MBL
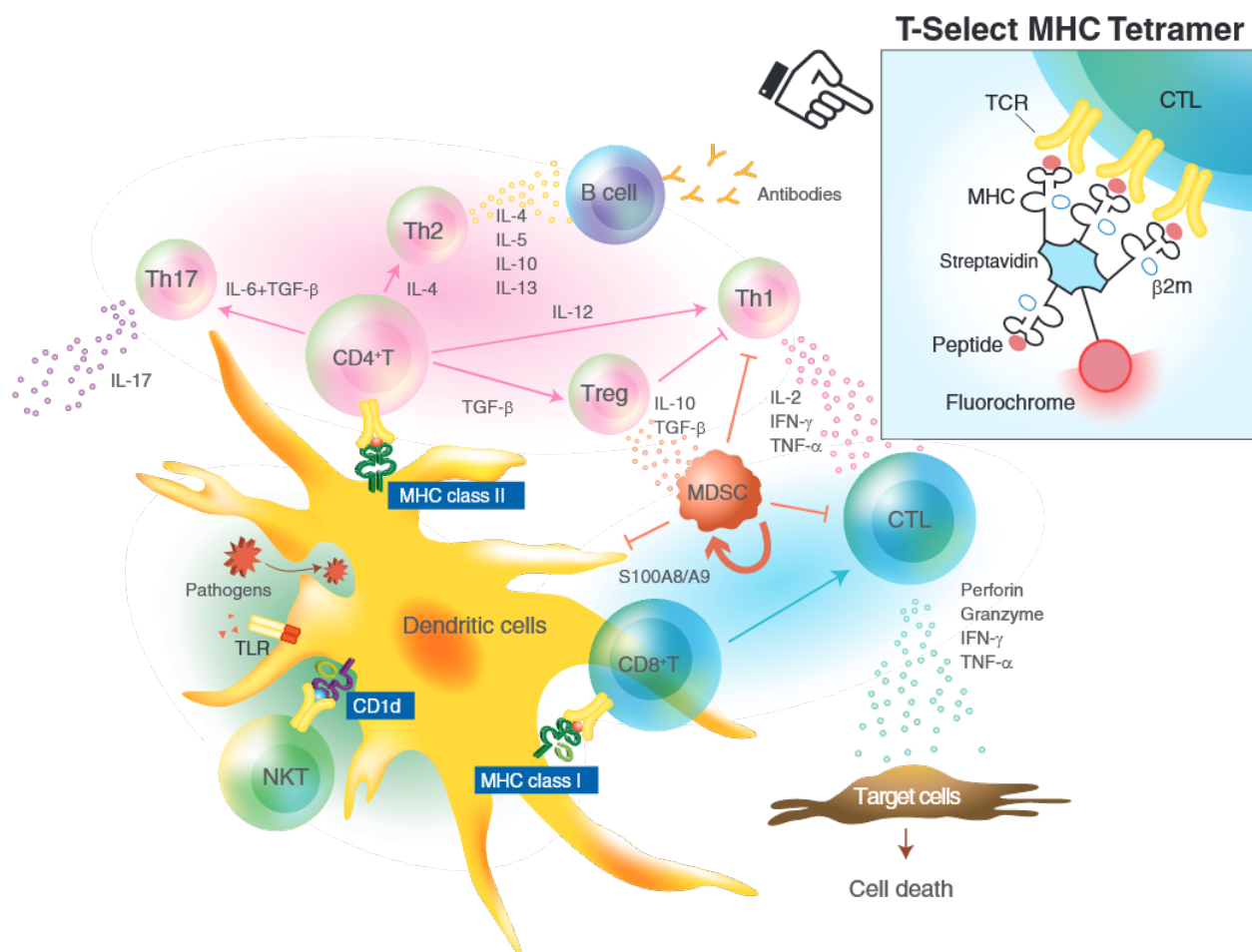A JSR Life Sciences Company

T-Select MHC Tetramer Product Catalog

T-Select MHC Tetramer

Class I and class II major histocompatibility complex (MHC) tetramer reagents allow rapid and simple detection of antigen-specific T cells. MHC tetramer technology is based on the ability of MHC–peptide molecules to recognize antigen-specific T cells at the single cell level. This breakthrough technology enables researchers to precisely measure targeted T-cell responses in infectious diseases, cancer, and autoimmune diseases.

T-Select MHC Tetramers are available as ready-to-use reagents conjugated to a fluorochrome such as fluorescein isothiocyanate (FITC), phycoerythrin (PE), or allophycocyanin (APC). Whole blood or isolated peripheral blood mononuclear cells (PBMCs) are appropriate sample types for tetramer analysis. MHC tetramers are used in combination with other T-cell-specific monoclonal antibody reagents and flow cytometry to provide exquisite specificity and sensitivity for the identification and isolation of rare-event, antigen-specific T cells. Monitoring of antigen-specific T cell immune responses is thought to be the most important and relevant outcome of anti-tumor or anti-viral responses during the development of vaccines and therapies. These antigen-specific T cells can be detected using MBL T-Select MHC Tetramers.

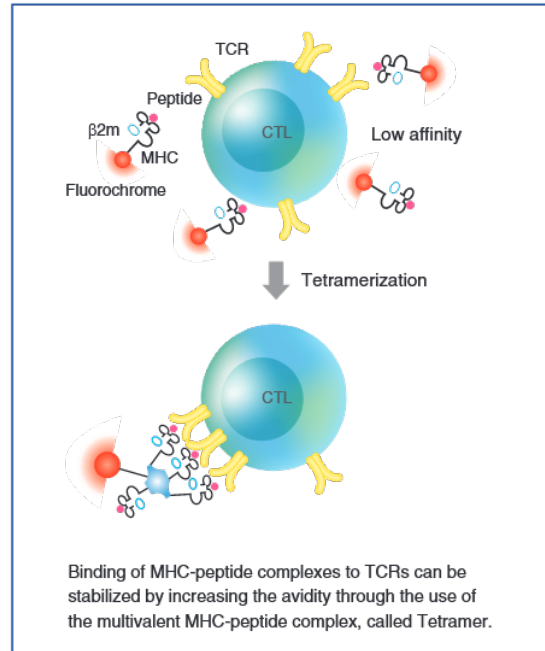# Contents

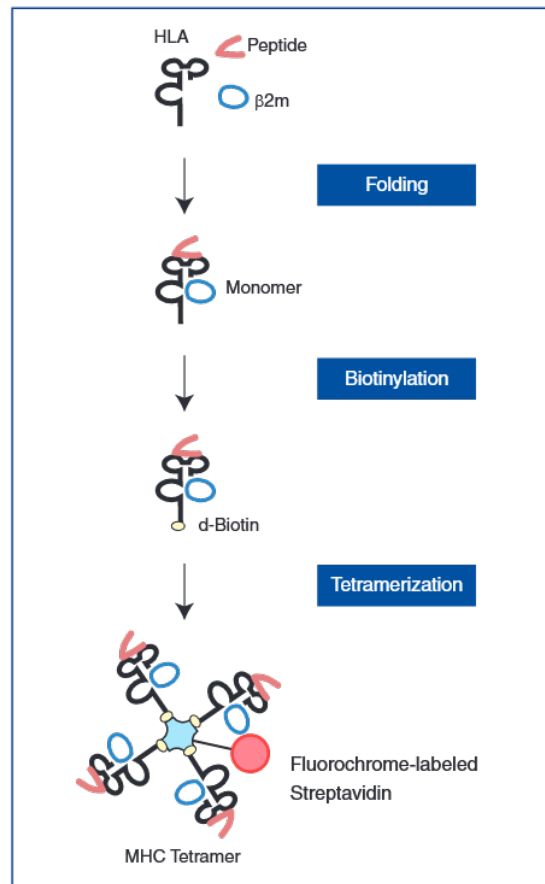# 1. Principle of Detection of Antigen-specific T cells by MHC Tetramers

Most T lymphocytes express a clonal and highly specific antigen receptor (TCR) on the cell surface. TCR (T cell receptor) binds to the MHC-peptide complex and recognizes the antigenic peptide presented in the context of MHC molecules. Such interaction initiates the generation of adaptive cellular immunity such as cell-mediated and humoral immunity. Because TCR has a low avidity and fast off-rates for MHC-peptide complexes, recombinant soluble monomeric MHC-peptide complexes have not been used to detect antigen-specific T cells. Altman et al.[1] have introduced MHC-peptide tetrameric complexes (so-called MHC tetramers) for detection of antigen-specific T cells. MHC tetramers have increased avidity for their cognate TCRs and are successfully used to directly visualize antigen-specific T cells *ex vivo* by flow cytometry.

Binding of MHC-peptide complexes to TCRs can be stabilized by increasing the avidity through the use of the multivalent MHC-peptide complex, called Tetramer.

2. Preparation of MHC class I Tetramers

MHC Tetramers are complexes of four MHC molecules, associated with a specific peptide and bound to a fluorochrome. To greatly reduce non-specific binding, MBL's class I MHC Tetramers specific to human alleles have a proprietary mutation in the α3 domain. MHC Tetramers are generated essentially as described by Altman et al.[1]. The manufacture process is outlined in the figure on the right side.

Purified recombinant MHC (heavy chain) and human β2-microglobulin (β2m) are refolded in molar excess of the appropriate 8- to 10-mer peptide for several days. High performance liquid chromatography (HPLC) has been extensively used to monitor the generation of monomeric MHC-peptide complexes (monomers). The refolded monomer is biotinylated with a single biotin by the BirA enzyme at the *C*-terminal end of the heavy chain. The biotinylated monomers are purified by streptavidin–agarose affinity column chromatography. Subsequently, purified monomers are linked by the addition of fluorochrome-labeled streptavidin.

<Reference>
1) Altman JD *et al.* Science 274, 94–96 (1996)

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MHC Tetramer staining method

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H-2Kᵇ OVA Tetramer

CD1d Tetramer

QuickSwitch™ Custom Tetramer Kits

MHC Class I custom Tetramer

MHC Class II custom Tetramer

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3. High specificity of T-Select HLA class I Tetramers

The T cell surface CD8 enhances T cell antigen recognition by binding to HLA class I molecules. Therefore, we produced T-Select HLA class I Tetramers with one point mutation (Ala245Val) at the HLA α3 domain known to reduce the CD8-HLA interaction. These mutated tetramers showed a greatly diminished nonspecific binding but retained specific binding[3]. Alterations of CD8 binding by mutation of the HLA greatly improved the specificity of HLA-peptide multimers, thus providing efficient tools to sort specific human T cells for immunotherapy.
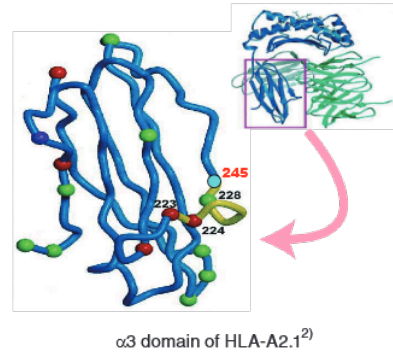
<Reference>
2) Gao GF *et al*. Nature 387, 630–634 (1997)
3) Bodinier M *et al*. Nat. Med. 6, 707–710 (2000)

α3 domain of HLA-A2.1[2]

The example of staining 1: HLA-A*02:01 CMV pp65 Tetramer

Native Tetramer

Non-mutated tetramers can bind CTLs via CD8, independent of TCR, leading to detection of antigen-non-specific irrelevant CTLs by undesired binding between CD8 and MHC molecules.

T-Select HLA class I Tetramer

Antigen-specific TCR can be detected selectively.

Conventional non-mutated Tetramer (HLA-A*02:01 CMV pp65)

A — Cell population containing HLA*02:01 CMV pp65 specific CTL
B — Cell population NOT containing HLA*02:01 CMV pp65 specific CTL

α3 mutated Tetramer (HLA-A*02:01 CMV pp65)

C — Cell population containing HLA*02:01 CMV pp65 specific CTL
D — Cell population NOT containing HLA*02:01 CMV pp65 specific CTL

CMV Positive Sample: T-Select Tetramers show substantially reduced non-specific CD8 binding (C) compared to Native (wild type) Tetramers (A).
CMV Negative Sample: T-Select HLA Tetramers produce accurate results (D) compared to Native Tetramers that produce false-positive results (B).

The example of staining 2: HLA-A*24:02 EBV BRLF1 Tetramer

PBMC were separated from peripheral blood of HLA-A*24:02 positive healthy subjects. EBV BRLF1-specific CTLs were induced by the MLPC method (see page 7) and stained with PE-labeled HLA-A*24:02 Negative (Code No. TS-M007-1), or EBV BRLF1 Tetramer with (Code No. TS-M002-1) or without the α3 mutation. Non-specific staining of CD8 positive cells was observed in the sample stained with the conventional, but not the α3-mutated, tetramer.

EBV BRLF1 Tetramer

Negative Tetramer | With α3 mutation | Without α3 mutation
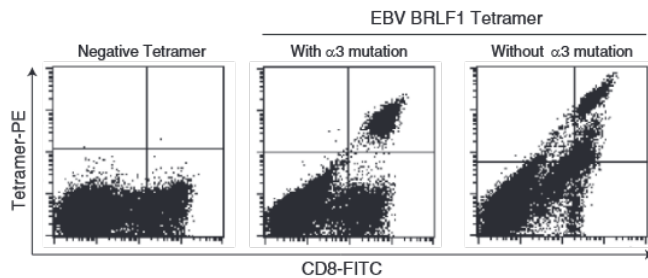
This data was analyzed within the lymphocyte gate and 7-AAD negative gate.
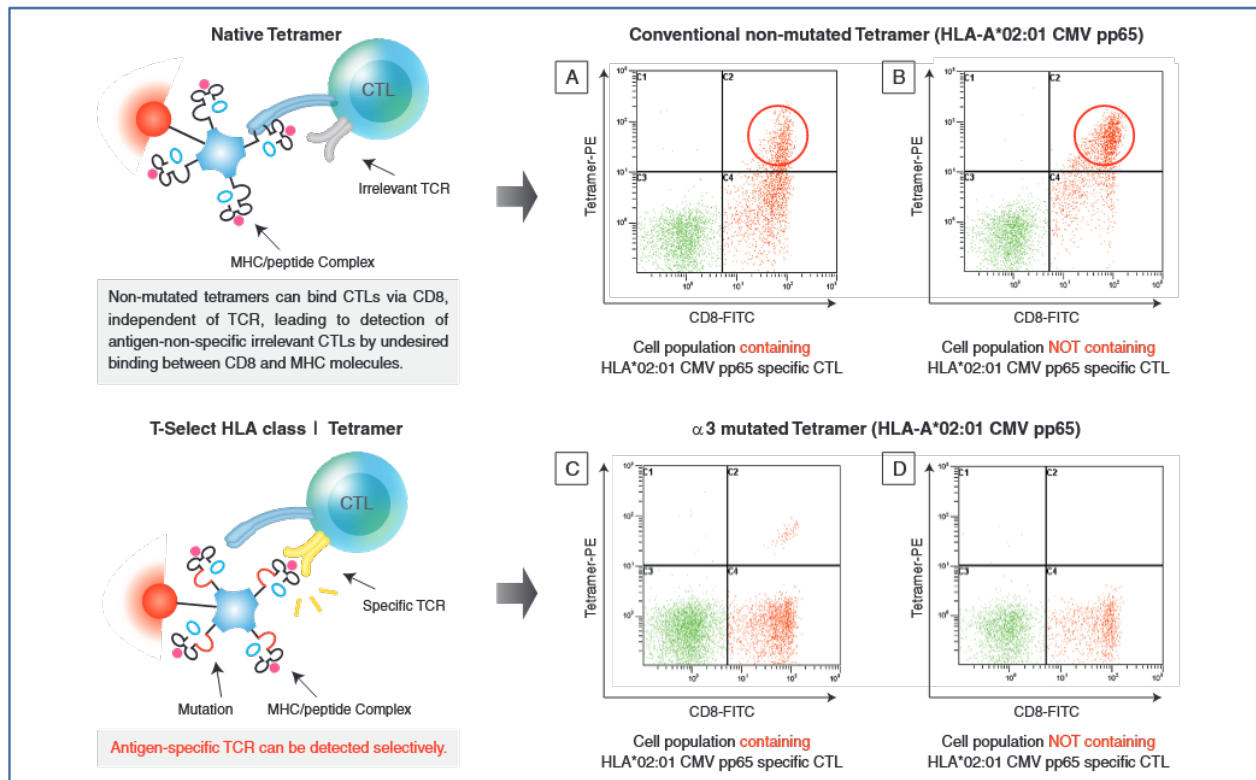
Principle of Detection of Antigen-specific T cells by MHC Tetramers
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4. MHC Tetramer staining method

4-a. Procedure for whole blood samples (Human)

1. Collect blood by venipuncture into a blood collection tube containing an appropriate anti-coagulant.
2. Add 10 μL of T-Select MHC Tetramer to each 12 x 75 mm test tube.
3. Add 200 μL of whole blood into each test tube.
4. Vortex gently.
5. Incubate for 30-60 minutes at 2-8°C or room temperature (15-25°C) protected from light.
6. Add any additional antibodies (e.g. anti-CD8) and vortex gently.
7. Incubate for 30 minutes at 2-8°C protected from light.
8. Lyse red blood cells using commercially available reagents.
9. Prepare samples according to description of the package insert.
10. Store prepared samples at 2-8°C protected from light for a minimum of 1 hour (maximum 24 hours) prior to analysis by flow cytometry.

