Miltenyi Biotec

MACSPlex Cytokine 10 Standard

mouse

Order no. 130-106-198

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1. Description

This product is for research use only.

Components	1 vial MACSPlex Cytokine 10 Standard, mouse: Lyophilized mixture of 2 ng recombinant IL-4, IL-5, IL-17A, and IL-23 and 10 ng recombinant lL-2, IL-10, IL-12p70, IFN-γ, TNF-α, and GM-CSF.
Size	For 2 standard curves.
Product format	MACSPlex Cytokine 10 Standard contains stabilizer.
Storage	Store protected from light at –20 °C. The expiration date is indicated on the vial label.

1.1 Background information

MACSPlex Assays have been designed for determining concentrations of soluble analytes in a single sample. The analysis is based on MACSPlex Capture Beads, which display defined fluorescence properties and can be identified using standard flow cytometry techniques.
The MACSPlex Cytokine 10 Standard in combination with MACSPlex Cytokine Reagents Kits, mouse and the MACSPlex Cytokine Basic Kit allows the simultaneous flow cytometric detection of up to seven soluble mouse cytokines in a single sample.

1.2 Applications

- The MACSPlex Cytokine 10 Standard has been developed as a standard control for the quantification of the analytes within the unknown samples.

1.3 Reagent and instrument requirements

- MACSPlex Cytokine Basic Kit (# 130-109-701)
- Up to seven MACSPlex Cytokine Reagents Kits, mouse of choice.
- MACSQuant® Analyzer, MACSQuant Analyzer 10 (# 130-096-343), or other flow cytometers equipped with blue (488 nm) and red (635 nm) lasers able to discriminate FITC, PE, and APC fluorescence.

▲ **Note:** The MACSQuant VYB cannot be used.

2. Protocol

▲ Reconstitute and dilute MACSPlex Cytokine 10 Standard with MACSPlex Buffer provided in the MACSPlex Cytokine Basic Kit, or use the same media as is used for the dilution of the unknown sample.

▲ Only use freshly prepared MACSPlex Cytokine 10 Standard solutions. Do not store or reuse reconstituted or diluted standards.

▲ Use polypropylene or polystyrene reagent tubes. Do not use glass vials.

The generation of standard curves requires eight samples: seven samples of the MACSPlex Cytokine 10 Standard, and one blank control. These samples will be measured as duplicates.

1. Thaw one vial containing the lyophilized MACSPlex Cytokine 10 Standard.
2. Open the vial and add 200 µL of MACSPlex Buffer or media to the pellet. Mix gently. This is the stock solution (10, 000 pg/mL for IL-4, IL-5, IL-17A, IL-23 and 50, 000 pg/mL for IL-2, IL-10, IL-12p70, IFN-γ, TNF-α, GM-CSF).
3. Label six reagent tubes and arrange them in the following order:
   1:5 (1:5^1; 2,000 pg/mL or 10,000 pg/mL)
   1:25 (1:5^2; 400 pg/mL or 2000 pg/mL)
   1:125 (1:5^3, 80 pg/mL or 400 pg/mL)
   1:625 (1:5^4; 16 pg/mL or 80 pg/mL)
   1:3125 (1:5^5; 3.2 pg/mL or 16 pg/mL)
   1:15,625 (1:5^6; 0.6 pg/mL or 3.2 pg/mL).
4. Pipette 200 µL of MACSPlex Buffer or media into each tube.
5. Perform a 1:5 dilution by transferring 50 µL from the stock solution to the tube labeled 1:5 and mix thoroughly. Continue making 1:5 serial dilutions by transferring 50 µL from the tube labeled 1:5 to the tube labeled 1:25 and so on to the tube labeled 1:15,625. Mix each dilution before performing the next transfer.
6. Keep 200 µL MACSPlex Buffer or media as blank control (0 pg/mL).

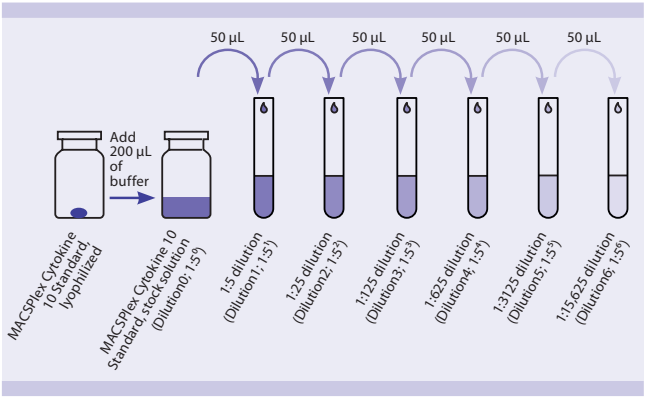


Figure 1: Serial dilution of the MACSPlex Cytokine 10 Standard.

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7. Perform the MACSPlex Cytokine Assay according to the protocol of the MACSPlex Cytokine Basic Kit.

Refer to **www.miltenyibiotec.com** for all data sheets and protocols. Miltenyi Biotec provides technical support worldwide. Visit www.miltenyibiotec.com/local to find your nearest Miltenyi Biotec contact.

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