Determination (R²): 0.9996
- dScl1A: LOD (RU/ml) = 0.036, Slope: 2.97, Coefficient of Determination (R²): 0.9996
- P0: LOD (RU/ml) = 0.036, Slope: 3.62, Coefficient of Determination (R²): 0.9996
- Nuc: LOD (RU/ml) = 0.036, Slope: 3.67, Coefficient of Determination (R²): 0.9996

Dilutional linearity:

Fitted regression equation:

- nRNP/Sm: y = 1.0099x + 0.4681, R² = 0.9991
- Sm: y = 1.0092x - 0.5244, R² = 0.9993
- SSA: y = 0.9845x - 2.3532, R² = 0.9968
- SCL-70: y = 0.1026x - 2.9842, R² = 0.9991
- PM-Scl: y = 0.9952x - 1.3453, R² = 0.9996
- Jo-1: y = 1.0674x + 1.588, R² = 0.9991
- PCNA: y = 1.0011x + 0.5494, R² = 0.9999
- dScl1A: y = 1.0116x - 1.3337, R² = 0.9986
- P0: y = 0.9911x - 1.1099, R² = 0.9994
- Nuc: y = 1.0049x + 1.7652, R² = 0.9992
- Observed: y = 0.9901x - 2.1922, R² = 0.9989

Conclusion:

The BioCLIA® 1200 chemiluminescent immunoassay system utilizes streptavidin-coated magnetic nanoparticle with lots of greater surface binding areas to improve sensitivity. The magnetic nanoparticles enable the separation and washing procedures; the bounded autoantibodies reacts with selected biotin-labeled autoimmune antigen and is detected by the anti human IgG conjugated alkaline phosphate that triggers the chemiluminescence substrate for quantitative determination of autoantibodies in serum.