Blood collection	Tetramer reaction Incubation	Hemolysis Fixation

MHC Tetramer-PE   10 μL
CD8-FITC   20 μL

Suspend in PBS

Analysis by FCM
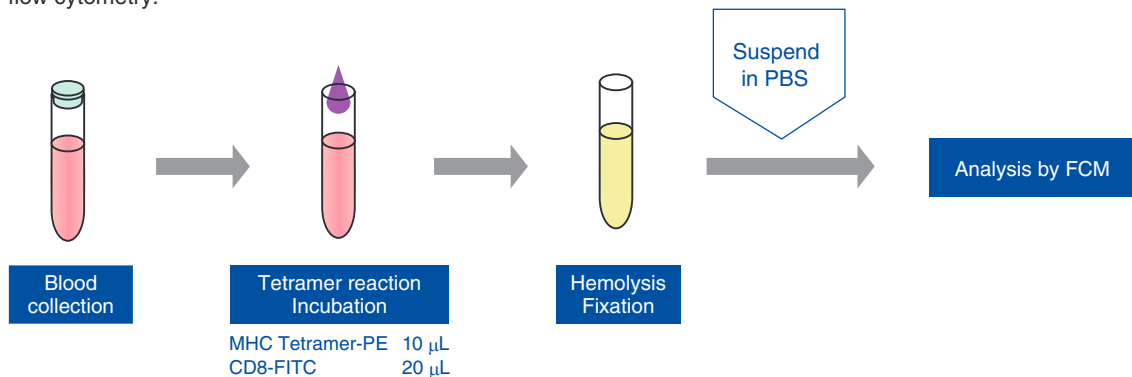
* If hemolysis is incomplete, a non-specific staining pattern caused by diffuse reflection from erythrocytes may be observed. Inclusion of a CD45 antibody can help identify the true lymphocyte population.

lymphocyte gating   CD45 gating

SSC-H / FSC-H   CD45-PC5

Gating out of diffuse reflection from erythrocytes by CD45 staining resulted in reduced noise.

HLA-A*24:02 BRLF1   HLA-A*24:02 Negative

Tetramer-PE

CD8(T8)-FITC

Only lymphocyte gating

Lymphocyte gating + CD45 gating

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4-b. Procedure for Peripheral Blood Mononuclear Cells (PBMC) samples (Human)

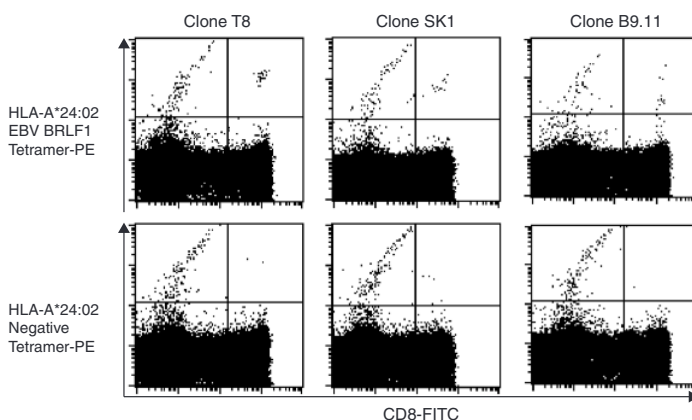1. Prepare peripheral blood mononuclear cells (PBMC) according to established procedures. Cells should be re-suspended at a concentration of 2×10^7 cells/mL. 50 μL of sample is required for each T-Select MHC Tetramer determination.
2. Add 10 μL of Clear Back (human FcR blocking reagent, Code No. MTG-001) to each 12 x 75 mm test tube.
3. Add 50 μL PBMC into each test tube (e.g. 1×10^6 cells per tube).
4. Incubate for 5 minutes at room temperature.
5. Add 10 μL of T-Select MHC Tetramer and vortex gently.
6. Incubate for 30-60 minutes at 2-8°C or room temperature (15-25°C) protected from light.
7. Add any additional antibodies (e.g. anti-CD8) and vortex gently.
8. Incubate for 30 minutes at 2-8°C protected from light.
9. Add 3 mL of PBS or FCM buffer (2% FCS/0.09% NaN$_3$/PBS).
10. Centrifuge tubes at 400 x g for 5 minutes.
11. Aspirate or decant the supernatant.
12. Resuspend the pellet in 500 μL of PBS with 0.5% formaldehyde.
13. Store prepared samples at 2-8°C protected from light for a minimum of 1 hour (maximum 24 hours) prior to analysis by flow cytometry.

4-c. Procedure for Mouse spleen samples

1. Collect lymph node, spleen or thymus and prepare a single-cell suspension according to an established protocol. Cells should be re-suspended at a concentration of 2×10^7 cells/mL. 50 μL of sample is required for each T-Select MHC Tetramer determination.
2. To each 12 x 75 mm test tube add 10 μL of Clear Back (human FcR blocking reagent, Code No. MTG-001).
3. Add 50 μL cell suspension into each test tube (e.g. 1×10^6 cells per tube).
4. Incubate for 5 minutes at room temperature.
5. Add 10 μL of T-Select MHC Tetramer and vortex gently.
6. Incubate for 30-60 minutes at 2-8°C protected from light.
7. Add any additional antibodies (e.g. anti-CD8) and vortex gently.
8. Incubate for 30 minutes at 2-8°C protected from light.
   If red blood cell lysis is necessary, proceed to step 8-16 in the **Procedure for Whole Blood** section. If red blood cell lysis is not necessary, continue to step 9 below.
9. Add 3 mL of PBS or FCM buffer (2% FCS/0.09% NaN$_3$/PBS).
10. Centrifuge tubes at 400 x g for 5 minutes.
11. Aspirate or decant the supernatant.
12. Resuspend the pellet in 500 μL of PBS with 0.5% paraformaldehyde or formalin.
13. Store at 4°C protected from light for a minimum of 1 hour (maximum 24 hours) prior to analysis by flow cytometry.

4-d. Staining differences among human CD8 antibody clones

PBMC separated from peripheral blood of HLA-A*24:02 positive healthy subjects were simultaneously stained with HLA-A*24:02 EBV BRLF1 Tetramer-PE (Code No. TS-M002-1) or HLA-A*24:02 Negative Tetramer-PE (Code No. TS-M007-1) with three different human CD8 antibody clones: T8, SK1, and B9.11. A distinct population of specific CTL was better resolved when clone T8 was used compared to the other two clones.

<Reference>
Rita C *et al*. Int. Immunol. 14, 39–44 (2002)

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5. Induction and detection of antigen specific CTL

5-a. Induction of CTL from peripheral blood using the Mixed Lymphocyte-Peptide Culture method

   MHC Tetramers are used as detection reagents for viral or cancer antigen-specific CTL. Because of their relative rarity compared with viral antigen-specific CTL, cancer antigen-specific CTL can be difficult to detect immediately following blood collection. Using the Mixed Lymphocyte-Peptide Cultures (MLPC) method, Karaniks *et al*. found that increased specific CTL in peripheral blood resulted from vaccine therapy to melanoma patients (J. Immunol., 2003, 171, 4898). The MLPC method expands antigen-specific CTL via culture of buffy coat cells with a stimulating peptide, thus allowing for the enumeration of low frequency CTLs, which are below detection in freshly isolated blood. Using the following modified MLPC protocol, MBL has successfully induced cancer antigen-specific CTL from peripheral blood of healthy subjects.

Heparinized blood
Centrifugation RT
3,000 rpm
10 min.

Separation of plasma
Add equal value
of serum free
RPMI1640

Separation of Lymphocyte

Stimulation by Antigen

Tetramer staining

Virus antigen — Several hours – 15 days
Cancer antigen — 12–15 days — 96 well plate
DAY 0
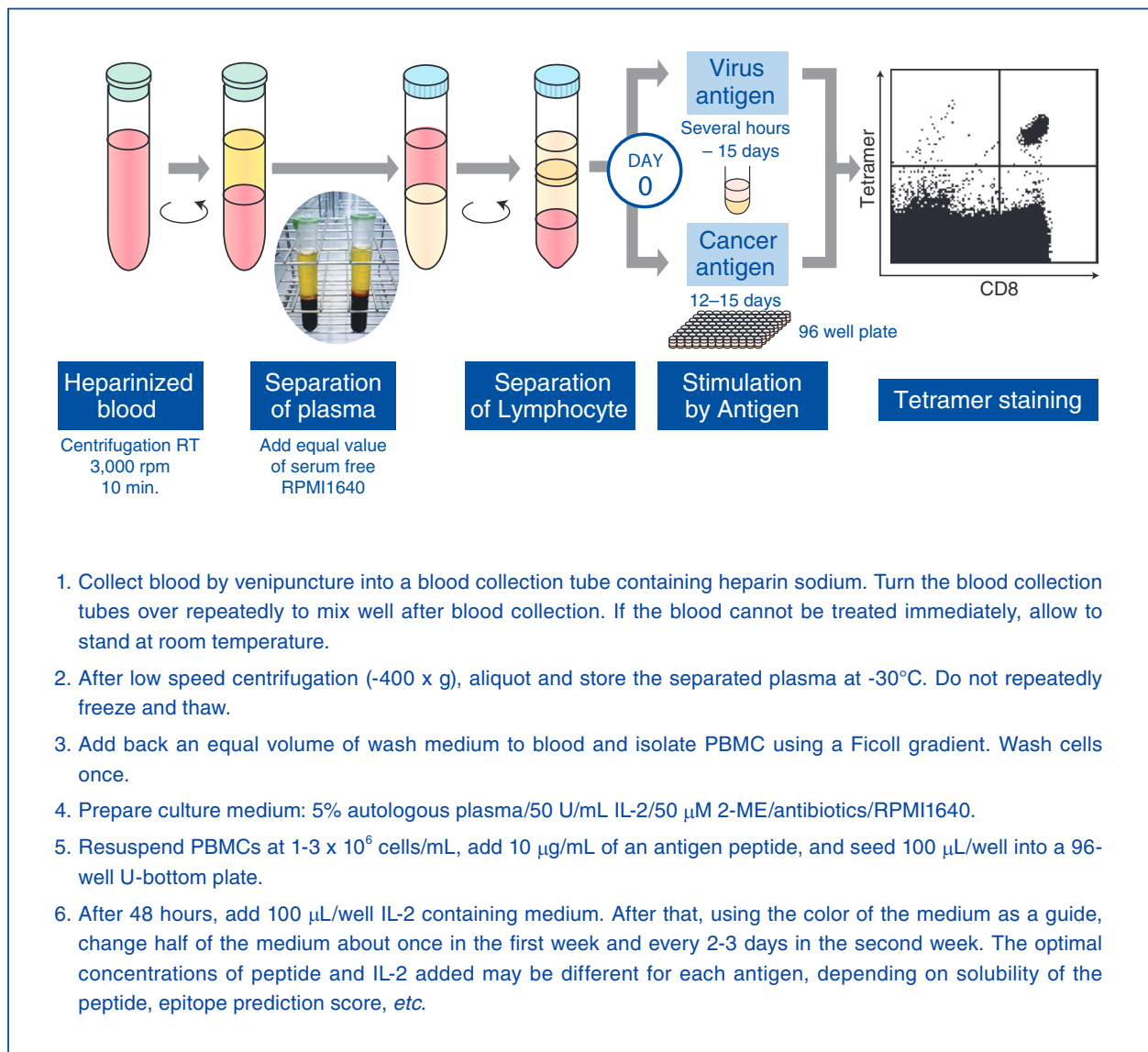
1. Collect blood by venipuncture into a blood collection tube containing heparin sodium. Turn the blood collection tubes over repeatedly to mix well after blood collection. If the blood cannot be treated immediately, allow to stand at room temperature.

2. After low speed centrifugation (-400 x g), aliquot and store the separated plasma at -30°C. Do not repeatedly freeze and thaw.

3. Add back an equal volume of wash medium to blood and isolate PBMC using a Ficoll gradient. Wash cells once.

4. Prepare culture medium: 5% autologous plasma/50 U/mL IL-2/50 μM 2-ME/antibiotics/RPMI1640.

5. Resuspend PBMCs at 1-3 x 10^6 cells/mL, add 10 μg/mL of an antigen peptide, and seed 100 μL/well into a 96-well U-bottom plate.

6. After 48 hours, add 100 μL/well IL-2 containing medium. After that, using the color of the medium as a guide, change half of the medium about once in the first week and every 2-3 days in the second week. The optimal concentrations of peptide and IL-2 added may be different for each antigen, depending on solubility of the peptide, epitope prediction score, *etc*.

Note:
Induction of viral antigen-specific CTL can often be achieved by the MLPC method using tubes. In order to induce cancer antigen-specific CTL, the 96-well plate MLPC method is recommended because the frequency of cancer antigen-specific CTL is low.

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CD1d Tetramer

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MHC Class II custom Tetramer

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5-b. Induction and detection of cancer antigen specific CTL

PBMC were separated from peripheral blood of healthy subjects and cancer antigen-specific CTL were induced by the 96-well MLPC method. Fourteen days after peptide stimulation, the cells were stained with Tetramer as follows: First, an aliquot of cells from each of 8 wells in a column of a 96-well plate were collected to make 12 pooled samples. These samples were stained separately with tetramer.

Pools yielding antigen-specific CTL were further deconvoluted by testing for tetramer binding to cells from individual wells that contributed to the positive pool.

Based on the number of positive wells, the frequency of antigen-specific CTL at blood collection was calculated using the following equation.

$$= \frac{(\text{Number of Tetramer positive well})}{(\text{Cell number / well}) \times (\text{Total well number}) \times (\text{CD8 positive cell rate in day 0})}$$

$$= \frac{2}{2 \times 10^5 \times 96 \times 0.35} = 2.98 \times 10^{-7} \text{of whole blood CD8 T cell}$$

Note:
Induction of viral antigen-specific CTL can often be achieved by the MLPC method using tubes. In order to induce cancer antigen-specific CTL, the 96-well plate MLPC method is recommended because the frequency of cancer antigen-specific CTL is low.
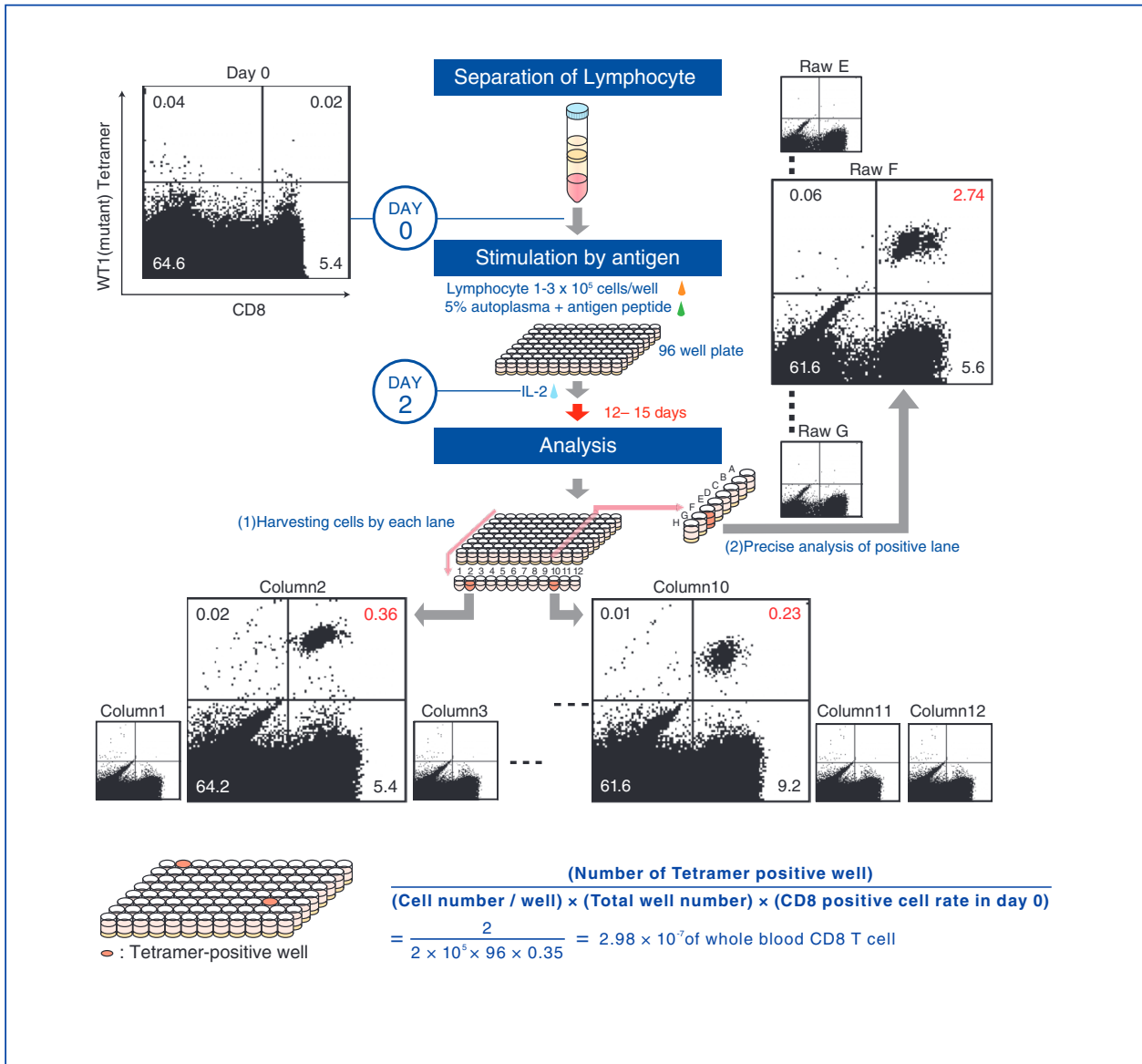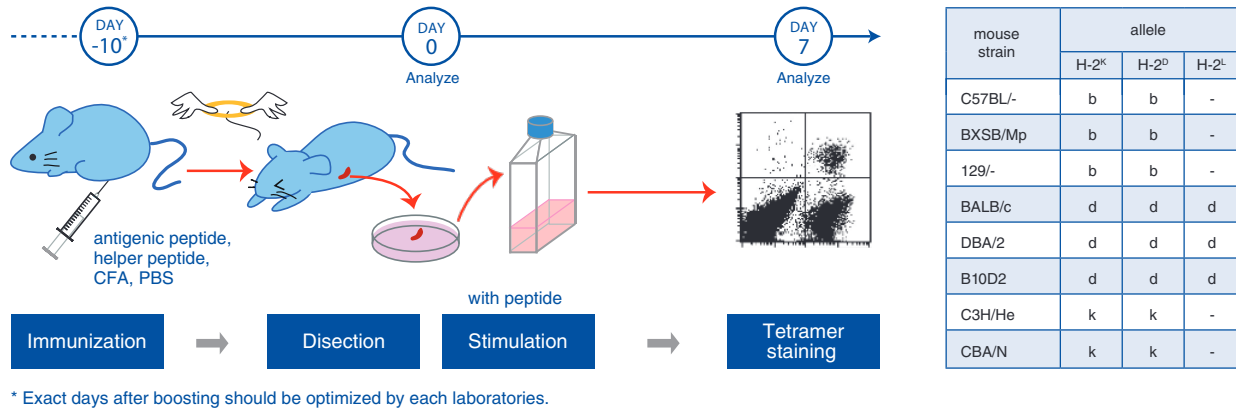
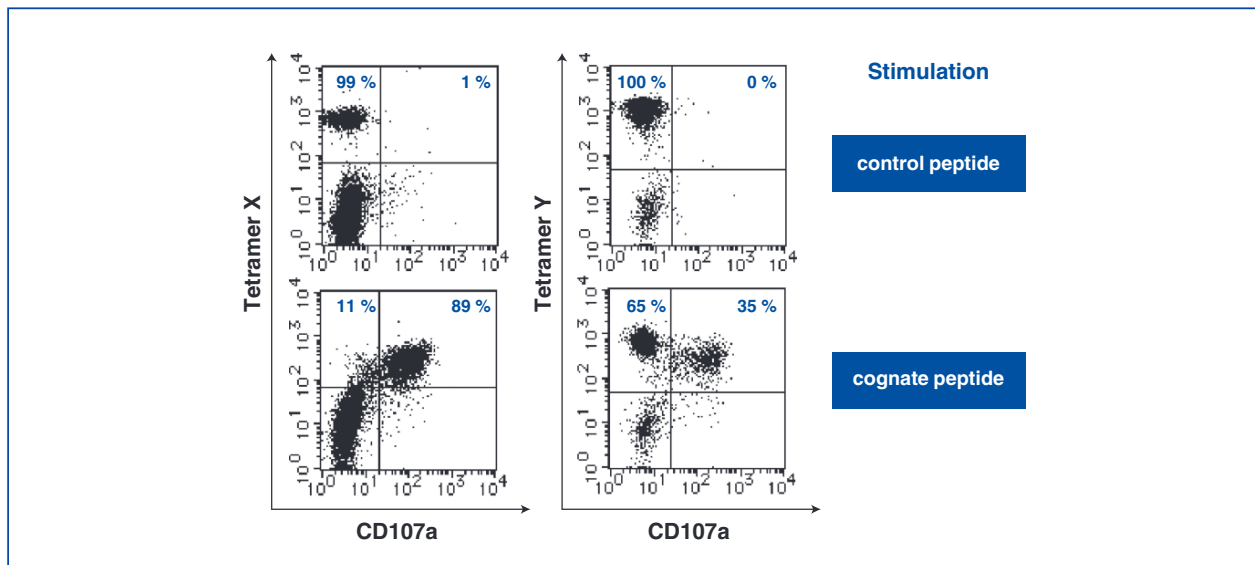## 5-c. Induction method for antigen-specific murine CTL

