

Magnetic Luminex® Performance Assay Human IL-6 Kit

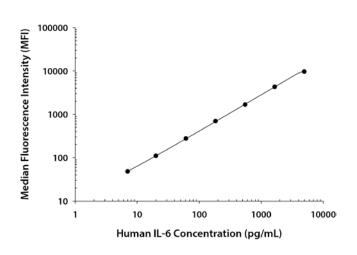
Catalog Number: LUHM206 Pack Size: 100 Tests

SPECIFICATIONS AND USE

| Recommended Sample Types Microparticle Region Components | • • | Cell culture supernates, serum, EDTA plasma, and heparin plasma. Region-27 Microparticle Concentrate (Part 894436) is supplied as a 100X concentrated stock (0.075 mL) with preservatives. |
|--|--------|---|
| | ٠ | Biotin-Antibody Concentrate (Part 892622) is supplied as a 100X concentrated stock solution (0.075 mL) with preservatives. |
| Other Supplies Required | ٠ | Magnetic Luminex Performance Assay Human Base Kit A (Catalog Number LUHM000). |
| Storage | • • | Store the unopened kit at 2-8 °C. Do not use past the expiration date on the label. Avoid freezing microparticles. Protect microparticles from light. |
| Instructions for Use | • | Refer to the Base Kit insert for the Luminex Performance Assay procedure. |

TYPICAL DATA

This human IL-6 standard curve is provided only for demonstration. A standard curve must be generated each time an assay is run, utilizing values from the Standard Value Card included in the Base Kit.



| Standard | pg/mL | MFI | Average | Corrected |
|----------|-------|--------------|---------|-----------|
| Blank | 0 | 15 16 | 16 | |
| 1 | 4950 | 9515 9802 | 9659 | 9643 |
| 2 | 1650 | 4308 4337 | 4323 | 4307 |
| 3 | 550 | 1680 1693 | 1687 | 1671 |
| 4 | 183 | 709 714 | 712 | 696 |
| 5 | 61 | 292 293 | 292 | 277 |
| 6 | 20 | 125 126 | 126 | 110 |
| 7 | 7 | 63 64 | 64 | 48 |

PERFORMANCE CHARACTERISTICS

All data were collected with assays run as a multiplex. Data obtained with polystyrene and magnetic beads were equivalent.

Sensitivity - The Minimum Detectable Dose (MDD) was determined by adding two standard deviations to the MFI of twenty zero standard replicates and calculating the corresponding concentration.

Forty-three assays were evaluated, and the MDD of human IL-6 ranged from 0.10-1.11 pg/mL. The mean MDD was 0.36 pg/mL.

Intra-assay Precision (precision within an assay) - Three samples of known concentration were tested twenty times on one plate to assess precision within an assay.

Inter-assay Precision (precision between assays) - Three samples of known concentration were tested in twenty-five separate assays to assess precision between assays.

| | Int | ra-assay Precisi | on | Inter-assay Precision | | | |
|--------------------|------|------------------|------|-----------------------|------|------|--|
| Sample | 1 | 2 | 3 | 1 | 2 | 3 | |
| n | 20 | 20 | 20 | 25 | 25 | 25 | |
| Mean (pg/mL) | 30.2 | 180 | 903 | 34 | 189 | 900 | |
| Standard Deviation | 1.43 | 7.78 | 41.9 | 3.00 | 13.0 | 53.0 | |
| % CV | 4.7 | 4.3 | 4.6 | 7.9 | 6.7 | 5.9 | |

Recovery and Linearity – Samples containing and/or spiked with high concentrations of IL-6 were evaluated for recovery and were serially diluted to evaluate assay linearity.

