HUMAN MxA
A SPECIFIC LABORATORY MARKER
OF VIRAL INFECTION

Human MxA Protein ELISA

- Differentiation between patients with viral and bacterial infection
- Monitoring of IFN-beta bioactivity in treatment of patients with multiple sclerosis
- Manufactured under the licence from Kyowa Medex Corporation, Ltd.

VIRAL INFECTION
MULTIPLE SCLEROSIS
Human MxA protein (Myxovirus resistance protein 1), the product of the MX1 gene, is a 76-kDa protein consisting of 662 amino acid residues and belonging to the dynamic superfamily of large GTPase.

MxA protein plays an important role in antiviral activity in cells against a wide variety of viruses, including influenza, parainfluenza, measles, coxsackie, hepatitis B virus, and Thogoto virus. The viruses are inhibited by MxA protein at an early stage in their life cycle, soon after host cell entry and before genome amplification. The mouse Mx1 protein (mouse analog of human MxA protein) accumulates in the cell nucleus where it associates with nuclear bodies and inhibits influenza and Thogoto viruses known to replicate in the nucleus. The human MxA protein accumulates in the cytoplasm and endoplasmic reticulum as well. The membrane compartment of endoplasmatic reticulum seems to provide an interaction platform that facilitates viral target recognition. MxA appears to detect viral infection by sensing and trapping nucleocapsid-like structures. As a consequence, the viral components become unavailable for the generation of new virus particles.

The expression of viral MxA protein is induced exclusively and in a dose-dependent manner by IFN-alpha and IFN-beta, but not by IFN-gamma, IL-1, TNF-alpha or other cytokines.

In clinical diagnostics, MxA protein may offer advantages as a marker for viral infection over the other induced proteins such as 2', 5'-oligoadenylate synthetase, because of its very low basal concentration and long half-life. Several clinical studies have reported on the possible use of MxA protein expression in peripheral blood mononuclear cells as a marker for distinguishing viral from bacterial disease, and as a reliable marker for type I IFN bioavailability during IFN treatment of chronic viral hepatitis and multiple sclerosis. Myxovirus resistance protein A (MxA) can be used as a marker of the bioactivity of interferon-beta (IFN-beta) therapy in patients with multiple sclerosis (MS). Two to forty per cent of IFN-beta-treated multiple sclerosis (MS) patients develop IFN-beta-neutralizing antibodies (NAb) with subsequent attenuation of MxA protein induction. MxA ELISA could be used for assessment of raised NAb.
**Intended use**
The RD194349200R Human MxA Protein ELISA is a sandwich enzyme immunoassay for the quantitative measurement of human Myxovirus resistance protein 1.
- The total assay time is less than 3 hours
- The kit measures MxA protein in human whole blood (cell lysate)
- Assay format is 96 wells
- Standard is recombinant protein based
- Components of the kit are provided ready-to-use, concentrated or lyophilized

**Clinical application**
- Viral infection
- Multiple sclerosis (MS)

**Test principle**
In the BioVendor Human MxA Protein ELISA, Standards and samples are incubated in microtitration wells pre-coated with monoclonal anti-human MxA protein antibody. After 60 minutes incubation followed by washing, biotin labelled monoclonal anti-human MxA protein antibody is added and incubated with the captured MxA protein for 60 minutes. After another washing, streptavidin-HRP conjugate is added. After 30 minutes incubation and the last washing step, the remaining conjugate is allowed to react with the substrate solution (TMB). The reaction is stopped by addition of acidic solution and absorbance of the resulting yellow product is measured. The absorbance is proportional to the concentration of MxA protein. A standard curve is constructed by plotting absorbance values against MxA protein concentrations of Standards and concentrations of unknown samples are determined using this standard curve.