Mouse models are commonly used to study various *in vivo* immune responses. Mouse MHC Tetramers can detect murine antigen-specific CTL. MBL conducted a study showing that antigen-specific CTL can be induced rapidly and easily at low cost using the following method. First, the antigen peptide of interest was mixed with a second "helper" peptide designed to induce T helper activity. This mixture was emulsified with adjuvant, and intraperitoneal (IP) immunizations were performed. Spleens were harvested 7-11 days after the final immunization, and splenocytes were stimulated by the peptides *in vitro* for 1 week. The times of immunization depend on the antigen used. In a study conducted by MBL, antigen-specific CTL could be induced in 1-4 rounds of immunization. Immunization of two or more mice per antigen is recommended due to individual differences. Please refer to the data sheet of each tetramer product for additional information.

mouse strain	allele		
	H-2^K	H-2^D	H-2^L
C57BL/-	b	b	-
BXSB/Mp	b	b	-
129/-	b	b	-
BALB/c	d	d	d
DBA/2	d	d	d
B10D2	d	d	d
C3H/He	k	k	-
CBA/N	k	k	-

* Exact days after boosting should be optimized by each laboratories.

6. Measurement methods for antigen-specific CTL using MBL Kits

6-a. IMMUNOCYTO CD107a Detection Kit

When activated by antigen stimulation, CTLs release cytotoxic factors including perforin and granzyme from intracellular granules. During this process, CD107a (LAMP-1) in the inner membrane of intracellular granules is exposed on the cell membrane. IMMUNOCYTO CD107a Detection Kit (Code No. 4844) enables indirect measurement of CTL cytotoxicity by detecting CD107a mobilization. While intracellular flow staining for IFN-γ has been found to be more sensitive than CD107a mobilization, a recent study conducted by MBL identifying new CTL epitopes showed the latter better correlated with Tetramer staining. In addition to being more specific, the CD107a mobilization assay is more rapid than cytokine flow cytometry.

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6-b. IMMUNOCYTO Cytotoxicity Detection Kit

*This kit does not include CFSE

T cells and NK cells (effector cells) directly recognize virus-infected and tumor cells (target cells) and can respond with cytotoxic activity to injure or kill target cells. While the ^{51}Cr release assay has been a reliable method to measure cytotoxic activity, special training and facilities for radioisotopes are required. Therefore, methods to measure cytotoxic activity without using radioisotopes have been developed. The principle to detect cytotoxic activity of effector cells using IMMUNOCYTO Cytotoxicity Detection Kit (Code No. AM-1005M) is to double stain the target cells with both CFSE and Kusabira-Orange labeled Annexin V, and measure cytotoxic activity by flow cytometry. Target cell death (as a consequence of effector cell cytotoxicity) is measured based on the ratio of cells stained with Annexin V to cells stained with CFSE, as described below. Because the effector cells are not stained with CFSE, they are easily distinguished by flow cytometry for analysis. This non-radioactive method is reported to have high correlation with the ^{51}Cr release assay making it a safer alternative.

Principle of apoptosis detection by Annexin V

Phosphatidylserine (PS) located on the inner side of the lipid bilayer is exposed on the cell surface in apoptotic cells due to change in membrane structure (right figure). Annexin V binds with high affinity to PS in a Ca^{2+} dependent manner, effectively labeling apoptotic cells (Figure 1). By using fluorescently-labeled Annexin V, the target cells damaged by effector cells can be detected by flow cytometry. IMMUNOCYTO Cytotoxicity Detection Kit contains fluorescent protein Kusabira-Orange (KO; Excitation Max 548 nm/Emission Max 559 nm)-labeled Annexin V.

Figure 1

Living cells / **Apoptotic cells**
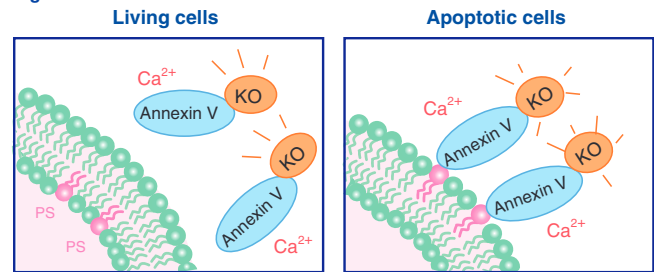
PS of living cells is not exposed on the cell surface and does not bind to Annexin V.

In apoptotic cells, the membrane structure is destroyed, PS is exposed on the surface of the cells, and PS binds to Annexin V.

Data analysis by flow cytometry

Cytotoxic activity measured by IMMUNOCYTO Cytotoxicity Detection Kit is shown for specific and negative control targets. Effector cells (E) and target cells (T) are indicated in Figure 2. When killing takes place, CFSE-single positive target cells become dual positive for CFSE and Annexin V. Cytotoxic activity can be calculated from percentages of single and dual positive cells using the following equation:

Cytotoxicity (%) = [(ET-T_0)/(100-T_0)]×100

ET: % CFSE+ Annexin V+ cells when effector cells and target cells are co-cultured.
T_0: % CFSE+ Annexin V+ cells when only target cells are cultured.

Figure 2

Cytotoxicity: 84.64% / Cytotoxcity: 0.32%
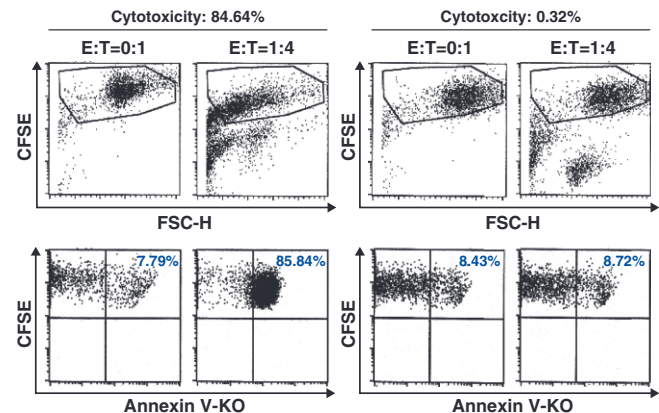
E: A*24:02 EBV BRLF1 CTL line
T: BRLF1 peptide pulsed HLA-A24 LCL

E: A*24:02 EBV BRLF1 CTL line
T: HLA-A2 LCL

Graphs showing cytotoxic activity

Graphs showing cytotoxic activity by HLA-A*24:02 CMV pp65 CTL line against various targets illustrate the specificity of CTL killing, as well as the effect of different E:T ratios (Figure 3).

Figure 3
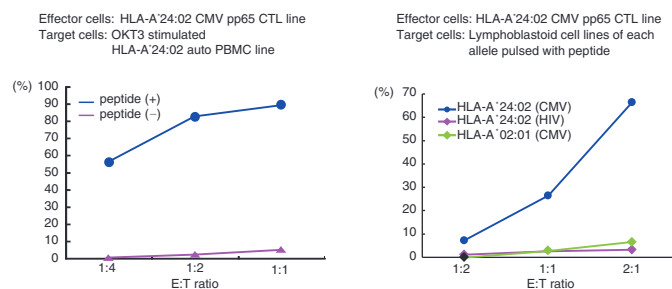
Effector cells: HLA-A*24:02 CMV pp65 CTL line
Target cells: OKT3 stimulated HLA-A*24:02 auto PBMC line

peptide (+)
peptide (−)

Effector cells: HLA-A*24:02 CMV pp65 CTL line
Target cells: Lymphoblastoid cell lines of each allele pulsed with peptide

HLA-A*24:02 (CMV)
HLA-A*24:02 (HIV)
HLA-A*02:01 (CMV)

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7. MHC Tetramer related products

7-a. Blocking reagent for tetramer staining

When a very rare cell population is detected by a tetramer reagent, it is important to reduce non-specific and Fc-specific staining. Clear Back (Code No. MTG-001) is a reagent that blocks non-specific binding to both human and murine cells in flow cytometry and fluorescence microscopy experiments. As shown in Figure 1, Clear Back reduces Fc-specific staining of monocytes by a mouse IgG2a. Clear Back also reduced background staining of PBMC with HLA-A*24:02 EBV BRLF1 Tetramer in the tetramer-positive CD8-negative region (Figure 2).

Effect of Clear Back on peripheral blood mononuclear cells

Figure 1

mouse IgG2a-FITC (5 μg/mL)

Red：Clear Back treatment
Green：No Clear Back treatment

Lymphocyte   **monocytes**

Clear Back treatment reduced non-specific staining of isotype control by monocytes.

Figure 2

Clear Back

Without Clear Back

Clear Back treatment reduced non-specific binding of CD8 negative Tetramer positive cells.
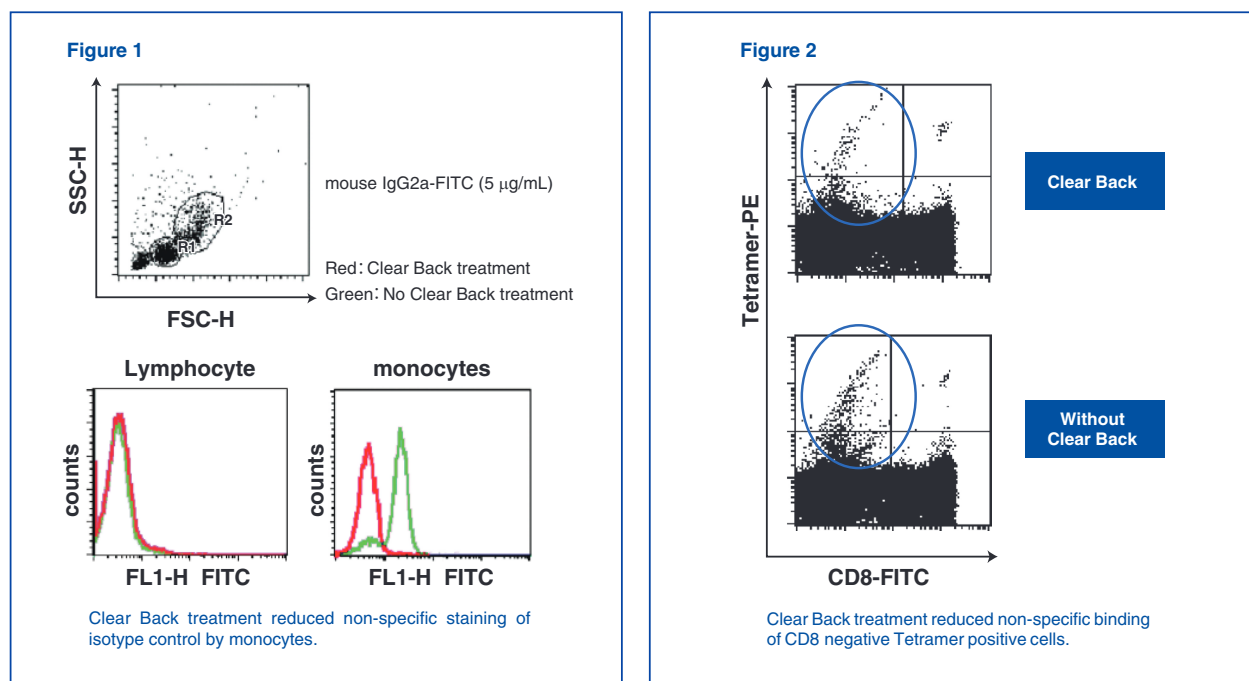
7-b. Peptides for antigen-specific CTL induction

MBL offers ready-to-use epitope peptides in liquid form for inducing CTL by the MLPC method. These are the same peptides that are used to produce MHC Tetramers.

Peptides for inducing murine CTL and antigenic peptides that induce T-helper immunity are also available. Please refer to the product list for product lineup.

Concentration	Volume	Solvent	Purity *
10 mg/mL	1 mg (100 μL)	DMSO	≧ 95%

*Some peptides do not apply the purity of ≧ 95%.

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8. H-2K^b OVA Tetramer

8-a. Introduction; OVA

Ovalbumin (OVA), the major protein found in chicken egg whites, is a T cell-dependent antigen commonly used as a model protein for studying antigen-specific immune responses in mice. H-2K^b OVA Tetramer detects OVA-specific T cells in C57BL/6 and other mouse strains expressing the class I allele, H-2K^b.

8-b. Effect of CD8 antibody clones on H-2K^b OVA Tetramer staining: "Cells from OT-I mice"

Splenocytes from mice transgenic for OVA-specific T cells (OT-I) were used to explore staining differences among mouse CD8 antibody clones in combination with H-2K^b Tetramers. OT-I mouse spleen cells (1×10^6 cells/sample) were stained with H-2K^b OVA Tetramer-PE (10 μL/sample) and serially diluted anti-mouse CD8 (clone KT15 or clone 53.6.7) antibody in a final assay volume of 100 μL. H-2K^b β-galactosidase (β-gal) Tetramer-PE (10 μL/sample) was used as a negative tetramer to assess non-specific binding. When anti-CD8 clone 53.6.7 was used, positive staining was observed on CD8 positive cells with both OVA Tetramer and β-gal Tetramer. Even when 53.6.7 was diluted, the cells were stained with both tetramers similarly, suggesting the tetramer staining was not specific.

When anti-CD8 clone KT15 was used, however, CD8 positive cells among OT-I mouse spleen cells were specifically stained only with OVA Tetramer at all antibody concentrations tested. Cells stained with β-gal Tetramer were negative. Titration of KT15 revealed an optimal signal to noise ratio at 0.63 μL/sample for OT-I splenocytes. In summary, when staining cells with H-2K^b tetramers it is important to use appropriately titrated anti-CD8 clone KT15 and include a negative control tetramer in the experiment.

