MHC Multimer Proficiency Testing 2023

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INTRODUCTION TO PROFICIENCY TESTING

Originally developed at the initiative of CIC (the US Cancer Immuno-therapy Consortium of the CRI) and CIMT (the European Association for Cancer Immunotherapy), Immudex has since 2013 offered Proficiency Testing as a service to help researchers and clinicians worldwide evaluate and benchmark their immune monitoring performance with MHC Multimers and T-cell ELISpot assays. The proficiency Testing is open to any laboratory, independent of geographic location or field of interest. Read more about Proficiency Testing here.

In the MHC Multimer Proficiency Testing 2023, the proficiency of participants in detection of antigen specific cells in a HPBMC sample was evaluated relative to other participating laboratories.

Each participant received one pretested PBMC sample and HLA matched MHC multimers presenting two EBV epitopes that detect low and medium abundant antigen-specific T cells respectively. In addition, participants were given the choice of enumerating MAIT cells in the same sample with MR1-Dextramer®

The PBMC sample was pretested at Immudex to ensure consistent results between vials and to check viability of the cells. The viability of the lymphocyte population of the tested PBMC sample was 98%. Each participant measured the percentages of antigen-specific CD8+ T cells in the PBMC samples according to the instructions, but with their own choice of materials (multimer, antibodies, viability marker etc.), and following their own assay protocol (staining tubes/plates, washing buffer etc.).

This report shows the test results and overall performance of the participants without revealing their names and affiliations.

In this Multimer Proficiency Test:

- 8 laboratories from 6 different countries participated.
- All the participants chose to use MHC Dextramer® reagents.
- 4 participants were from Academia, and 4 participants were from industry.
- 7 out of 8 participating laboratories got a proficiency score of > 2.0.

ANALYSES

Table 1 MHC Multimer reagents used in the MHC Multimer Proficiency Testing 2023:

MHC Multimer reagent	MHC Multimer specificity
HLA- B*0801/FLRGRAYGL	EBV-specific
HLA- B*0801/RAKFKQLL	EBV-specific
HLA-B*0801/Neg. Control MHC Dextramer®	
hMR1/5-OP-RU	MAIT cell-specific
hMR1/6-FP	Negative control

Each participant:

- Was assigned a confidential laboratory identification number (Lab ID)
- Received instructions on how to perform the MHC Multimer proficiency test (Appendix 1)
- Received one pretested PBMC sample (HHU20181002)
- Received MHC Dextramer®
- Was recommended to look at the "Assay Harmonization Guidelines" (Appendix 2)
- Was encouraged to analyze samples with their own standard protocol to reflect routine sample analysis conducted in their laboratory.
- Reported the following numbers for each of the 6 analyses with MHC Dextramer®:
    - The number of CD8+ cells
    - The number of CD8+ Multimer+ cells
    - The % of CD8+ Multimer+ cells out of the CD8+ cell population
- Reported the following numbers for each of the 4 analyses with MR1 Dextramer®:
    - The number of CD3+ CD161+ cells
    - The number of CD3+ CD161+ MR1 multimer+ cells
    - The % of CD3+ CD161+ MR1 multimer+ cells out of the CD3+ CD161+ population.
- Reported FCS files, compensation files, and "PowerPoint Dot plot" file.

RESULTS

In this year's MHC Multimer Testing, all 8 participants reported their data.

The reported results are summarized in Figures 1-3 on the following pages and listed in appendix 3. All measurements were done in duplicate. Percentage of antigen-specific CD8+ T cells were calculated out of the total number of CD8+ T cells and similarly the percentage of antigen specific CD3+ CD161+ MAIT cells were calculated in the analyses with MR1 Dextramer®

The raw data are presented in Appendix 3.