**HUMAN MxA PROTEIN ELISA**
**CAT. NO.: RD194349200R**

<table>
<thead>
<tr>
<th>Assay format</th>
<th>Sandwich ELISA, Biotin-labelled antibody, 96 wells/kit</th>
</tr>
</thead>
<tbody>
<tr>
<td>Samples</td>
<td>Whole blood (cell lysate)</td>
</tr>
<tr>
<td>Standards</td>
<td>0.375 to 12 ng/ml</td>
</tr>
<tr>
<td>Limit of detection</td>
<td>0.03 ng/ml</td>
</tr>
</tbody>
</table>
**Precision**

Intra-assay (Within-Run) (n=8)

<table>
<thead>
<tr>
<th>Sample</th>
<th>Mean (ng/ml)</th>
<th>SD (ng/ml)</th>
<th>CV (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>38.54</td>
<td>0.7</td>
<td>1.9</td>
</tr>
<tr>
<td>2</td>
<td>11.56</td>
<td>0.9</td>
<td>7.4</td>
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</table>

Inter-assay (Run-to-Run) (n=8)

<table>
<thead>
<tr>
<th>Sample</th>
<th>Mean (ng/ml)</th>
<th>SD (ng/ml)</th>
<th>CV (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>37.07</td>
<td>1.6</td>
<td>4.3</td>
</tr>
<tr>
<td>2</td>
<td>10.46</td>
<td>1.0</td>
<td>9.6</td>
</tr>
</tbody>
</table>

**Spiking recovery**

Serum samples were spiked with different amounts of human MxA protein and assayed.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Observed (ng/ml)</th>
<th>Expected (ng/ml)</th>
<th>Recovery O/E (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>3.05</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>6.52</td>
<td>6.77</td>
<td>96.3</td>
</tr>
<tr>
<td></td>
<td>4.66</td>
<td>4.90</td>
<td>95.2</td>
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<tr>
<td></td>
<td>3.93</td>
<td>3.95</td>
<td>99.4</td>
</tr>
<tr>
<td>2</td>
<td>1.36</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>4.97</td>
<td>5.11</td>
<td>97.3</td>
</tr>
<tr>
<td></td>
<td>3.15</td>
<td>3.23</td>
<td>97.5</td>
</tr>
<tr>
<td></td>
<td>2.58</td>
<td>2.29</td>
<td>112.7</td>
</tr>
</tbody>
</table>

**Linearity**

Serum samples were serially diluted with Dilution Buffer and assayed.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Dilution</th>
<th>Observed (ng/ml)</th>
<th>Expected (ng/ml)</th>
<th>Recovery O/E (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>-</td>
<td>9.13</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>2x</td>
<td>4.50</td>
<td>4.57</td>
<td>98.6</td>
</tr>
<tr>
<td></td>
<td>4x</td>
<td>2.11</td>
<td>2.28</td>
<td>92.4</td>
</tr>
<tr>
<td>2</td>
<td>-</td>
<td>4.68</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>2x</td>
<td>2.36</td>
<td>2.34</td>
<td>100.9</td>
</tr>
<tr>
<td></td>
<td>4x</td>
<td>1.06</td>
<td>1.17</td>
<td>90.6</td>
</tr>
</tbody>
</table>

**Interference Study**

Interference by lipids, direct bilirubin, indirect bilirubin, hemoglobin and various anticoagulants was examined by the addition of the test metabolite to aliquots of whole blood.

**Summary of protocol**

- Reconstitute Master Standard and prepare set of Standards
- Dilute whole blood samples (10x)
- Add 100 µl Standards and samples
  - Incubate at RT for 1 hour/300 rpm
  - Wash plate 3 times
  - Add 100 µl Biotin Labelled Antibody
  - Incubate at RT for 1 hour/300 rpm
  - Wash plate 3 times
  - Add 100 µl Streptavidin-HRP Conjugate
  - Incubate at RT for 30 min/300 rpm
  - Wash plate 5 times
  - Add 100 µl Substrate Solution
  - Incubate at RT for 20 min
  - Add 100 µl Stop Solution
  - Read absorbance and calculate results
MxA protein levels in whole blood of healthy controls and patients with viral or bacterial infection:

- Patients with etiologically diagnosed viral infection (N = 17; respiratory syncytial virus: 10, influenza virus A/B: 3, adenovirus: 3, rotavirus: 1)
- Patients with clinically diagnosed viral infection (N = 14; upper respiratory infection: 10, gastroenteritis: 4)
- Patients with bacterial infection (N = 11)
- Healthy individuals (N = 18)

Horizontal bars and values indicate the median for each group.


Clinical Relevance

Related products

- RGP019R IP-10 (CXCL10) Human ELISA
- RD191006200R Procalcitonin Human ELISA
References

7. Simon et al., Interferon-regulated Mx gene are not responsive to IL-1, TNF and other cytokins. J. Virol., 65, 968-971 (1991)

References to this product


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