Staining with anti-mouse CD8 (Clone KT15)-FITC/Tetramer -PE
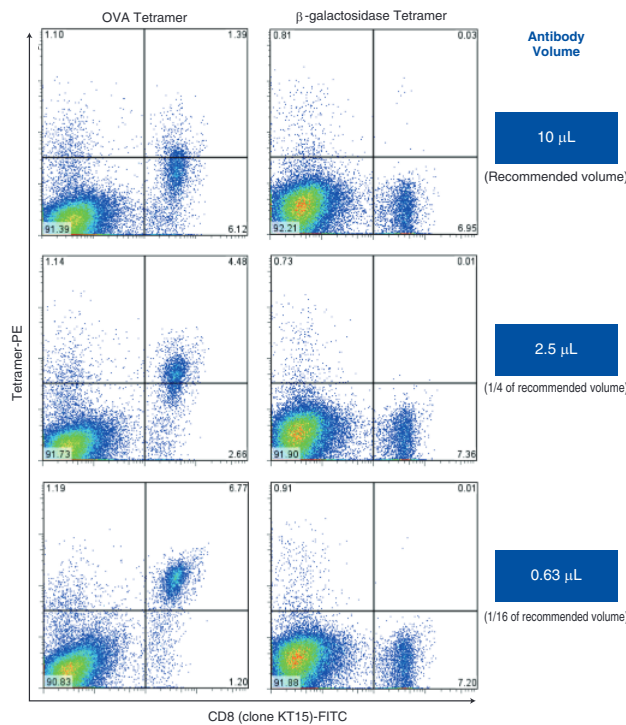
Staining with anti-mouse CD8 (Clone 53.6.7)-FITC/Tetramer -PE
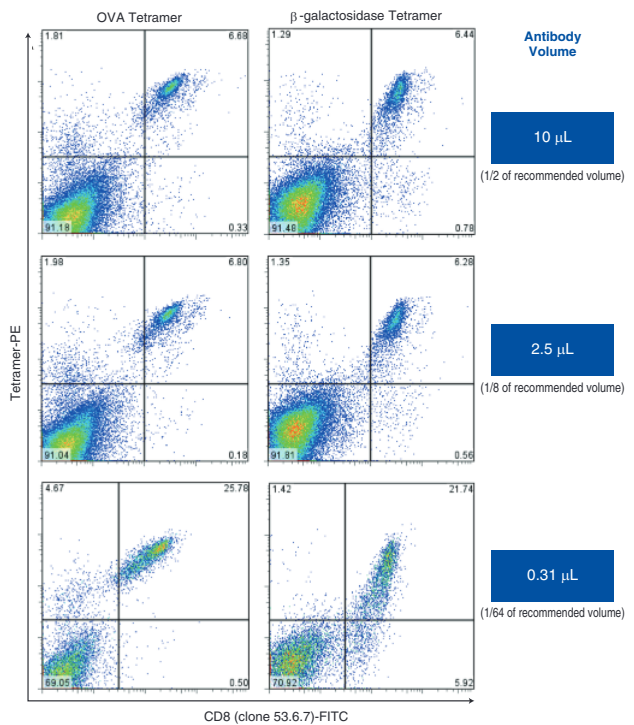
8-c. Effect of CD8 antibody clones on H-2Kb OVA Tetramer staining: "Cells from peptide-immunized mice"

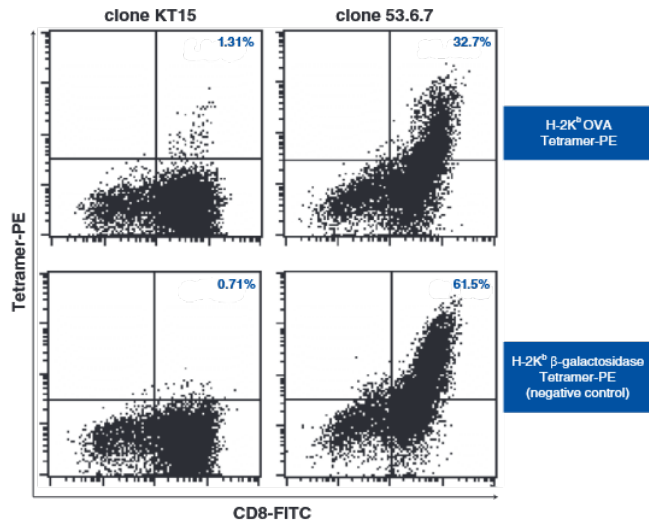Mouse spleen cells prepared using peptide immunization for OVA-specific CTL induction were stained with H-2K^b OVA Tetramer-PE (10 μL/sample) and anti-mouse CD8 (clone KT15 or clone 53.6.7) H-2K^b β-galactosidase Tetramer-PE (10 μL/sample) was used as a negative tetramer to assess non-specific. When KT15 was used, OVA-specific tetramer positive cells were observed compared with the negative tetramer, while when 53.6.7 was used, marked non-specific staining was observed in both H-2K^b β-galactosidase Tetramer samples clone 53.6.7 is known to have poor compatibility with OVA and other H-2K^b Tetramers. MBL recommends the use of KT15 for murine CD8 staining.

clone KT15 | clone 53.6.7 — 1.31% | 32.7% — H-2K^b OVA Tetramer-PE — 0.71% | 61.5% — H-2K^b β-galactosidase Tetramer-PE (negative control). Tetramer-PE (y-axis), CD8-FITC (x-axis).

9. CD1d Tetramer

CD1d is a membrane protein non-covalently bonded to β2-microglobulin (β2m) and shows high homology between human and mice. CD1d can present α-galactosylceramide (α-GalCer), a glycolipid extracted and isolated from the marine sponge, and this complex can activate human and murine CD1d-restricted NKT cells. CD1d Tetramer-PE is a reagent prepared by tetramerization of complexes of CD1d and β2m by PE- or APC- labeled streptavidin. Binding this reagent to α-GalCer enables highly sensitive detection of CD1d-restricted NKT cells and can be combined with antibodies to study NKT cell function by flow cytometry.

<Reference>
1) Stephane S *et al*. J. Immunol. Methods 268, 107–121 (2002)
2) Matsuda JL *et al*. J. Exp. Med. 192, 741–754 (2000)
3) Benlagha K *et al*. J. Exp. Med. 191, 1895–1903 (2000)
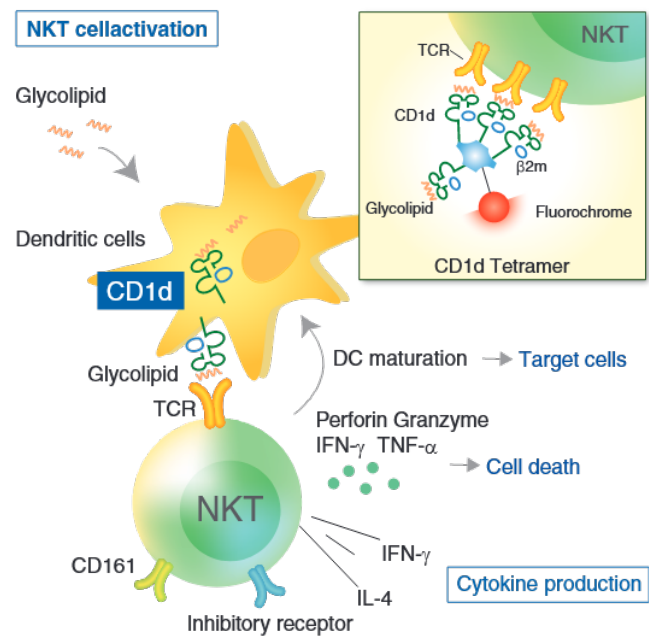
Dissolving and loading of α-GalCer used with CD1d Tetramer

*This product does NOT include α-GalCer.

Pyridine — α-GalCer stock solution (1 mg/mL) → 0.9% NaCl/0.5% Tween-20 α-GalCer diluted solution (200 μg/mL) → Add diluted α-GalCer into CD1d-tetramer-PE → Incubate for 12-18 hr at RT (20-30°C) **or** 37°C, protected from light → Store at 4 °C

Principle of Detection of Antigen-specific T cells by MHC Tetramers

Preparation of class I MHC I Tetramers

High specificity of T-Select HLA class I Tetramers

MHC Tetramer staining method

Induction and detection of antigen specific CTL

Measurement methods for antigen-specific CTL using MBL Kits

MHC Tetramer related products

H-2K^b OVA Tetramer

CD1d Tetramer

QuickSwitch™ Custom Tetramer Kits

MHC Class I custom Tetramer

MHC Class II custom Tetramer

Products List

Principle of Detection of
Antigen-specific T cells
by MHC Tetramers

Preparation of class
MHC I Tetramers

High specificity of
T-Select HLA class I
Tetramers

MHC Tetramer
staining method

Induction and detection
of antigen specific CTL

Measurement methods
for antigen-specific
CTL using MBL Kits

MHC Tetramer
related products

H-2K^b OVA Tetramer

CD1d Tetramer

QuickSwitch™
Custom Tetramer Kits

MHC Class I
custom Tetramer

MHC Class II
custom Tetramer

Products List

Detection of NKT cells with CD1d Tetramer

PBMC were separated from peripheral blood of healthy subjects and incubated at room temperature for 5 minutes with 40 μL of Clear Back (Human Fc receptor blocking reagent, Code No. MTG-001). CD3-FITC and human CD1d Tetramer-PE (with or without binding of α-GalCer) were added and incubated for 30 minutes at 4°C protected from light, and cells were analyzed by flow cytometry. Results showed total cells contained 0.05% NKT cells, as defined by CD1d/CD3 dual positivity.

C57BL/6 mouse splenocytes were stained with CD4-FITC and mouse CD1d Tetramer-PE (with or without binding of α-GalCer) for 30 minutes at 4°C protected from light. Measurement by flow cytometry resulted in detection of NKT cells corresponding to 2.33% of all the cells.
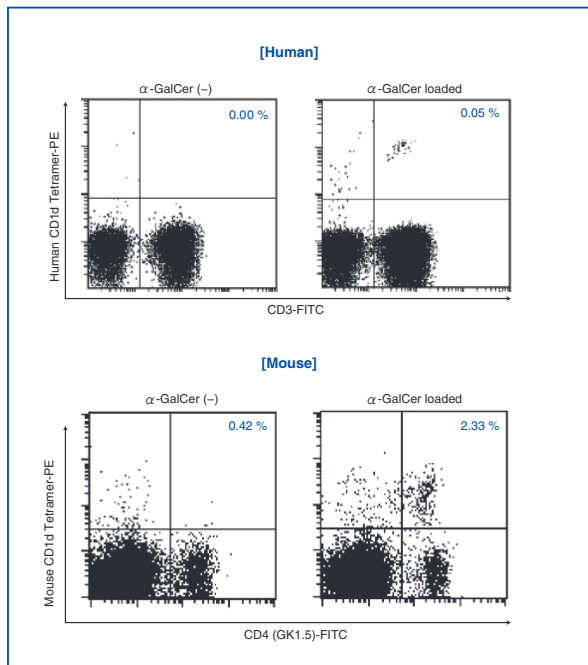
CD1d Tetramer Products

Product name	α-GalCer unloaded	α-GalCer loaded	Size
	Code No.	Code No.	
Human CD1d Tetramer-PE	TS-HCD-1	TS-HCG-1	50 tests
Human CD1d Tetramer-APC	TS-HCD-2	TS-HCG-2	50 tests
Mouse CD1d Tetramer-PE	TS-MCD-1	TS-MCG-1	50 tests
Mouse CD1d Tetramer-APC	TS-MCD-2	TS-MCG-2	50 tests

CD1d Tetramer FAQ

Q1. Can I use DMSO, instead of pyridine, to dissolve α-GalCer?

A1. We recommend using pyridine. In an evaluation at MBL, no positive cells were detected when α-GalCer was dissolved in DMSO and heated at 80°C for 10 seconds before loading the mouse CD1d Tetramer-PE.

Q2. Can I store α-GalCer after dilution to 200 μg/mL?

A2. Do not store α-GalCer diluted to 200 μg/mL because storage reduces the reactivity of the diluted α-GalCer. Instead, store at –20°C as a 1 mg/mL solution in pyridine, and dilute to 200 μg/mL immediately before loading the CD1d Tetramer.
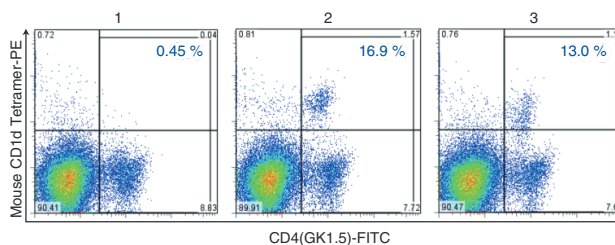
The value in the top right in the figure indicates the percentage of CD1d tetramer positive cells of CD4 positive cells (%).

1. Mouse CD1d Tetramer not loaded with α-GalCer.
2. Mouse CD1d Tetramer loaded with α-GalCer which was diluted to 200 μg/mL on the day before the staining.
3. Mouse CD1d Tetramer loaded on the day before staining with α-GalCer diluted to 200 μg/mL and stored at –30°C for 3 months.

Q3. Can I store CD1d Tetramer at 4°C after it is loaded with α-GalCer?

A3. Yes. Both human and mouse CD1d Tetramers remain reactive for at least 12 months.
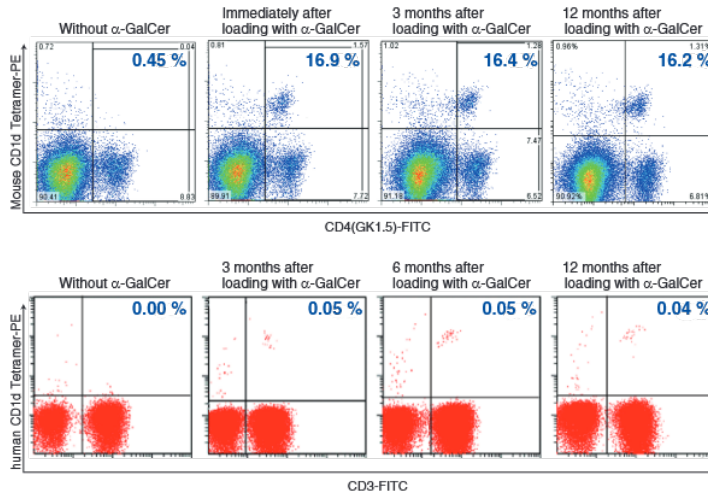
The number in the upper right corner of each panel indicates the percentage of mouse CD1d Tetramer-positive cells that are also CD4-positive.

The number in the upper right corner of each panel indicates the percentage of human CD1d Tetramer-positive cells among all cells.

In all cases, reactivity was comparable to CD1d Tetramer stored at 4°C without α-GalCer, then loaded with α-GalCer on the day before staining.

Q4. What is the expected positive rate of CD1d Tetramer?

A4. Typically, 0.01-0.05% of PBMCs in freshly collected blood from healthy individuals are positive. However, variation between individuals is large, and some individuals may have no positive cells. When performing flow cytometry, we recommend gating on lymphocytes to identify about 100,000 cells for analysis because of the low frequencies. When mouse splenocytes were stained, approximately 5% (BALB/c) and 20% (C57BL/6) of CD4-positive cells were also CD1d Tetramer-positive (test results at MBL).

Q5. Can you recommend a blocking agent for use with CD1d Tetramer?

A5. We recommend Clear Back (Human Fc receptor blocking reagent, Code No. MTG-001). It will prevent non-specific uptake of the tetramer reagents by monocyte-derived cells in both humans and mice.

### Q6. Human and mouse CD1d proteins have a high homology. Do the CD1d Tetramer reagents show species cross-reactivity between human and mouse?

A6. We recommend matching the reagent to the species from which the cells were derived. Staining human PBMC with mouse CD1d Tetramer is not recommended because of high non-specific staining. Staining mouse splenocytes with human CD1d Tetramer instead of mouse CD1d Tetramer results in a lower percentage of positive cells.
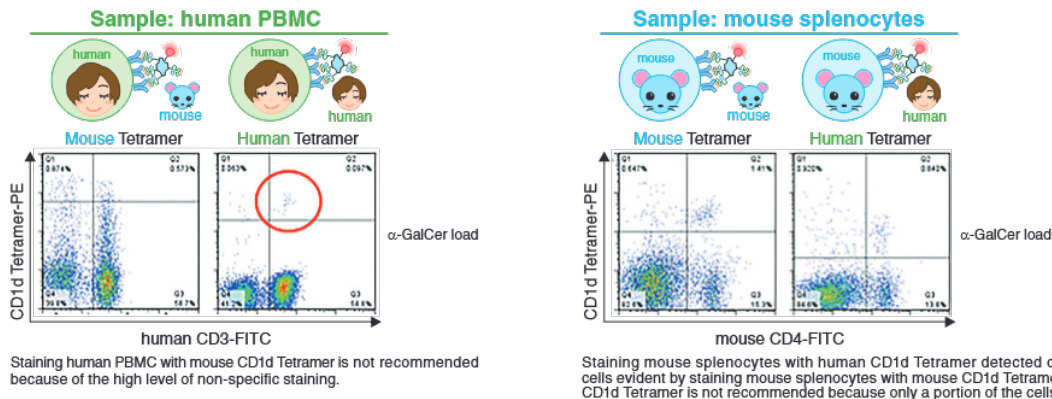
Staining human PBMC with mouse CD1d Tetramer is not recommended because of the high level of non-specific staining.

Staining mouse splenocytes with human CD1d Tetramer detected only 60–70% of NKT cells evident by staining mouse splenocytes with mouse CD1d Tetramer. The use of human CD1d Tetramer is not recommended because only a portion of the cells are detected.

