PRODUCT INFORMATION AND MANUAL

FlowCytomix
Human MIP-1α Simplex Kit

BMS82029FF

For research use only.
Not for diagnostic or therapeutic procedures.
96 Tests
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This Human MIP-1α Simplex Kit must be used in combination with FlowCytomix Basic Kit BMS8420FF. For test procedure, measurement and calculation of results please refer to FlowCytomix Basic Kit BMS8420FF manual.

1 REAGENTS PROVIDED

1 vial (175 µl) Fluorescent Beads (20x) coated with polyclonal antibody to MIP-1α, Bead Population A9
2 vials MIP-1α Standard (lyophilized): 200 ng/ml upon reconstitution
1 vial (350 µl) Biotin-Conjugate (20x) anti-MIP-1α polyclonal antibody

2 INTENDED USE

BMS82029FF is a bead based Analyte Detection System for quantitative detection of MIP-1α in cell culture supernatants, serum and plasma (EDTA, citrate) by Flow Cytometry. BMS82029FF is for research use only. Not for use in diagnostic or therapeutic procedures.

3 SUMMARY

Chemokines are cytokines that induce chemotaxis of inflammatory cells. They are able to induce leukocyte chemotaxis and adhesion to endothelial cells. Macrophage inflammatory protein 1 alpha (MIP-1alpha) and beta (MIP-1beta) belong to the family of cysteine-cysteine (cc) chemokines, RANTES being another prominent member thereof. The chemokines self-associate to form high molecular mass aggregates while the monomers are low mass polypeptides. Both MIP-1alpha and MIP-1beta are not only chemoattractants but also coactivators of macrophages acting in concert with IFN-γ as type 1 cytokines. MIP-1alpha and MIP-1beta are distinct but highly homologous chemokines produced by a variety of host cells in response to various external stimuli and share affinity for their receptor CCR5. The roles of MIP-1alpha and MIP-1beta have been elucidated in response to their affects on cellular and humoral immune response.
MIP-1alpha was shown to stimulate strong antigen specific responses, while MIP-1beta promotes antibody responses. Both macrophage inflammatory proteins are however strictly associated with type 1 immune response. Determination of the expression levels of the MIP-1s turned out to provide important information regarding numerous diseases such as multiple myeloma, allergic asthmatic disorders, acute experimental autoimmune encephalomyelitis, HIV infection, sarcoidosis and sepsis. An important role of the chemokines has further been shown in the pathogenesis of hemophagocytic syndrome, in the active demyelinating of multiple sclerosis lesions, the modulating of the process of apical periodontitis, the inflammation associated with atopic dermatitis. Stimulatory effects by MIP-1 on synovial fibroblasts play a potential role in the regulation of T-cells in rheumatoid joints in case of rheumatoid arthritis. The chemokine concentrations correlate with infection in lyme borreliosis. High expression was further detected in gastric cancers.
4 STORAGE INSTRUCTIONS

Store kit and components at 2 to 8°C. The expiry of the kit components can only be guaranteed if the components are stored properly, and if, in case of repeated use of one component, the reagent is not contaminated by the first handling.

5 REPRESENTATIVE STANDARD CURVE

Table 1

Representative standard curve.

Do not use this curve to derive test results. A standard curve must be run for each group of samples assayed.

<table>
<thead>
<tr>
<th>Concentration</th>
<th>Fluorescent Intensity</th>
</tr>
</thead>
<tbody>
<tr>
<td>pg/ml</td>
<td>MIP-1α</td>
</tr>
<tr>
<td>10000,0</td>
<td>43,97</td>
</tr>
<tr>
<td>3333,3</td>
<td>26,57</td>
</tr>
<tr>
<td>1111,1</td>
<td>13,05</td>
</tr>
<tr>
<td>370,4</td>
<td>5,12</td>
</tr>
<tr>
<td>123,5</td>
<td>3,27</td>
</tr>
<tr>
<td>41,2</td>
<td>2,31</td>
</tr>
<tr>
<td>13,7</td>
<td>2,08</td>
</tr>
<tr>
<td>0</td>
<td>1,97</td>
</tr>
</tbody>
</table>
6 PERFORMANCE CHARACTERISTICS

6.1 Sensitivity

The limit of detection of MIP-1α defined as the concentration resulting in a fluorescent intensity significantly higher than that of the dilution medium (mean + 2 standard deviations) was determined to be 6.44 pg/ml.

The value shown depends on the type of flow cytometer used for analysis as well as on the respective instrument setup. The value shown is for guidance only. Optimum results for each machine can be achieved by following the instrument set up process.

6.2 Reproducibility

6.2.1 Intra-assay

Reproducibility within the assay was evaluated in 3 independent experiments. Each assay was carried out with 6 replicates of 3 serum samples containing different concentrations of MIP-1α (high, medium and low concentration). 2 standard curves were run on each plate. Data below show the mean intra-assay coefficient of variation for MIP-1α (see Table 2). It has been calculated to be 8.6%.

Individual user data may vary due to differences in protein content of serum/plasma pools or individual donor serum/plasma.

Table 2

The coefficient of variation of the MIP-1α concentration calculated for each sample.

<table>
<thead>
<tr>
<th>Sample</th>
<th>CV high (%)</th>
<th>CV medium (%)</th>
<th>CV low (%)</th>
<th>Mean intra-assay CV (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>MIP-1α</td>
<td>7.5</td>
<td>9.3</td>
<td>9.1</td>
<td>8.6</td>
</tr>
</tbody>
</table>

6.2.2 Inter-assay

Assay to assay reproducibility within one laboratory was evaluated in 3 independent experiments. Each assay was carried out with 6 replicates of 3 serum samples containing different concentrations of MIP-1α (high,
medium and low concentration). 2 standard curves were run on each plate. Data below (see Table 3) show the mean inter-assay coefficient of variation for MIP-1α, calculated on 12 determinations of each sample. It has been calculated to be 4.6%.
Individual user data may vary due to differences in protein content of MIP-1α serum/plasma pools or individual donor serum/plasma.

Table 3
The coefficient of variation of the MIP-1α concentration calculated for each sample.

<table>
<thead>
<tr>
<th>Sample</th>
<th>CV Sample 1 high (%)</th>
<th>CV Sample 2 medium (%)</th>
<th>CV Sample 3 low (%)</th>
<th>Mean inter-assay CV (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>MIP-1α</td>
<td>3.3</td>
<td>6.9</td>
<td>3.7</td>
<td>4.6</td>
</tr>
</tbody>
</table>

6.3 Specificity
There was no detectable crossreactivity observed for other combinable analytes of Simplex and Multiplex Assays from Bender MedSystems. (For detailed information refer to “Combination Table” on www.bendermedsystems.com.)

6.4 Hook effect
Samples with expected concentrations two fold higher than the concentration of highest standard should be diluted 10 fold in Assay Buffer (1x) before assay performance to prevent false negative results due to “hook effects”.

7 BIBLIOGRAPHY


BMS82029FF Human MIP-1α Simplex Kit


For literature update refer to www.bendermedsystems.com
8 ORDERING INFORMATION

For orders please contact:

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**Cat.No. BMS82029FF Human MIP-1α Simplex Kit**