| Recovery | | | | Linearity | | | | | |
|-------------------------------|-----------------------|-----------|-----|-----------------------|-----------------------|----------------------------|---------|----------------|-------------------|
| Sample Type | Average % Recovery | Range (%) | | | | Cell culture supernates | Serum | EDTA Plasma | Heparin Plasma |
| Cell culture 96 supernates | 01 100 | | 1.2 | Average % of Expected | 101 | 106 | 103 | 99 | |
| | 96 | 91-100 | | 1:2 | Range (%) | 87-117 | 104-109 | 82-124 | 86-110 |
| Serum 1 | 100 | 95-118 | | 1:4 | Average % of Expected | 103 | 112 | 122 | 101 |
| | 108 | | | | Range (%) | 86-127 | 103-117 | 116-129 | 85-114 |
| EDTA plasma 1 | 102 | 95-116 | | 1.0 | Average % of Expected | 102 | 115 | 126 | 100 |
| | 102 | | | 1:8 | Range (%) | 82-130 | 97-126 | 121-131 | 87-110 |
| Heparin plasma | 109 | 96-129 | | | | · | | · | |

Specificity - This assay recognizes natural and recombinant human IL-6. The assay was tested for cross-reactivity and interference with the following factors. Less than 0.5% cross-reactivity and interference was observed.

| Recombinant human: | | | Recombinant mouse: | | Recombinant rat: | Recombinant porcine: | Recombinant human multiplex partners: | |
|-----------------------|-----------|---------------------|-----------------------|--------|---------------------|----------------------|--|--------|
| 6Ckine | IL-1 RII | IL-17 | G-CSF | IL-8 | GM-CSF | GM-CSF | ENA-78 | IL-8 |
| CNTF | IL-2 Ra | IL-18 | GM-CSF | IL-10 | IFN-7 | IL-1α | FGF basic | IL-10 |
| β-ECGF | IL-2 Rβ | LIF | IFN-γ | IL-17 | IL-1α | IL-1β | G-CSF | IL-17 |
| FGF acidic | IL-2 Rγ | LIF R | IL-1α | MIP-1a | IL-1β | IL-2 | GM-CSF | MCP-1 |
| FGF-4 | IL-3 Ra | MIP-1a | IL-1ra | MIP-1β | IL-2 | IL-4 | IFN-γ | MIP-1a |
| FGF-5 | IL-4 R | MIP-3a | IL-1 | RANTES | IL-4 | IL-6 | IL-1α | MIP-1β |
| FGF-6 | IL-5 Ra | MIP-3β | IL-2 | Тро | IL-6 | IL-8 | IL-1β | RANTES |
| FGF-9 | IL-6 R | MCP-2 | IL-4 | TNF-α | IL-10 | IL-10 | IL-1ra | Тро |
| FGF-10 | IL-10 R | MCP-3 | IL-5 | VEGF | TNF-α | Leptin | IL-2 | TNF-α |
| FGF-18 | IL-3 | MCP-4 | IL-6 | | | TNF-a | IL-4 | VEGF |
| GCP-2 | IL-7 | M-CSF | | | | | IL-5 | |
| $GR0\alpha$ | IL-9 | TNF RI | | | | | | |
| GR0β | IL-11 | TNF-α | | | | | | |
| GR0γ | IL-12 p40 | VEGF ₁₂₁ | | | | | | |
| I-309 | IL-12 p70 | VEGF ₁₆₅ | | | | | | |
| IGF-I | IL-13 | VEGF-D | | | | | | |
| IGF-II | IL-15 | | | | | | | |
| IL-1 RI | IL-16 | | | | | | | |

TECHNICAL HINTS

- Protect the microparticles and streptavidin-PE from light at all times.
- Refer to the Base Kit Standard Value Card for reconstitution volume and values of the reconstituted standard.
- Diluted microparticles cannot be stored. Make a fresh dilution of microparticles each time the assay is run.
- The use of a magnetic device made to accommodate a microplate is necessary for washing.
- Discrepancies may exist in values obtained for the same analyte utilizing different technologies.

Luminex Performance Assays afford the user the benefit of multianalyte analysis of biomarkers in a complex sample. For each sample type, a single, multipurpose diluent is used to optimize recovery, linearity, and reproducibility. Such a multipurpose diluent may not optimize any single analyte to the same degree that a unique diluent selected for analysis of that analyte can optimize conditions. Therefore, some performance characteristics may be more variable than those for assays designed specifically for single analyte analysis.