Q7. There is sometimes non-specific staining when CD3-negative cells are stained with human CD1d tetramer without α-GalCer. What should I do?

A7. Human CD1d Tetramer without α-GalCer is known to bind to ILT4 (CD85d) protein expressed in CD14-positive cells. CD14-positive cells can be eliminated by gating out after co-staining them with CD14 antibody. For details, please refer to Li D *et al.* J. Immunol. 182, 1033-1040 (2009) PMID: 19124746.

10. QuickSwitch™ Custom Tetramer Kits

Kits for preparation of custom tetramers in the laboratory using our proprietary peptide exchange technology

◎ **Prepare custom tetramers in 4 hours**
◎ **No UV lamp or special instrument required**
◎ **Quantify peptide exchange efficiency (Quant Tetramer Kits)**
◎ **Select ready-to-use tetramer in PE, APC, or BV421**

Principle of the peptide exchange reaction

Peptide Exchange Factor → Custom peptide → 4 hours at room temperature / dark → Exiting peptide
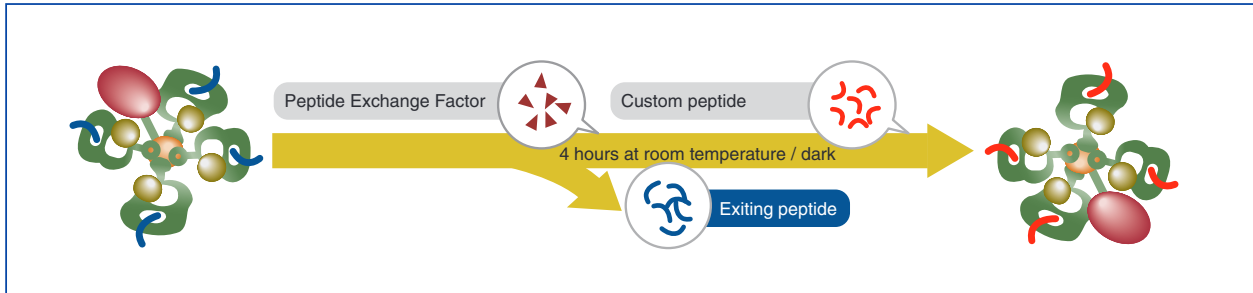
MHC tetramers in QuickSwitch™ Custom Tetramer Kits are pre-bound with "exiting peptide" (shown in blue) to maintain structural integrity. Exchange of the exiting peptide with custom peptide (shown in red) is initiated upon addition of the custom peptide and a peptide-exchange factor (reaction time is 4 hours).

The efficiency of peptide exchange depends on the sequence of the custom peptide. QuickSwitch™ Quant Tetramer Kits contain reagents for determination of the peptide exchange efficiency (see the reverse side for details).

Tetramer preparation and cell staining using QuickSwitch™ Tetramer Kits

■ CTL staining with HLA-A*02:01 Influenza M1 (GILGFVFTL) tetramers

Human PBMCs were stained with tetramer prepared using QuickSwitch™ Tetramer Kit or with MBL's equivalent tetramer product (the number in the upper right corner of each panel indicates the percentage of tetramer-positive cells that are also CD8-positive).

The peptide exchange efficiency of influenza M1 (GILGFVFTL) was 89% (data not shown).

Negative Tetramer — CD3+ Lymphocytes (++ : 0.00%)
Classically folded tetramer Infulenza M1 GILGFVFTL — CD3+ Lymphocytes (++ : 0.28%)
HLA-A*02:01 QuickSwitch™ Infulenza M1 GILGFVFTL — CD3+ Lymphocytes (++ : 0.25%)
Tetramer-PE / CD8 FITC
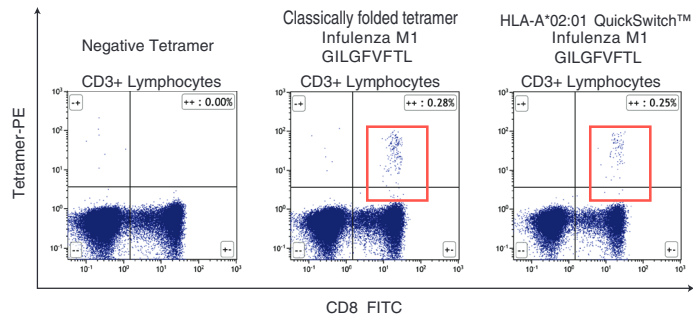
■ CTL staining with H-2K^b OVA (SIINFEKL) tetramers

Spleens were harvested from OT-I TCR transgenic mice, and splenocytes (1.2×10^5 cells/test) were stained with 0.5 or 0.1 μg of tetramer reagents.

Green: H-2K^b TRP-2 Tetramer (negative control)
Red: H-2K^b OVA SIINFEKL Tetramer
(prepared using QuickSwitch™ Tetramer kit)
Blue: H-2K^b OVA SIINFEKL Tetramer
(MBL's equivalent tetramer product)

0.5 μg tetramer/test — 0.1 μg tetramer/test
Tetramer-APC / CD8 FITC
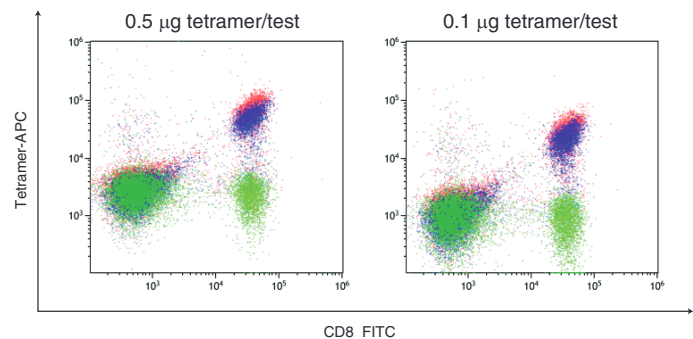
See page 26 for the list of QuickSwitch™ Custom Tetramer Kit.

Principle of Detection of Antigen-specific T cells by MHC Tetramers
Preparation of class MHC I Tetramers
High specificity of T-Select HLA class I Tetramers
MHC Tetramer staining method
Induction and detection of antigen specific CTL
Measurement methods for antigen-specific CTL using MBL kits
MHC Tetramer related products
H-2K^b OVA Tetramer
CD1d Tetramer
QuickSwitch™ Custom Tetramer Kits
MHC Class I custom Tetramer
MHC Class II custom Tetramer
Products List

11. MHC Class I custom Tetramer

Available alleles

HLA-A	HLA-B	HLA-C
A*01:01	B*07:02	Cw*01:02
A*02:01	B*08:01	Cw*03:03
A*02:06	B*15:01	Cw*03:04
A*02:07	B*15:02	Cw*04:01
A*03:01	B*27:05	Cw*06:02
A*11:01	B*35:01	Cw*08:01
A*23:01	B*40:01	Cw*12:02
A*24:02	B*40:06	Cw*15:02
A*26:01	B*42:01	
A*29:02	B*52:01	**HLA-E**
A*31:01	B*54:01	E*01:01
	B*57:01	E*01:03

Mouse
H-2K^b
H-2K^d
H-2D^b
H-2D^d
H-2D^k
H-2L^d
H-2K^K
A2K^b (chimera)
A24K^b (chimera)
Qa-1b

Chicken
BF2*1201
BF2*1501

Rhesus Macaque
Mamu-A*90120-5
Mamu-A*01

Macaca Fascicularis
Mafa-A1*063
Mafa-B*104:01

Steps for custom MHC class I tetramer

Folding

MHC class I heavy chain, β2m, and a peptide are folded to generate a MHC class I/peptide complex (monomer).

Monomer

Biotinylation

The MHC class I heavy chain is biotinylated and the monomers are pulified by column chromatography.

d-Biotin

Tetramarization

Tetramerization using fluorescent labeled Streptavidin

Fluorescent labeled Streptavidin

MHC Tetramer

12. MHC Class II custom Tetramer

Available alleles

Human Class II	
DRB1*01:01	DRB1*11:01
DRB1*03:01	DRB1*12:02
DRB1*04:01	DRB1*14:01
DRB1*04:05	DRB1*14:06
DRB1*04:10	DRB1*15:01
DRB1*07:01	DRB1*15:02
DRB1*08:02	DRB4*01:01
DRB1*08:03	DPB1*04:01
DRB1*09:01	

Mouse Class II
I-A^d
I-A^b
I-A^{g7}

Steps for custom Human HLA MHC class II tetramer

α-chain   β-chain

Folding

α-chain, β-chain, and a peptide are folded to generate a MHC class II/peptide complex (monomer).

Peptide

Biotinylation

d-Biotin

Tetramarization

Tetramerization using fluorescent labeled Streptavidin

Fluorescent labeled Streptavidin
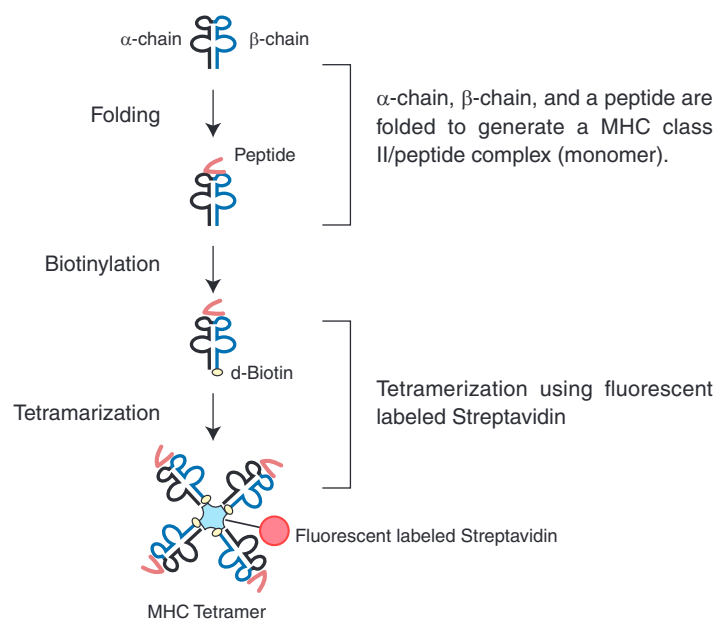
MHC Tetramer

We can manufacture a custom MHC tetramer based on your needs.
Contact us with the allele and peptide sequence which you wish to order.
Any questions according to our MHC tetramers are welcome.

✉ **Contact information: support@mbl.co.jp**

Principle of Detection of Antigen-specific T cells by MHC Tetramers

Preparation of class I MHC I Tetramers

High specificity of T-Select HLA class I Tetramers

MHC Tetramer staining method

Induction and detection of antigen specific CTL

Measurement methods for antigen-specific CTL using MBL Kits

MHC Tetramer related products

H-2K^b OVA Tetramer

CD1d Tetramer

QuickSwitch™ Custom Tetramer Kits

MHC Class I custom Tetramer

MHC Class II custom Tetramer

Products List

13. Products List

Human class I Tetramer

Antigen	MHC Allele	Sequence	Location (aa)	PE-labeled (50 tests)	APC-labeled (50 tests)	BV421-labeled (50 tests)	FITC-labeled (50 tests)	Peptide
Cancer (Human)								
ACC-1	A*24:02	DYLQYVLQI	15-23	TS-M141-1	TS-M141-2			
AIM-2	A*01:01	RSDSGQQARY	-	TS-M137-1	TS-M137-2			
BA46	A*02:01	GLQHWVPEL	97-106	TB-0151-1	TB-0151-2	TB-0151-4		
BCR-ABL	A*02:01	GVRGRVEEI	bcr-abl fusion	TB-0169-1	TB-0169-2	TB-0169-4		
CA9	A*24:02	EYRALQLHL	219-227	TS-M112-1	TS-M112-2			
CD20	A*02:01	SLFLGILSV	188-196	TB-0158-1	TB-0158-2	TB-0158-4		
CD33	A*02:01	AIISGDSPV	65-73	TS-M101-1	TS-M101-2			
CD33 A65Y	A*02:01	YIISGDSPV	65-73	TS-M102-1	TS-M102-2			
CEA	A*02:01	YLSGANLNL	605-613	TS-M103-1	TS-M103-2			
CEA N6D	A*02:01	YLSGADLNL	605-613	TS-M080-1	TS-M080-2			
EphA2	A*02:01	TLADFDPRV	883-891	TS-M084-1	TS-M084-2			
EZH2	A*02:01	YMCSFLFNL	666-674	TB-0164-1	TB-0164-2	TB-0164-4		
gp100	A*02:01	KTWGQYWQV	154-162	TB-0035-1	TB-0035-2	TB-0035-4		
gp100	A*02:01	YLEPGPVTA	280-288	TB-M082-1	TB-M082-2			
gp100	A*02:01	YLEPGPVTV	280-288	TB-0120-1	TB-0120-2	TB-0120-4		
gp100	A*03:01	ALLAVGATK	17-25	TB-0166-1	TB-0166-2	TB-0166-4		
gp100 (wild)	A*02:01	ITDQVPFSV	209-217	TS-0014-1C	TB-0014-2	TB-0014-4		
gp100 (mutant)	A*02:01	IMDQVPFSV	209-217	TS-0013-1C	TB-0013-2	TB-0013-4		
gp100-intron 4	A*24:02	VYFFLPDHL	170-178	TS-M089-1	TS-M089-2			
GPC3	A*02:01	FVGEFFTDV	144-152	TB-0134-1	TB-0134-2	TB-0134-4		
GPC3	A*24:02	EYILSLEEL	298-306	TB-0140-1	TB-0140-2	TB-0140-4		
Her-2/neu	A*02:01	RLLQETELV	689-697	TB-0016-1	TB-0016-2	TB-0016-4		
Her-2/neu E75	A*02:01	KIFGSLAFL	369-377	TB-0015-1	TB-0015-2	TB-0015-4		
HM1.24	A*02:01	KLQDASAEV	126-134	TS-M083-1	TS-M083-2			
hTERT	A*02:01	ILAKFLHWL	540-548	TS-M115-1	TS-M115-2			
hTERT	A*02:01	ALLTSRLRFI	615-624	TB-0128-1	TB-0128-2	TB-0128-4		
hTERT	A*02:01	GLLGASVLGL	674-683	TB-0150-1	TB-0150-2	TB-0150-4		
hTERT	A*02:01	YLFFYRKSV	572-580	TB-0113-1	TB-0113-2	TB-0113-4		
hTERT	A*02:01	YLQVNSLQTV	988-997	TB-0114-1	TB-0114-2	TB-0114-4		
hTERT	A*03:01	KLFGVLRLK	973-981	TB-0105-1	TB-0105-2	TB-0105-4		
hTERT	A*24:02	VYGFVRACL	461-469	TS-M010-1				
LIVIN	A*02:01	QLCPICRAPV	175-184	TB-0156-1	TB-0156-2	TB-0156-4		
LY6K	A*24:02	RYCNLEGPPI	177-186	TB-0167-1	TB-0167-2	TB-0167-4		
MAGE-A1	A*01:01	EADPTGHSY	161-169	TS-M114-1	TS-M114-2			
MAGE-A1	A*02:01	KVLEYVIKV	278-286	TB-M070-1	TB-M070-2	TB-M070-4		
MAGE-A1	B*07:02	RVRFFFPSL	289-297	TS-M071-1	TS-M071-2			
MAGE-A10	A*02:01	GLYDGMEHL	254-262	TS-M078-1	TS-M078-2			
MAGE-A2	A*02:01	YLQLVFGIEV	157-166	TB-M072-1	TB-M072-2			
MAGE-A2	A*24:02	EYLQLVFGI	156-164	TS-M073-1	TS-M073-2			
MAGE-A3	A*01:01	EVDPIGHLY	168-176	TB-M074-1	TB-M074-2	TB-M074-4		
MAGE-A3	A*02:01	KVAELVHFL	112-120	TB-M075-1	TB-M075-2			
MAGE-A3	A*02:01	FLWGPRALV	271-279	TB-M076-1	TB-M076-2			
MAGE-A3	A*24:02	IMPKAGLLI	195-203	TS-M077-1	TS-M077-2			
MAGE-C1	A*02:01	ILFGISLREV	959-968	TS-M138-1	TS-M138-2			
Mart-1	A*02:01	ELAGIGILTV	26-35	TB-0009-1	TB-0009-2	TB-0009-4		TS-0009-P
MCPyV large T Ag	A*24:02	EWWRSGGFSF	92-101	TS-M091-1	TS-M091-2			
Mesothelin	A*02:01	SLLFLLFSL	20-28	TB-0110-1	TB-0110-2	TB-0110-4		
Mesothelin	A*02:01	VLPLTVAEV	530-538	TB-0112-1	TB-0112-2	TB-0112-4		
MUC1	A*02:01	LLLLTVLTV	12-20	TB-M088-1	TB-M088-2			TS-M088-P
MUC1	A*02:01	LLLTVLTVV	13-21	TB-0153-1	TB-0153-2	TB-0153-4		
NY-ESO-1	A*02:01	SLLMWITQC	157-165	TB-M011-1	TB-M011-2	TB-M011-4		TS-M011-P
NY-ESO-1 C9V	A*02:01	SLLMWITQV	157-165	TB-M105-1	TB-M105-2	TB-M105-4		
NY-ESO-1	B*35:01	MPFATPMEA	94-102	TB-0129-1	TB-0129-2	TB-0129-4		
P2X5	B*07:02	TPNQRQNVC	110-118	TS-M109-1	TS-M109-2			
p53	A*02:01	LLGRNSFEV	264-272	TS-M081-1	TS-M081-2			
p53	A*02:01	KLCPVQLWV	139-147	TB-0152-1	TB-0152-2	TB-0152-4		
p53	A*02:01	GLAPPQHLIRV	187-197	TB-0136-1	TB-0136-2	TB-0136-4		
p53	A*02:01	SLPPPGTRV	149-157	TB-0159-1	TB-0159-2	TB-0159-4		
p53	A*02:01	RMPEAAPPV	65-73	TB-0157-1	TB-0157-2	TB-0157-4		

