# High Quality MHC and TCR Monomers for Immunological Research

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# Introduction

Production of critical components such as peptide-major histocompatibility complexes and T-cell receptors (TCRs) with correct folding is challenging due to their complex structure and low-affinity ligands. By relying on more than 10 years' experience in protein expression, our team of experts has set up an effective manufacturing process with stringent quality control, providing you with high-quality pMHC and TCR Monomers. This poster describes the quality control checks used to demonstrate the production of high-quality pMHC and TCR constructs and explores the associated applications for MHC and TCR Monomers within the dynamically evolving field of immunology.

# High Quality Ready-to-Use Monomers

A broad range of ready-to-use MHC and TCR Monomers are available, both with or without biotin. MHC I and II Monomers are available both ready-to-use, and loadable (see peptide-receptive, below).

iNKT Cells

MHC I and MHC II Monomers

CD8<sup>+</sup> and CD4<sup>+</sup> T Cells

HLA B\*07:02

✓ HLA B\*07:02

h<sup>β</sup>2m-chain

α-chain

Immudex

Ladder Monomer

50

30

25 20

MHC Monomers: Immudex Quality Control

**MR1** Monomers

MAIT Cells

• Analysis of the performance of starting materials by

Analysis of peptide-MHC (pMHC) complex integrity

RPHERNGFTVL, cat. no. M-WH02135) run on an SDS-

PAGE Gel under reducing conditions. The high purity of

flow cytometry - MHC heavy chain and β2

using a fluorescence and binding assay.

Analysis of peptide-MHC complex purity and

biotinylation using an SDS gel-shift assay.

the MHC Monomer is evidenced by absence of

Left: 1µg of MHC Monomer (HLA B\*07:02 /

microglobulin (B2M).

degradation products.

**CD1d Monomers** 

**HLA-E & HLA-G Monomers** 

NK Cells, Tregs and more

TCR Dextramer<sup>®</sup> PE

FUNCTIONALITY AND SPECIFICITY ASSAY

**TCR Monomers** 











TCR

Biotin

For investigation of Antigen-Presenting Cells

- SDS-PAGE gel confirms TCR alpha and beta chains are present at the correct sizes (23 and 29 kDa, respectively).
- Testing functionality and specificity of TCR by generating TCR Dextramer<sup>®</sup> reagents and analyzing them by flow cytometry.
- Orange peak = negative control pMHC (irrelevant pMHC that should not interact with the TCR Dextramer<sup>®</sup>)
- Blue peak = the shift indicates the binding of the TCR Dextramer<sup>®</sup> to its cognate pMHC

# Peptide-Receptive MHC Monomers

easYmers<sup>®</sup> and U-Load<sup>®</sup> MHC II are formulations of peptide-receptive MHC I and II monomers, which can be used to generate specific pMHC complexes by loading your peptide of choice.

# Flexibility to Assemble Your Own MHC Multimers

easYmers<sup>®</sup> or U-Load<sup>®</sup> MHC II-peptide complexes can easily be loaded onto fluorescentlylabeled U-Load Dextramer<sup>®</sup>, U-Load dCODE Dextramer<sup>®</sup>, or other lab-developed MHC multimers, for detection of antigen-specific CD8<sup>+</sup> or CD4<sup>+</sup> T cells using flow cytometry<sup>1</sup> or single-cell multi-omics<sup>2</sup>. 

# 1,2K . 100 90 80 900 -Count 009 70 40 α-chain β-chain 300 25 **20** -10

SDS-PAGE

# **TCR Monomers: Immudex Quality Control**





# Broadest Allele Coverage

To satisfy the diverse research needs of our customers, our list of MHC alleles is constantly expanding.

Human	Primate	Mouse
64 MHC I 7 MHC II MR1 CD1d HLA-E HLA-G	3 MHC I	7 MHC I

# Applications of MHC and TCR Monomers

### Immunogenicity Assessment

For protein drugs, *in vitro* assays are employed to evaluate the binding of (neo)-epitopes with MHC molecules<sup>1</sup>.

### **Epitope and TCR Discovery**

Multiplex screening of large epitope panels using a library of dCODE Dextramer<sup>®</sup> reagents assembled using loadable MHC Monomers. Simultaneously identify the TCR sequences as well as the epitopes with which the TCRs interact. For applications including TCR-T cell therapy.

### **TCR:pMHC Binding Affinity**



### **pMHC Binding Affinity**

Use plates coated with peptide-receptive MHC monomers and load with candidate epitopes to assess the strength of binding affinity between the MHC and each peptide.

### **TCR Validation**

Screen for functionality of candidate TCRs against target pMHC Monomers.



### **TCR Cross-Reactivity**

More than 65 additional Human, Primate and Mouse alleles are available through Immudex Custom Solutions and Services.

# MHC Monomers [GMP]

MHC Monomers (GMP) are manufactured by Immudex following good manufacturing practice and meet the requirements for medical devices defined by ISO13485 and 21CFR820.

### References

1. Pai et al., 2023, Cancer Cell 41(4), 776-790. 2. Minervina et al., 2022, Nature Immunology 23, 781–790. 3. Hölzemer *et al.*, 2015, PLoS Med 12(11): e1001900.

Use pMHC and soluble TCR monomers to assess TCR:pMHC binding affinity by Surface Plasmon Resonance<sup>3</sup>, lateral flow assays or plate-based assays.



Screen TCR specificity using plate-based assays/ELISA with immobilized soluble TCR Monomers and libraries of pMHC monomers including potential off-target antigens.

## **Identification of TCR-like antibodies**



Immunization of pMHC complexes in animal models to identify TCR-like antibodies recognizing the pMHC.

Use pMHC complexes to screen libraries of specific TCR-like antibodies.



Use pMHC complexes to stimulate CD8<sup>+</sup> and CD4<sup>+</sup> T cells for applications including TCR-T and CAR-T cell therapy.

**T-Cell Stimulation or Enrichment** 

### **Assembly of MHC Multimers**

Assemble your own MHC multimers, like a Dextramer<sup>®</sup>, with our U-Load Dextramer<sup>® 1</sup> and U-Load dCODE Dextramer<sup>®</sup> reagents<sup>2</sup>.

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