Miltenyi Biotec

# MHC MACSimer H-2Db FleX Kit, mouse, PE

| | |
|---|---|
| 50 tests | 130-141-813 |
| 100 tests | 130-141-829 |

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## 1. Description

**This product is for research use only.**

| | |
|---|---|
| **Components** | **One kit for 50 tests contains:**<br>1× 150 µL MHC MACSimer H-2Db FleX Reagent, mouse, PE (100 pmol)<br>1× 100 µL MHC MACSimer H-2Db FleX Loading Buffer<br>**One kit for 100 tests contains:**<br>2× 150 µL MHC MACSimer H-2Db FleX Reagent, mouse, PE (100 pmol)<br>2× 100 µL MHC MACSimer H-2Db FleX Loading Buffer |
| **Class** | MHC Class I |
| **Allele** | H-2Db |
| **Antigen** | custom |
| **Peptide sequence** | custom |
| **Species reactivity** | mouse |
| **Release feature** | yes |
| **Control** | MHC MACSimer H-2Db Control, mouse, PE |
| **Product format** | MHC MACSimer FleX Reagents are supplied in buffer containing stabilizer and 0.05% sodium azide. MHC MACSimer FleX Loading Buffer contains 0.05% sodium azide. |
| **Storage** | Store protected from light at +2 to +8 °C. Do not freeze. The expiration date is indicated on the vial label. |

### 1.1 Background information

MHC MACSimer FleX Kits have been designed for the highly flexible analysis of antigen-specific T cells. The antigenic peptide of interest, such as a disease-associated peptide, is loaded onto the provided MHC MACSimer FleX Reagent for subsequent analysis of antigen-specific T cells. With the kit, a single epitope of interest can be easily analyzed in multiple samples. Alternatively, a large number of different (potential) epitopes can be screened in parallel. For sequential multi-sorting or further downstream applications, MHC MACSimers of certain allelic variants can be released from the antigen-specific T cells after staining using REAlease® Technology.

The MHC MACSimer FleX Kits allow maximum flexibility for various applications, such as neoantigen discovery, screening of potential cancer-, autoimmune-, or pathogen-related T cell epitopes, as well as monitoring of antigen-specific T cell responses.

For more information about the MHC multimer technology, refer to the MACS Handbook at **www.miltenyibiotec.com/MHC-multimers-handbook**.

### 1.2 Applications

- Detection and analysis of antigen-specific T cells by flow cytometry
- Sorting and enrichment of antigen-specific T cells by flow sorting or MACS® Technology (magnetic enrichment using Anti-PE MicroBeads)

### 1.3 Reagent and instrument requirements

- Flow cytometer equipped with a red (640 nm), a blue (488 nm), and a violet (405 nm) laser, e.g., MACSQuant® Analyzer 10 (# 130-096-343) or MACSQuant Analyzer 16 (# 130-109-803)
- (Optional) REAlease Support Kit (# 130-120-675)
- (Optional) Cell sorter equipped with a red (640 nm), a blue (488 nm), and a violet (405 nm) laser, e.g., MACSQuant Tyto® (# 130-103-931)
- (Optional) Anti-PE MicroBeads (# 130-048-801) or Anti-PE MicroBeads UltraPure (# 130-105-639)
- (Optional) MHC MACSimer H-2Db Control, mouse, PE, (# 130-141-871, # 130-141-881)
- (Optional) Spleen Dissociation Kit, mouse, (# 130-095-926)
- (Optional) Fluorochrome-conjugated antibodies for flow cytometric analysis. Recommended antibodies for the staining cocktail are listed in **Table 1**. For more information about antibodies refer to **www.miltenyibiotec.com/antibodies**.

| Antibody | Clone |
|---|---|
| CD3 Antibody, anti-mouse, REAfinity™ | REA641 |
| CD8a Antibody, anti-mouse, REAfinity™ | REA601 |
| CD11b Antibody, anti-mouse, REAfinity™ | REA592 |
| CD19 Antibody, anti-mouse, REAfinity™ | REA749 |

**Table 1:** Recommended antibodies for the staining cocktail.


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## 2. Protocol

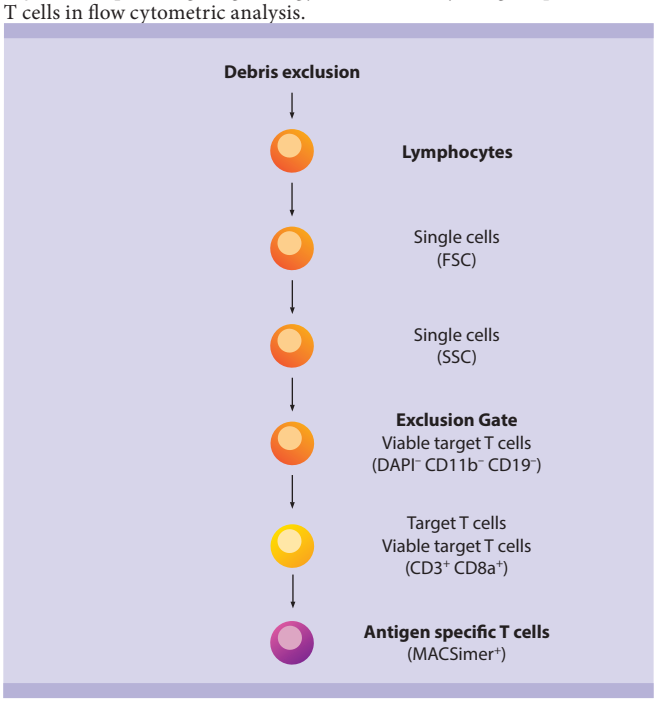
▲ The detailed protocol and an exemplary MHC MACSimer staining data set are available at **www.miltenyibiotec.com/peptide-loaded-MHC-class-1-MACSimer-staining.html.**

| | 50 tests | 100 tests |
|---|---|---|
| Number of loading reactions | 5 with 50 µL each | 10 with 50 µL each |
| Number of staining reactions | ≥50 with $10^6$ cells each | ≥100 with $10^6$ cells each |

**Table 2:** Kit capacities for loading and staining reactions.

## 3. Example of Gating Tree

**Figure 1:** Sequential gating strategy used to identify antigen-specific $CD8^+$ T cells in flow cytometric analysis.




Refer to **www.miltenyibiotec.com** for all data sheets and protocols. Miltenyi Biotec provides technical support worldwide. Visit www.miltenyibiotec.com for local Miltenyi Biotec Technical Support contact information.

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