Antigen	MHC Allele	Sequence	Location (aa)	PE-labeled (50 tests)	APC-labeled (50 tests)	BV421-labeled (50 tests)	FITC-labeled (50 tests)	Peptide
				Code No.				
p53	A*02:01	YLGSYGFRL	103-111	TB-0163-1	TB-0163-2	TB-0163-4		
PAP	A*02:01	ALDVYNGLL	299-307	TB-M107-1	TB-M107-2			
PP2A	A*02:01	SLLPAIVEL	402-410	TS-M095-1	TS-M095-2			
PRAME	A*02:01	ALYVDSLFFL	300-309	TS-M116-1	TS-M116-2			
PRAME	A*02:01	VLDGLDVLL	100-108	TS-M117-1	TS-M117-2			
PRAME	A*02:01	SLLQHLIGL	425-433	TS-M118-1	TS-M118-2			
PRAME	A*02:01	SLYSFPEPEA	142-151	TS-M119-1	TS-M119-2			
Proteinase 3 (PR-1)	A*02:01	VLQELNVTV	169-177	TB-0017-1	TB-0017-2	TB-0017-4		
PSA	A*02:01	FLTPKKLQCV	141-150	TS-M120-1	TB-M120-2			
PSA	A*02:01	KLQCVDLHV	146-154	TS-M087-1	TS-M087-2			
PSA	A*24:02	CYASGWGSI	153-161	TB-0139-1	TB-0139-2	TB-0139-4		
PSCA	A*02:01	AILALLPAL	105-113	TB-0127-1	TB-0127-2	TB-0127-4		
PSMA	A*02:01	VLAGGFFLL	27-38	TB-0161-1	TB-0161-2	TB-0161-4		
RHAMM	A*02:01	ILSLELMKL	165-173	TS-M104-1	TS-M104-2			
SSX-2	A*02:01	KASEKIFYV	41-49	TS-M079-1	TS-M079-2			
Survivin T2M	A*02:01	LMLGEFLKL	96-104	TS-M085-1	TS-M085-2			
Survivin	A*02:01	LTLGEFLKL	96-104	TB-0155-1	TB-0155-2	TB-0155-4		
Survivin (modK)	A*03:01	RISTFKNWPK	18-27	TB-0108-1	TB-0108-2	TB-0108-4		
Survivin-2B	A*24:02	AYACNTSTL	80-88	TS-M025-1	TS-M025-2			TS-M025-P
topo II	A*02:01	FLYDDNQRV	828-836	TB-0132-1	TB-0132-2	TB-0132-4		
Tyrosinase	A*02:01	YMDGTMSQV	368-376	TS-0019-1C	TS-0019-2C	TB-0019-4		
Tyrosinase	A*24:02	AFLPWHRLF	206-214	TS-M090-1	TS-M090-2			
Tyrosinase	A*01:01	KSDICTDEY	243-251	TB-0126-1	TB-0126-2	TB-0126-4		
Tyrosinase	B*07:02	LPWHRLFLL	208-216	TB-0119-1	TB-0119-2	TB-0119-4		
WT1	A*02:01	RMFPNAPYL	126-134	TS-M016-1	TS-M016-2	TB-M016-4		
WT1	A*02:01	VLDFAPPGA	37-45	TS-M140-1	TS-M140-2			TS-M140-P
modified WT1	A*24:02	CYTWNQMNL	235-243	TS-M014-1	TS-M014-2	TB-M014-4		
Virus and Bacteria (Human)								
AdV 11 Hexon	A*02:01	YLLFEVFDV	913-921	TS-M058-1	TS-M058-2			
AdV 11 Hexon	A*02:01	LLFEVFDVV	914-922	TB-M059-1	TB-M059-2			
AdV 11 Hexon	A*24:02	TYFNLGNKF	37-45	TS-M062-1	TS-M062-2			
AdV 11 Hexon	A*24:02	VYSGSIPYL	696-704	TS-M064-1	TS-M064-2			
AdV 5 Hexon	A*24:02	TYFSLNNKF	37-45	TS-M063-1	TS-M063-2			
AdV Hexon	A*02:01	YVLFEVFDV	917-925	TS-M061-1	TS-M061-2			
AdV Hexon	B*07:02	KPYSGTAYNSL	114-124	TS-M065-1	TS-M065-2			
AdV Hexon	B*07:02	KPYSGTAYNAL	114-124	TS-M066-1	TS-M066-2			
AdV Hexon	B*35:01	MPNRPNYIAF	320-329	TS-M067-1	TB-M067-2			
AdV Hexon	B*35:01	IPYLDGTFY	705-713	TS-M068-1	TB-M068-2			
EBV BALF4	A*02:01	FLDKGTYTL	276-284	TB-0131-1	TB-0131-2	TB-0131-4		
EBV BMLF1	A*02:01	GLCTLVAML	280-288	TB-0011-1	TB-0011-2	TB-0011-4		TS-0011-P
EBV BMLF1	A*24:02	DYNFVKQLF	320-328	TS-M003-1				TS-M003-P
EBV BRLF1	A*24:02	TYPVLEEMF	198-206	TS-M002-1				TS-M002-P
EBV BRLF1	A*03:01	RVRAYTYSK	148-156	TS-M124-1	TS-M124-2			
EBV BZLF1	B*08:01	RAKFKQLL	190-197	TS-M036-1	TS-M036-2			
EBV BZLF1	B*35:01	EPLPQGQLTAY	54-64	TB-M037-1	TB-M037-2			
EBV EBNA1	B*35:01	HPVGEADYFEY	407-417	TB-0168-1	TB-0168-2	TB-0168-4		
EBV EBNA1	C*03:03	FVYGGSKTSL	508-517	TS-M150-1	TS-M150-2			
EBV EBNA3A	A*03:01	RLRAEAQVK	603-611	TS-M033-1	TS-M033-2			
EBV EBNA3A	A*24:02	RYSIFFDYM	246-254	TS-M004-1				TS-M004-P
EBV EBNA3A	B*07:02	RPPIFIRRL	379-387	TS-M142-1	TS-M142-2			
EBV EBNA3A	B*08:01	FLRGRAYGL	325-333	TS-M123-1	TS-M123-2			
EBV EBNA3B	A*11:01	AVFDRKSDAK	399-408	TS-M028-1				
EBV EBNA3B	A*11:01	IVTDFSVIK	416-424	TS-M029-1				
EBV EBNA3B	A*24:02	TYSAGIVQI	217-225	TS-M005-1				TS-M005-P
EBV EBNA4	B*15:01	GQGGSPTAM	831-839	TB-0101-1	TB-0101-2	TB-0101-4		
EBV EBNA6	B*07:02	QPRAPIRPI	881-889	TB-0123-1	TB-0123-2	TB-0123-4		
EBV LMP1	A*02:01	YLQQNWWTL	159-167	TS-M006-1				
EBV LMP1	A*02:01	YLLEMLWRL	125-133	TB-0146-1	TB-0146-2	TB-0146-4		
EBV LMP2	A*02:01	TVCGGIMFL	243-251	TS-M030-1	TS-M030-2			
EBV LMP2	A*02:01	LLWTLVVLL	329-337	TS-M031-1	TS-M031-2			
EBV LMP2	A*02:01	CLGGLLTMV	426-434	TS-M032-1	TS-M032-2			
EBV LMP2	A*02:01	FLYALALLL	356-364	TS-M069-1	TS-M069-2			TS-M069-P
EBV LMP2	A*11:01	SSCSSCPLSK	340-349	TS-M111-1	TS-M111-2			TS-M111-P

Principle of Detection of Antigen-specific T cells by MHC Tetramers

Preparation of class I MHC Tetramers

High specificity of T-Select HLA class I Tetramers

MHC Tetramer staining method

Induction and detection of antigen specific CTL

Measurement methods for antigen-specific CTL using MBL Kits

MHC Tetramer related products

H-2K^b OVA Tetramer

CD1d Tetramer

QuickSwitch™ Custom Tetramer Kits

MHC Class I custom Tetramer

MHC Class II custom Tetramer

Products List

Antigen	MHC Allele	Sequence	Location (aa)	Code No. PE-labeled (50 tests)	APC-labeled (50 tests)	BV421-labeled (50 tests)	FITC-labeled (50 tests)	Peptide
EBV LMP2 S9T	A*11:01	SSCSSCPLTK	340-349	TS-M135-1	TS-M135-2			TS-M135-P
EBV LMP2	A*24:02	IYVLVMLVL	222-230	TS-M001-1	TS-M001-2			TS-M001-P
EBV LMP2	A*24:02	PYLFWLAAI	131-139	TS-M034-1	TS-M034-2			
EBV LMP2	A*24:02	TYGPVFMSL	419-427	TS-M035-1	TS-M035-2			TS-M035-P
EBV LMP2	A*24:02	TYGPVFMCL	419-427	TS-M154-1	TS-M154-2	TB-0117-4		
EBV LMP2	B*35:01	MGSLEMVPM	1-9	TB-M038-1	TB-M038-2			
EBV LMP2 (mutant)	A*02:01	SLGGLLTMV	426-434	TS-M155-1	TS-M155-2			
EBV LMP2 (mutant)	A*02:01	CLGGLITMV	426-434	TS-M156-1	TS-M156-2			
EBV LMP2 (mutant)	A*02:01	FLCALALLL	356-364	TS-M157-1	TS-M157-2			
EBV LMP2	A*02:01	QLSPLLGAV	264-272	TS-M159-1	TS-M159-2			
EBV LMP2	A*02:01	GLGTLGAAL	293-301	TS-M160-1	TS-M160-2			
EBV LMP2 (mutant)	A*02:01	TVCGGMMFL	243-251	TS-M161-1	TS-M161-2			
EBV Mix	A*24:02	–	-	TS-M009-1				
HBV core	A*02:01	FLPSDFFPSV	18-27	TB-0018-1	TB-0018-2	TB-0018-4		TS-0018-P
HBV core	A*24:02	EYLVSFGVW	117-125	TB-0022-1	TB-0022-2	TB-0022-4		TS-0022-P
HBV env	A*02:01	WLSLLVPFV	335-343	TB-M051-1	TB-M051-2			
HBV env	A*02:01	GLSPTVWLSV	348-357	TS-M052-1	TS-M052-2			
HBV pol	A*02:01	FLLSLGIHL	575-583	TB-M053-1	TB-M053-2			
HBV pol	A*24:02	KYTSFPWLL	756-764	TS-0023-1C	TS-0023-2C	TB-0023-4		
HBV S protein	A*02:01	FLLTRILTI	183-191	TB-0122-1	TB-0122-2	TB-0122-4		
HCMV IE1	A*02:01	VLEETSVML	316-324	TB-M057-1	TB-M057-2			
HCMV IE1	A*03:01	KLGGALQAK	184-192	TS-M100-1	TS-M100-2			
HCMV IE1	B*08:01	ELRRKMMYM	199-207	TS-0026-1C	TS-0026-2C	TB-0026-4		
HCMV IE1	B*08:01	QIKVRVDMV	88-96	TB-0147-1	TB-0147-2	TB-0147-4		
HCMV pp50	A*01:01	VTEHDTLLY	245-253	TS-0024-1C	TS-0024-2C	TB-0024-4		
HCMV pp65	A*02:01	NLVPMVATV	495-503	TS-0010-1C	TS-0010-2C	TB-0010-4		TS-0010-P
HCMV pp65	A*11:01	ATVQGQNLK	501-509	TS-M012-1				TS-M012-P
HCMV pp65	A*24:02	QYDPVAALF	341-349	TS-0020-1C	TS-0020-2C	TB-0020-4		TS-0020-P
HCMV pp65	B*07:02	RPHERNGFTVL	265-275	TB-M099-1	TB-M099-2			
HCMV pp65	B*07:02	TPRVTGGGAM	417-426	TS-0025-1C	TB-0025-2	TB-0025-4		
HCMV pp65	B*15:01	KMQVIGDQY	215-223	TS-M013-1				
HCMV pp65	B*35:01	IPSINVHHY	123-131	TB-0027-1	TB-0027-2	TB-0027-4		
HCV E2	A*24:02	EYVLLLFLL	717-725	TS-M044-1	TS-M044-2			
HCV NS3	A*02:01	CINGVCWTV	1073-1081	TB-M039-1	TB-M039-2	TB-M039-4		
HCV NS3	A*02:01	KLVALGINAV	1406-1415	TB-M040-1	TB-M040-2			TS-M040-P
HCV NS3	A*02:01	CVNGVCWTV	1073-1081	TB-0118-1	TB-0118-2	TB-0118-4		
HCV NS3	B*08:01	HSKKKCDEL	1395-1403	TB-0145-1	TB-0145-2	TB-0145-4		
HCV NS4B	A*02:01	VLSDFKTWL	1992-2000	TS-M041-1	TS-M041-2			
HCV NS5B	A*02:01	ALYDVVTKL	2594-2602	TS-M042-1	TS-M042-2			
HCV NS5B	A*02:01	ALYDVVSKL	2594-2602	TS-M043-1	TS-M043-2			
HCV polyprotein	A*01:01	ATDALMTGY	1436-1444	TB-0125-1	TB-0125-2	TB-0125-4		
HHV-6B U54	A*02:01	ILYGPLTRI	129-137	TS-M143-1	TS-M143-2			
HHV-6B U54	A*24:02	PFHCSFHTI	267-275	TS-M164-1	TS-M164-2			
HHV-8 gB	A*02:01	LMWYELSKI	492-500	TB-0154-1	TB-0154-2	TB-0154-4		
HIV env gp160	A*24:02	RYLRDQQLL	584-592	TS-M007-1	TS-M007-2		TS-M007-3	TS-M007-P
HIV env	A*24:02	RYLKDQQLL	67-75	TB-0130-1	TB-0130-2	TB-0130-4		
HIV env	B*27:05	GRAFVTIGK	103-111	TB-0148-1	TB-0148-2	TB-0148-4		
HIV gag	A*02:01	SLYNTVATL	77-85	TS-M027-1	TS-M027-2	TB-M027-4	TS-M027-3	TS-M027-P
HIV gag	A*02:01	TLNAWVKVV	19-27	TS-M139-1	TS-M139-2			
HIV p17 gag	A*03:01	RLRPGGKKK	20-28	TB-0109-1	TB-0109-2	TB-0109-4		
HIV gag	B*27:05	KRWIILGLNK	265-274	TB-0124-1	TB-0124-2	TB-0124-4		
HIV gag	B*27:05	KRWIIMGLN	265-273	TB-0106-1	TB-0106-2	TB-0106-4		
HIV gag	B*07:02	GPGHKARVL	223-231	TB-0142-1	TB-0142-2	TB-0142-4		
HIV gag	B*57:01	ISPRTLNAW	147-155	TB-0104-1	TB-0104-2	TB-0104-4		
HIV nef	A*24:02	RYPLTFGW	134-141	TS-M110-1	TS-M110-2			
HIV nef	A*03:01	QVPLRPMTYK	73-82	TB-0138-1	TB-0138-2	TB-0138-4		
HIV nef	B*07:02	TPGPGVRYPL	128-137	TS-M054-1	TS-M054-2			
HIV nef	B*35:01	VPLRPMTY	74-81	TB-M106-1	TB-M106-2			
HIV pol	A*02:01	ILKEPVHGV	476-484	TS-0008-1C	TB-0008-2	TB-0008-4		
HIV RT	B*35:01	NPDIVIYQY	175-183	TB-M055-1	TS-M055-2			
HIV RT	B*35:01	VPLDEDFRKY	273-282	TB-0149-1	TB-0149-2	TB-0149-4		
HIV Vif	A*02:01	GLADQLIHL	101-109	TB-0135-1	TB-0135-2	TB-0135-4		
HPV16 E6	A*02:01	KLPQLCTEL	18-26	TB-M047-1	TB-M047-2			

Antigen	MHC Allele	Sequence	Location (aa)	Code No.				
				PE-labeled (50 tests)	APC-labeled (50 tests)	BV421-labeled (50 tests)	FITC-labeled (50 tests)	Peptide
HPV16 E6	A*24:02	VYDFAFRDL	49-57	TS-M049-1	TS-M049-2			
HPV16 E6	A*03:01	KLCLRFLSK	64-72	TB-0137-1	TB-0137-2	TB-0137-4		
HPV16 E7	A*02:01	YMLDLQPET	11-19	TB-0031-1	TB-0031-2	TB-0031-4		
HPV16 E7	A*02:01	YMLDLQPETT	11-20	TS-M048-1	TS-M048-2			TS-M048-P
HPV16 E7	A*02:01	MLDLQPETT	12-20	TB-0173-1	TB-0173-2	TB-0173-4		
HPV16 E7	B*07:02	KPTLKEYVL	5-13	TB-0143-1	TB-0143-2	TB-0143-4		
HTLV bZIP	A*02:01	AVLDGLLSL	42-50	TB-0133-1	TB-0133-2	TB-0133-4		
HTLV-1 Env	A*24:02	FFQFCPLIF	11-19	TS-M022-1				
HTLV-1 Tax	A*02:01	LLFGYPVYV	11-19	TS-M017-1	TS-M017-2	TB-M017-4		TS-M017-P
HTLV-1 Tax	A*02:01	QLGAFLTNV	178-186	TS-M019-1				
HTLV-1 Tax	A*11:01	KVLTPPITH	88-96	TS-M023-1				
HTLV-1 Tax	A*11:01	QSSSFIFHK	272-280	TS-M024-1				
HTLV-1 Tax	A*24:02	LFGYPVYVF	12-20	TS-M020-1				
HTLV-1 Tax	A*24:02	PYKRIEELL	187-195	TS-M021-1				
HTLV-1 Tax	A*24:02	SFHSLHLLF	301-309	TS-M018-1	TS-M018-2			TS-M018-P
Influenza M1	A*02:01	GILGFVFTL	58-66	TS-0012-1C	TS-0012-2C	TB-0012-4		TS-0012-P
Influenza M1	A*02:01	ILGFVFTLTV	59-68	TS-M162-1	TS-M162-2	TB-0165-4		
Influenza NP	A*01:01	CTELKLSDY	44-52	TS-M045-1	TS-M045-2			
Influenza NP	A*03:01	ILRGSVAHK	265-273	TB-0103-1	TB-0103-2	TB-0103-4		
Influenza NP	B*27:05	SRYWAIRTR	383-391	TB-0111-1	TB-0111-2	TB-0111-4		
Influenza NP	B*35:01	LPFEKSTVM	418-426	TB-M046-1	TB-M046-2			
Influenza NS1	A*02:01	AIMDKNIIL	122-130	TS-M163-1	TS-M163-2			
Influenza PA	A*24:02	YYLEKANKI	130-138	TS-M144-1	TS-M144-2			
Influenza PB1	A*24:02	SYLIRALTL	216-224	TS-M145-1	TS-M145-2			
Influenza PB1	A*24:02	RYTKTTYWW	430-438	TS-M146-1	TS-M146-2			
Influenza PB1	A*24:02	SYINRTGTF	482-490	TS-M147-1	TS-M147-2			
Influenza PB1	A*24:02	RYGFVANF	498-505	TS-M148-1	TS-M148-2			
Influenza PB2	A*24:02	TYQWIIRNW	549-557	TS-M149-1	TS-M149-2			
LCMV GPC	A*02:01	YLVSIFLHL	447-455	TB-0116-1	TB-0116-2	TB-0116-4		
LCMV ND	A*02:01	ALPHIIDEV	10-18	TB-0121-1	TB-0121-2	TB-0121-4		
M. tuberculosis 16 kDa	A*02:01	GILTVSVAV	120-128	TS-M132-1	TS-M132-2			
M. tuberculosis 19 kDa	A*02:01	VLTDGNPPEV	88-97	TS-M133-1	TS-M133-2			
M. tuberculosis Ag85A	A*02:01	GLPVEYLQV	48-56	TS-M128-1	TS-M128-2			
M. tuberculosis Ag85A	A*02:01	KLIANNTRV	242-250	TS-M129-1	TS-M129-2			
M. tuberculosis Ag85B	A*02:01	KLVANNTRL	199-207	TS-M131-1	TS-M131-2			
M. tuberculosis Ag85C	A*02:01	WPTLIGLAM	204-212	TS-M134-1	TS-M134-2			
M. tuberculosis ESAT-6	A*02:01	AMASTEGNV	82-90	TS-M125-1	TS-M125-2			
M. tuberculosis Hsp65	A*02:01	KLQERLAKL	362-370	TS-M130-1	TS-M130-2			
M. tuberculosis MPT51	A*02:01	TLAGKGISVV	53-62	TS-M026-1				TS-M026-P
M. tuberculosis Rv1614	A*02:01	FLYELIWNV	197-205	TS-M127-1	TS-M127-2			
Measles virus HA	A*02:01	KLWCRHFCV	576-584	TS-M092-1	TS-M092-2			
RSV matrix protein	A*01:01	YLEKESIYY	229-237	TS-M056-1	TS-M056-2			
VZV IE62	A*02:01	ALWALPHAA	593-601	TB-M122-1	TB-M122-2			

Others (Human)

Antigen	MHC Allele	Sequence	Location (aa)	PE-labeled (50 tests)	APC-labeled (50 tests)	BV421-labeled (50 tests)	FITC-labeled (50 tests)	Peptide
BTG1	A*02:01	TLWVDPYEV	103-111	TS-M097-1	TS-M097-2			
H-Y	A*02:01	FIDSYICQV	311-319	TS-M094-1	TS-M094-2			
H-Y	B*07:02	SPSVDKARAEL	-	TB-0144-1	TB-0144-2	TB-0144-4		
HA-1	A*02:01	VLHDDLLEA	137-145	TS-M093-1	TS-M093-2			
HA-2	A*02:01	YIGEVLVSV	41-49	TS-M108-1	TS-M108-2			
HA-8	A*02:01	RTLDKVLEV	149-157	TS-M098-1	TS-M098-2			
Insulin B	A*02:01	HLVEALYLV	10-18	TB-0102-1	TB-0102-2	TB-0102-4		
IAPP	A*02:01	KLQVFLIVL	5-13	TB-0170-1	TB-0170-2	TB-0170-4		
IGRP	A*02:01	VLFGLGFAI	265-273	TB-0162-1	TB-0162-2	TB-0162-4		
IGRP	A*02:01	LNIDLLWSV	228-236	TB-0107-1	TB-0107-2	TB-0107-4		
RNA-dependent helicase	A*02:01	YLLPAIVHI	148-156	TS-M096-1	TS-M096-2			

Negative Tetramer (Human)

Antigen	MHC Allele	Sequence	Location (aa)	PE-labeled (50 tests)	APC-labeled (50 tests)	BV421-labeled (50 tests)	FITC-labeled (50 tests)	Peptide
HIV env gp160	A*24:02	RYLRDQQLL	584-592	TS-M007-1	TS-M007-2		TS-M007-3	TS-M007-P
Negative	A*02:01	-	-	TB-0029-1	TB-0029-2	TB-0029-4		TS-0029-P
Negative	A*02:01	ALAAAAAAV	-	TS-M151-1	TS-M151-2		TS-M151-3	TS-M151-P
Negative	A*11:01	ATAAAAAAK	-	TS-M152-1	TS-M152-2		TS-M152-3	TS-M152-P
Negative	A*24:02	AYAAAAAAL	-	TS-M153-1	TS-M153-2		TS-M153-3	TS-M153-P

Principle of Detection of Antigen-specific T cells by MHC Tetramers
Preparation of class I MHC Tetramers
High specificity of T-Select HLA class I Tetramers
MHC Tetramer staining method
Induction and detection of antigen specific CTL
Measurement methods for antigen-specific CTL using MBL Kits
MHC Tetramer related products
H-2K^b OVA Tetramer
CD1d Tetramer
QuickSwitch™ Custom Tetramer Kits
MHC Class I custom Tetramer
MHC Class II custom Tetramer
Products List

Human class II Tetramer

Antigen	MHC Allele	Sequence	Location (aa)	PE-labeled (20 tests)	APC-labeled (20 tests)	BV421-labeled (20 tests)	FITC-labeled (20 tests)	Peptide
Virus, Bacteria and Allergen								
Bet v 1	DRB1*15:01	ETLLRAVESYLLAHS	142-156	TS-M818-1	TS-M818-2			TS-M818-P
EBV EBNA1	DRB1*01:01	TSLYNLRRGTALA	515-527	TS-M803-1	TS-M803-2			TS-M803-P
Fel d 1	DRB1*01:01	LPVVLENARILKNCVDAK	49-66	TS-M813-1	TS-M813-2			TS-M813-P
GAD65	DRB1*04:01	NFFRMVISNPAAT	555-567	TS-M811-1	TS-M811-2			TS-M811-P
HIV gag	DRB1*01:01	DYVDRFYKTLRAE	295-307	TS-M802-1	TS-M802-2			TS-M802-P
HTLV-1 Tax	DRB1*01:01	YLYQLSPPITWPL	155-167	TS-M815-1	TS-M815-2			TS-M815-P
Influenza HA	DRB1*01:01	PKYVKQNTLKLAT	306-318	TS-M804-1	TS-M804-2			TS-M804-P
Influenza HA	DRB1*04:05	PKYVKQNTLKLAT	306-318	TS-M806-1	TS-M806-2			
Influenza HA	DRB1*04:01	PKYVKQNTLKLAT	306-318	TS-M810-1	TS-M810-2			
Influenza HA	DRB1*11:01	PKYVKQNTLKLAT	306-318	TS-M808-1	TS-M808-2			
Lol p 1	DRB1*04:01	APYHFDLSGHAFG	105-117	TS-M814-1	TS-M814-2			TS-M814-P
Tetanus Toxin	DRB1*11:01	MQYIKANSKFIGITEL	829-844	TS-M812-1	TS-M812-2			TS-M812-P
Negative Tetramer (Human)								
CLIP	DRB1*01:01	PVSKMRMATPLLMQA	103-117	TS-M801-1	TS-M801-2			TS-M801-P
CLIP	DRB1*04:01	PVSKMRMATPLLMQA	103-117	TS-M809-1	TS-M809-2			
CLIP	DRB1*04:05	PVSKMRMATPLLMQA	103-117	TS-M805-1	TS-M805-2			
CLIP	DRB1*11:01	PVSKMRMATPLLMQA	103-117	TS-M807-1	TS-M807-2			
CLIP	DRB1*15:01	PVSKMRMATPLLMQA	103-117	TS-M816-1	TS-M816-2			
CLIP	DRB1*15:02	PVSKMRMATPLLMQA	103-117	TS-M817-1	TS-M817-2			

Principle of Detection of Antigen-specific T cells by MHC Tetramers

Preparation of class II MHC I Tetramers

High specificity of T-Select HLA class I Tetramers

MHC Tetramer staining method

Induction and detection of antigen specific CTL

Measurement methods for antigen-specific CTL using MBL Kits

MHC Tetramer related products

H-2Kᵇ OVA Tetramer

CD1d Tetramer

QuickSwitch™ Custom Tetramer Kits

MHC Class I custom Tetramer

MHC Class II custom Tetramer

Products List

Mouse class I Tetramer

Antigen	MHC Allele	Sequence	Location (aa)	PE-labeled (50 tests)	APC-labeled (50 tests)	BV421-labeled (50 tests)	FITC-labeled (50 tests)	Peptide
Cancer (Mouse)								
Adpgk Neoepitope	H-2D^b	ASMTNMELM	-	TB-5113-1	TB-5113-2	TB-5113-4		
CEA	H-2D^b	EAQNTTYL	526-533	TS-M518-1				TS-M518-P
Copine-1 Neoepitope	H-2D^b	SSPYSLHYL	-	TB-5115-1	TB-5115-2	TB-5115-4		
Erk2 K136Q	H-2K^d	QYIHSANVL	136-144	TS-M545-1	TS-M545-2			
gp100 (human)	H-2D^b	KVPRNQDWL	25-33	TS-M505-1	TS-M505-2	TB-M505-4		TS-M505-P
gp100 (mouse)	H-2D^b	EGSRNQDWL	25-33	TS-M546-1	TS-M546-2			
HER2/neu	H-2K^d	TYLPTNASL	63-71	TS-M526-1				TS-M526-P
JAK1	H-2K^d	SYFPEITHI	367-375	TS-M544-1	TS-M544-2			
MAGE-A3	H-2K^d	SYVKVLHHM	-	TB-5105-1	TB-5105-2	TB-5105-4		
MAGE-A5	H-2K^b	HNTQYCNL	5-12	TS-M559-1	TS-M559-2			
MAGE-AX	H-2K^b	LGITYDGM	169-176	TS-M558-1	TS-M558-2			
P815A P1A	H-2L^d	LPYLGWLVF	35-43	TS-M519-1				TS-M519-P
pBM1	H-2K^b	INFDFNTI	207-214	TS-M563-1	TS-M563-2			
PSA(human)	H-2D^b	HCIRNKSVIL	65-74	TS-M561-1	TS-M561-2			
REPS1 Neoepitope	H-2D^b	AQLANDVVL	-	TB-5114-1	TB-5114-2	TB-5114-4		
Survivin	H-2K^b	MFFCFKEL	-	TB-5102-1	TB-5102-2	TB-5102-4		
Survivin	H-2D^b	ATFKNWPFL	20-28	TB-5108-1	TB-5108-2	TB-5108-4		
TERT(mouse)	H-2K^b	VGRNFTNL	198-205	TB-M562-1	TB-M562-2			
Tnpo3	H-2K^d	SYMLQALCI	-	TB-5104-1	TB-5104-2	TB-5104-4		
TRP-2	H-2K^b	SVYDFFVWL	180-188	TB-5004-1	TB-5004-2	TB-5004-4		TS-5004-P
WT1	H-2D^b	RMFPNAPYL	126-134	TS-M504-1	TS-M504-2			
Virus (Mouse)								
Adv5 E1A	H-2D^b	SGPSNTPPEI	234-243	TS-M564-1	TS-M564-2			
HBsAg	H-2L^d	IPQSLDSWWTSL	28-39	TS-M522-1				TS-M522-P
HBV core	H-2K^b	MGLKFRQL	93-100	TB-M537-1	TB-M537-2			
HBV HBsAg	H-2K^b	VWLSVIWM	190-197	TB-5110-1	TB-5110-2	TB-5110-4		
HCV NS3	H-2D^b	GAVQNEVTL	1629-1637	TS-M532-1	TS-M532-2			
HIV env	H-2D^d	IGPGRAFYA	315-323	TB-M536-1	TS-M536-2			
HIV gag	H-2K^d	AMQMLKETI	197-205	TB-5007-1	TB-5007-2	TB-5007-4		
HIV P18-I10	H-2D^d	RGPGRAFVTI	318-327	TS-M516-1				TS-M516-P
HPV16 E7	H-2D^b	RAHYNIVTF	49-57	TB-5008-1	TB-5008-2	TB-5008-4		TS-5008-P
HSV-1 gB	H-2K^b	SSIEFARL	498-505	TS-M523-1				TS-M523-P
HTLV-1 Tax	H-2D^k	ARLHRHALL	38-46	TS-M531-1				TS-M531-P
Influenza HA	H-2K^d	IYSTVASSL	533-541	TS-M520-1	TS-M520-2			TS-M520-P
Influenza HA	H-2K^d	LYQNVGTYV	204-212	TS-M535-1	TS-M535-2			
Influenza NP	H-2K^d	TYQRTRALV	147-155	TB-M534-1	TB-M534-2			TS-M534-P
Influenza NP	H-2D^b	ASNENMDAM	366-374	TS-M527-1	TS-M527-2			TS-M527-P
Influenza NP	H-2D^b	ASNENMDTM	366-374	TS-M502-1	TS-M502-2			TS-M502-P
Influenza NP	H-2D^b	ASNENMETM	366-374	TS-M508-1	TS-M508-2			TS-M508-P
Influenza NS2	H-2K^b	RTFSFQLI	114-121	TS-M566-1	TS-M566-2			
Influenza PA	H-2D^b	SSLENFRAYV	224-233	TS-M528-1	TS-M528-2			TS-M528-P
Influenza PB1	H-2K^b	SSYRRPVGI	703-711	TS-M533-1	TS-M533-2			
LCMV gp	H-2K^b	ISHNFCNL	118-125	TB-5012-1	TB-5012-2	TB-5012-4		
LCMV gp	H-2D^b	SGVENPGGYCL	276-286	TB-5009-1	TB-5009-2	TB-5009-4		
LCMV gp 33	H-2D^b	KAVYNFATC	33-41	TB-5002-1	TS-5002-2C	TB-5002-4		TS-5002-P
LCMV gp 33 C9M	H-2D^b	KAVYNFATM	33-41	TS-M512-1	TS-M512-2			TS-M512-P
LCMV gp	H-2K^b	AVYNFATC	34-41	TB-5010-1	TB-5010-2	TB-5010-4		
LCMV gp	H-2K^b	AVYNFATCGI	34-43	TB-5011-1	TB-5011-2	TB-5011-4		
LCMV L protein	H-2K^b	LEYDFNKL	825-832	TB-5014-1	TB-5014-2	TB-5014-4		
LCMV NP	H-2L^d	RPQASGVYM	118-126	TS-M514-1				TS-M514-P
LCMV NP	H-2K^b	YTVKYPNL	205-212	TB-5015-1	TB-5015-2	TB-5015-4		
LCMV NP	H-2D^b	FQPQNGQFI	396-404	TS-M513-1	TS-M513-2	TB-M513-4		TS-M513-P
MCMV IE1	H-2L^d	YPHFMPTNL	168-176	TS-M510-1				TS-M510-P
MCMV M45	H-2D^b	HGIRNASFI	985-993	TB-5109-1	TB-5109-2	TB-5109-4		
MCMV M164	H-2D^d	AGPPRYSRI	257-265	TB-5111-1	TB-5111-2	TB-5111-4		
MHV(MuHV-4) M2	H-2K^d	GFNKLRSTL	91-99	TS-M568-1	TS-M568-2			
MHV S protein	H-2D^b	CSLWNGPHL	510-518	TB-5101-1	TB-5101-2	TB-5101-4		
MoMSV	H-2D^b	(Abu)(Abu)L(Abu)LTVFL	85-93	TB-5016-1	TB-5016-2	TB-5016-4		
MuLV gp70	H-2L^d	SPSYVYHQF	423-431	TS-M521-1	TS-M521-2			TS-M521-P
MuLV p15E	H-2K^b	KSPWFTTL	604-611	TS-M507-1	TS-M507-2			TS-M507-P
Polyomavirus MT	H-2D^k	RRLGRTLLL	389-397	TS-M530-1				TS-M530-P

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MHC Tetramer related products
H-2K^b OVA Tetramer
CD1d Tetramer
QuickSwitch™ Custom Tetramer Kits
MHC Class I custom Tetramer
MHC Class II custom Tetramer
Products List

Antigen	MHC Allele	Sequence	Location (aa)	Code No.				
				PE-labeled (50 tests)	APC-labeled (50 tests)	BV421-labeled (50 tests)	FITC-labeled (50 tests)	Peptide
RSV F glycoprotein	H-2K^d	KYKNAVTEL	85-93	TS-M555-1	TS-M555-2			
RSV M protein	H-2D^b	NAITNAKII	187-195	TB-5018-1	TB-5018-2	TB-5018-4		
RSV M2 protein	H-2K^d	SYIGSINNI	82-90	TS-M506-1	TS-M506-2	TB-M506-4		TS-M506-P
RSV M2 protein	H-2K^d	SYIGINNI	-	TS-M567-1	TS-M567-2			
SeV NP	H-2D^b	FAPGNYPAL	324-332	TS-M509-1				TS-M509-P
SIV gag	H-2D^b	AAVKNWMTQTL	312-322	TB-5017-1	TB-5017-2	TB-5017-4		
SV40 large T Ag	H-2D^b	SAINNYAQKL	206-215	TB-M539-1	TB-M539-2	TB-M539-4		
SV40 large T Ag	H-2D^b	QGINNLDNL	489-497	TS-M540-1	TS-M540-2			
VACV B8R	H-2K^b	TSYKFESV	20-27	TB-M538-1	TB-M538-2			
VSV NP	H-2K^b	RGYVYQGL	52-59	TS-M529-1				TS-M529-P
Others (Mouse)								
BCG MPT51	H-2D^d	GGPHAVYLL	24-32	TS-M517-1				TS-M517-P
β-galactosidase	H-2L^d	TPHPARIGL	876-884	TS-M511-1	TS-M511-2			TS-M511-P
β-galactosidase	H-2K^b	DAPIYTNV	96-103	TS-M501-1	TS-M501-2			TS-M501-P
CD94/NKG2 Qdm	Qa-1b	AMAPRTLLL	-	TB-5106-1	TB-5106-2	TB-5106-4		
Chlamydia CrpA	H-2D^b	ASFVNPIYL	63-71	TS-M548-1	TS-M548-2			
EGFP	H-2K^d	HYLSTQSAL	200-208	TS-M525-1				TS-M525-P
HA-60	H-2K^b	LTFNYRNL	39-46	TS-M551-1	TS-M551-2			
HY Uty	H-2D^b	WMHHNMDLI	246-254	TS-M524-1	TB-M524-2	TB-M524-4		TS-M524-P
IGRP	H-2K^d	VYLKTNVFL	206-214	TB-M552-1	TB-M552-2			
InsB	H-2K^d	LYLVCGERL	15-23	TS-M554-1	TS-M554-2			
Listeria LLO	H-2K^d	GYKDGNEYI	91-99	TS-M503-1				TS-M503-P
Malaria (P. berghei) CSP	H-2K^d	SYIPSAEKI	252-260	TS-M515-1				TS-M515-P
Malaria (P. yoelii) CSP	H-2K^d	SYVPSAEQI	280-288	TB-M547-1	TB-M547-2			
M. tuberculosis 32a	H-2D^b	GAPINSATAM	309-318	TS-M549-1	TS-M549-2			
MimA2	H-2D^b	YAIENYLEL	-	TS-M557-1	TS-M557-2			
MTB Ag85A	H-2L^d	MPVGGQSSF	70-78	TB-5112-1	TB-5112-2	TB-5112-4		
NRP-V7	H-2K^d	KYNKANVFL	-	TB-M553-1	TB-M553-2			
OVA	H-2K^b	SIINFEKL	257-264	TS-5001-1C	TS-5001-2C	TB-5001-4		TS-5001-P
OVA E1	H-2K^b	EIINFEKL	257-264	TS-M541-1	TS-M541-2			
OVA G4	H-2K^b	SIIGFEKL	257-264	TS-M542-1	TS-M542-2			
OVA Q4H7	H-2K^b	SIIQFEHL	257-264	TS-M543-1	TS-M543-2			
TB10.4	H-2K^b	IMYNYPAM	4-11	TS-M550-1	TS-M550-2			
Toxoplasma gondii	H-2K^b	SVLAFRRL	-	TB-5103-1	TB-5103-2	TB-5103-4		
Trypanosoma cruzi trans-sialidase	H-2K^b	ANYKFTLV	380-387	TS-M560-1	TS-M560-2			
SIY (Negative)	H-2K^b	SIYRYYGL	designed peptide	TS-M008-1	TS-M008-2			TS-M008-P

Mouse class II Tetramer

Antigen	MHC Allele	Sequence	Location (aa)	Code No.				
				PE-labeled (20 tests)	APC-labeled (20 tests)	BV421-labeled (20 tests)	FITC-labeled (20 tests)	Peptide
BDC2.5 mimotope	I-A^{g7}	AHHPIWARMDA	-	TS-M727-1	TS-M727-2			TS-M727-P
chicken HEL	I-A^{g7}	AMKRHGLDNYRGYSL	11-25	TS-M718-1	TS-M718-2			TS-M718-P
Eα	I-A^b	ASFEAQGALANIAVDKA	52-68	TS-M706-1				
ESAT-6	I-A^b	MTEQQWNFAGIEAAASAIQG	1-20	TS-M707-1				TS-M707-P
FMLV	I-A^b	EPLTSLTPRCNTAWNRLKL	123-141	TS-M705-1				
human CLIP	I-A^b	PVSKMRMATPLLMQA	103-117	TS-M715-1	TS-M715-2			
human CLIP	I-A^d	PVSKMRMATPLLMQA	103-117	TS-M720-1	TS-M720-2			
human CLIP	I-A^{g7}	PVSKMRMATPLLMQA	103-117	TS-M717-1	TS-M717-2			
Influenza NP	I-A^b	QVYSLIRPNENPAHK	311-325	TS-M716-1	TS-M716-2			TS-M716-P
MOG	I-A^b	MEVGWYRSPFSRVVHLYRNGK	35-55	TS-M704-1	TS-M704-2			TS-M704-P
mouse 2W1S	I-A^b	EAWGALANWAVDSA	52-68	TS-M722-1	TS-M722-2			TS-M722-P
Mtb Ag85B	I-A^b	FQDAYNAAGGHNAVF	240-254	TS-M719-1	TS-M719-2			
OVA	I-A^b	ISQAVHAAHAEINEAGR	323-339	TS-M710-1	TS-M710-2			TS-M703-P
OVA	I-A^d	ISQAVHAAHAEINEAGR	323-339	TS-M703-1	TS-M703-2			
T. gondii CD4Ag28m	I-A^b	AVEIHRPVPGTAPPS	605-619	TS-M723-1	TS-M723-2			TS-M723-P

HLA-E Tetramer

Antigen	MHC Allele	Sequence	Location (aa)	Code No.				
				PE-labeled (50 tests)	APC-labeled (50 tests)	BV421-labeled (50 tests)	FITC-labeled (50 tests)	Peptide
HLA-A*02, A*24 leader	HLA-E*01:03	VMAPRTLVL	3-11	TS-ME01-1				
HLA-A*02, A*24 leader Negative	HLA-E*01:03	VMAPKTLVL	3-11	TS-ME02-1				
HLA-A*02, A*24 leader	HLA-E*01:01	VMAPRTLVL	3-11	TS-ME03-1				
HLA-A*02, A*24 leader Negative	HLA-E*01:01	VMAPKTLVL	3-11	TS-ME04-1				

Other species

Antigen	MHC Allele	Sequence	Location (aa)	Code No.				
				PE-labeled (50 tests)	APC-labeled (50 tests)	BV421-labeled (50 tests)	FITC-labeled (50 tests)	Peptide
Monkey								
SIV gag	Mamu-A*90120-5	SSVDEQIQW	241-249	TS-M901-1	TS-M901-2			
SIV gag	Mamu-A*01	CTPYDINQM	181-189	TB-5003-1	TB-5003-2	TB-5003-4		
SIV gag GW9	Mafa-A1*063	GPRKPIKCW	386-394	TB-5022-1	TB-5022-2	TB-5022-4		
SIV gag NA9	Mafa-B*104:01	NCVGDHQAA	192-200	TB-5024-1	TB-5024-2	TB-5024-4		
SIV nef RM9	Mafa-A1*063	RPKVPLRTM	103-111	TB-5021-1	TB-5021-2	TB-5021-4		
SIV nef HW8	Mafa-A1*063	HPAQTSQW	196-203	TB-5023-1	TB-5023-2	TB-5023-4		
SIV nef LT9	Mafa-B*104:01	LNMADKKET	254-262	TB-5025-1	TB-5025-2	TB-5025-4		
Chicken								
IBDV VP2	BF2*1201	ALRPVTLV	338-345	TS-M951-1	TS-M951-2			
IBV NP	BF2*1501	WRRQARYK	71-78	TS-M952-1	TS-M952-2			

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MHC Tetramer related products

H-2Kb OVA Tetramer

CD1d Tetramer

QuickSwitch™ Custom Tetramer Kits

MHC Class I custom Tetramer

MHC Class II custom Tetramer

Products List

CD1d Tetramer

Code No.	Product name	Size
TS-HCD-1	Human CD1d Tetramer-PE	50 tests
TS-HCD-2	Human CD1d Tetramer-APC	50 tests
TS-MCD-1	Mouse CD1d Tetramer-PE	50 tests
TS-MCD-2	Mouse CD1d Tetramer-APC	50 tests
TS-HCG-1	Human CD1d Tetramer (α-GalCer loaded)-PE	50 tests
TS-HCG-2	Human CD1d Tetramer (α-GalCer loaded)-APC	50 tests
TS-MCG-1	Mouse CD1d Tetramer (α-GalCer loaded)-PE	50 tests
TS-MCG-2	Mouse CD1d Tetramer (α-GalCer loaded)-APC	50 tests

QuickSwitch™

*QuickSwitch™ Quant Tetramer Kits contain reagents for the determination of peptide exchange efficiency.

Code No.	Product name	Size
TB-7300-K1	QuickSwitch™ Quant HLA-A*02:01 Tetramer Kit-PE	25 µg
TB-7300-K2	QuickSwitch™ Quant HLA-A*02:01 Tetramer Kit-APC	25 µg
TB-7300-K4	QuickSwitch™ Quant HLA-A*02:01 Tetramer Kit-BV421	25 µg
TB-7304-K1	QuickSwitch™ Quant HLA-A*11:01 Tetramer Kit-PE	25 µg
TB-7304-K2	QuickSwitch™ Quant HLA-A*11:01 Tetramer Kit-APC	25 µg
TB-7304-K4	QuickSwitch™ Quant HLA-A*11:01 Tetramer Kit-BV421	25 µg
TB-7302-K1	QuickSwitch™ Quant HLA-A*24:02 Tetramer Kit-PE	25 µg
TB-7302-K2	QuickSwitch™ Quant HLA-A*24:02 Tetramer Kit-APC	25 µg
TB-7302-K4	QuickSwitch™ Quant HLA-A*24:02 Tetramer Kit-BV421	25 µg
TB-7400-K1	QuickSwitch™ Quant H-2Kb Tetramer Kit-PE	25 µg
TB-7400-K2	QuickSwitch™ Quant H-2Kb Tetramer Kit-APC	25 µg
TB-7400-K4	QuickSwitch™ Quant H-2Kb Tetramer Kit-BV421	25 µg

*QuickSwitch™ Tetramer Kits do not include these reagents.

Code No.	Product name	Size
TB-7301-K1	QuickSwitch™ HLA-A*02:01 Tetramer Kit-PE	25 µg
TB-7301-K2	QuickSwitch™ HLA-A*02:01 Tetramer Kit-APC	25 µg
TB-7301-K4	QuickSwitch™ HLA-A*02:01 Tetramer Kit-BV421	25 µg
TB-7303-K1	QuickSwitch™ HLA-A*24:02 Tetramer Kit-PE	25 µg
TB-7303-K2	QuickSwitch™ HLA-A*24:02 Tetramer Kit-APC	25 µg
TB-7303-K4	QuickSwitch™ HLA-A*24:02 Tetramer Kit-BV421	25 µg
TB-7401-K1	QuickSwitch™ H-2Kb Tetramer Kit-PE	25 µg
TB-7401-K2	QuickSwitch™ H-2Kb Tetramer Kit-APC	25 µg
TB-7401-K4	QuickSwitch™ H-2Kb Tetramer Kit-BV421	25 µg

When using the standard protocol, each kit is sufficient for custom tetramers for 10 different peptide sequences. Each peptide sequence requires approximately 2.5 µg of tetramer molecules. The amount of custom tetramers to use for staining T cells in PBMCs needs to be determined for each peptide sequence.

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MHC Tetramer related products

H-2Kb OVA Tetramer

CD1d Tetramer

QuickSwitch™ Custom Tetramer Kits

MHC Class I custom Tetramer

MHC Class II custom Tetramer

Products List

Related products

Kits and Reagents

Code No.	Product name	Size
4844	IMMUNOCYTO CD107a Detection Kit	50 tests
AM-1005M	IMMUNOCYTO Cytotoxicity Detection Kit	50 tests
MTG-001	Clear Back (Human Fc receptor blocking reagent)	1 mL (50 tests) × 2

Mouse Helper Peptides

Code No.	Array	Immunogen (Antigen)	Location (aa)	MHC Allele	Size
TS-M701-P	TPPAYRPPNAPIL	HBc	128-140	I-A^b	100 μL (10 mg/ mL)
TS-M702-P	FNNFTVSFWLRVPKVSASHLE	TT p30	947-967	I-A^d	100 μL (10 mg/ mL)
TS-M703-P	ISQAVHAAHAEINEAGR	OVA	323-339	I-A^b, I-A^d	100 μL (10 mg/ mL)
TS-M704-P	MEVGWYRSPFSRVVHLYRNGK	MOG	35-55	I-A^b	100 μL (10 mg/ mL)
TS-M707-P	MTEQQWNFAGIEAAASAIQG	ESAT-6	1-20	I-A^b	100 μL (10 mg/ mL)
TS-M708-P	DGSTDYGILQINSRW	HEL	48-62	I-A^k	100 μL (10 mg/ mL)

HLA Antibodies

Code No.	Product name	Clone	Isotype	Application	Size
D367-3	Anti-HLA class I (HLA-A,B,C) (Human) mAb	EMR8-5	Mouse IgG1 κ	WB, FCM, IH	100 μg/100 μL
D370-3H	Anti-HLA class I (HLA-A,B,C) (Human) mAb	EMR8-5.1	Mouse IgG1 κ	IH	6 mL
K0186-3	Anti-HLA-A2 (Human) mAb	BB7.2	Mouse IgG2b	IP, FCM, IH	100 μg/100 μL
K0186-4	Anti-HLA-A2 (Human) mAb-FITC	BB7.2	Mouse IgG2b	FCM	50 μg/100 μL
K0186-5	Anti-HLA-A2 (Human) mAb-PE	BB7.2	Mouse IgG2b	FCM	1 mL (50 tests)
K0208-3	Anti-HLA-A24 (Human) mAb	17A10	Mouse IgG2b	FCM, IH*	100 μg/100 μL
K0208-4	Anti-HLA-A24 (Human) mAb-FITC	17A10	Mouse IgG2b	FCM	50 μg/100 μL
K0208-5	Anti-HLA-A24 (Human) mAb-PE	17A10	Mouse IgG2b	FCM	1 mL (50 tests)
K0208-A48	Anti-HLA-A24 (Human) mAb-Alexa Fluor ® 488	17A10	Mouse IgG2b	FCM	100 μg/100 μL
K0208-A64	Anti-HLA-A24 (Human) mAb-Alexa Fluor ® 647	17A10	Mouse IgG2b	FCM	100 μg/100 μL
K0209-3	Anti-HLA-A24 (Human) mAb	22E1	Mouse IgG2b	FCM	100 μg/100 μL
K0209-4	Anti-HLA-A24 (Human) mAb-FITC	22E1	Mouse IgG2b	FCM	50 μg/100 μL
K0209-5	Anti-HLA-A24 (Human) mAb-PE	22E1	Mouse IgG2b	FCM	1 mL (50 tests)
K0215-3	Anti-HLA-E (Human) mAb	4D12	Mouse IgG1	FCM, IH*	100 μg/100 μL
K0126-3	Anti-HLA-E (Human) mAb	MEM-E/02	Mouse IgG1	WB, IH	100 μg

CD8 Antibodies

Code No.	Product name	Clone	Isotype	Application	Size
K0226-4	Anti-CD8 (Human) mAb-FITC	Hit8a	Mouse IgG1 κ	FCM	1 mL (100 tests)
D271-4	Anti-CD8 (Mouse) mAb-FITC	KT15	Rat IgG2aλ	FCM	1 mL (100 tests)
D271-5	Anti-CD8 (Mouse) mAb-PE	KT15	Rat IgG2aλ	FCM	1 mL (100 tests)
D271-A64	Anti-CD8 (Mouse) mAb-Alexa Fluor ® 647	KT15	Rat IgG2aλ	FCM	50 μg/50 μL

WB: Western Blotting, IP: Immunoprecipitation, FCM, Flow Cytometry, IH: Immunohistochemistry
*: reported in articles (not confirmed by MBL)

Principle of Detection of Antigen-specific T cells by MHC Tetramers

Preparation of class I MHC Tetramers

High specificity of T-Select HLA class I Tetramers

MHC Tetramer staining method

Induction and detection of antigen specific CTL

Measurement methods for antigen-specific CTL using MBL Kits

MHC Tetramer related products

H-2K^b OVA Tetramer

CD1d Tetramer

QuickSwitch™ Custom Tetramer Kits

MHC Class I custom Tetramer

MHC Class II custom Tetramer

Products List

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