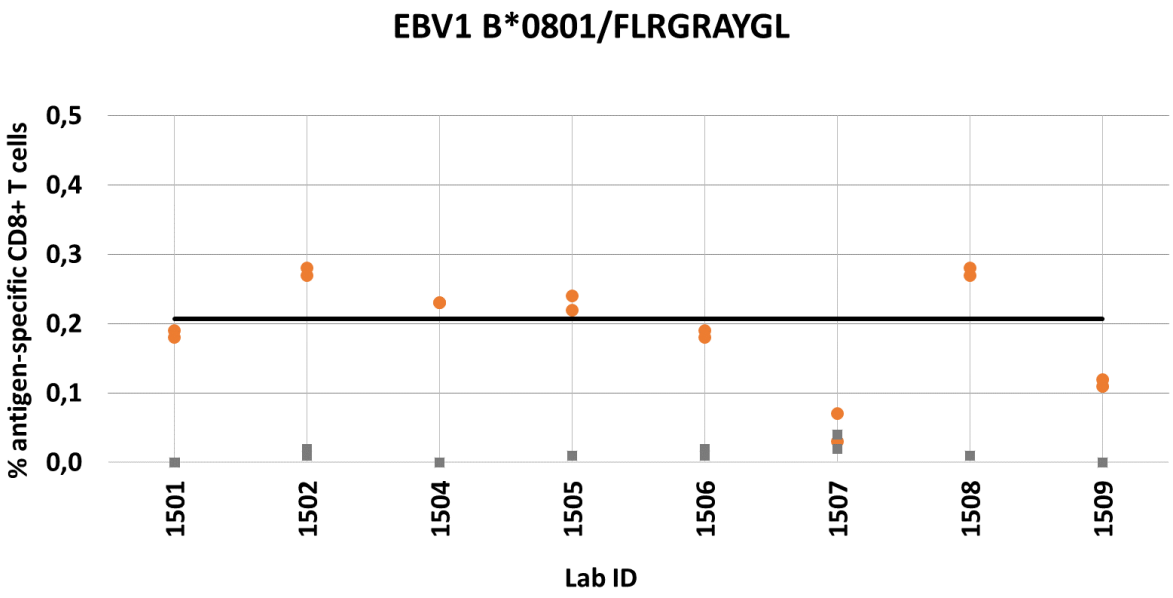

Figure 1 Results from analysis of sample PBMC HHU20181002 stained with EBV specific HLA-B*0801/FLRGRAYGL MHC Dextramer® (orange dots) and negative control MHC Dextramer® (grey squares). Median for EBV1-specific CD8+ T cells is 0,21%.

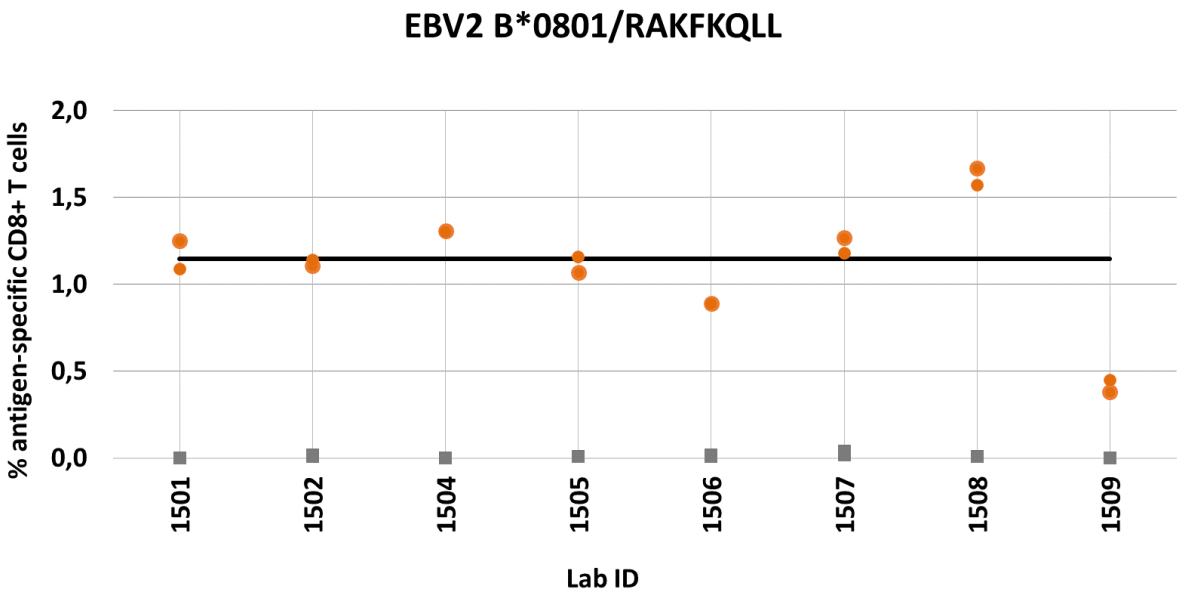

Figure 2 Results from analysis of samples PBMC HHU20181002 stained with EBV specific HLA- B*0801/RAKFKQLL MHC Dextramer® (orange dots) and negative control MHC Dextramer® (grey squares). Median for EBV2-specific CD8+ T cells is 1,15%.

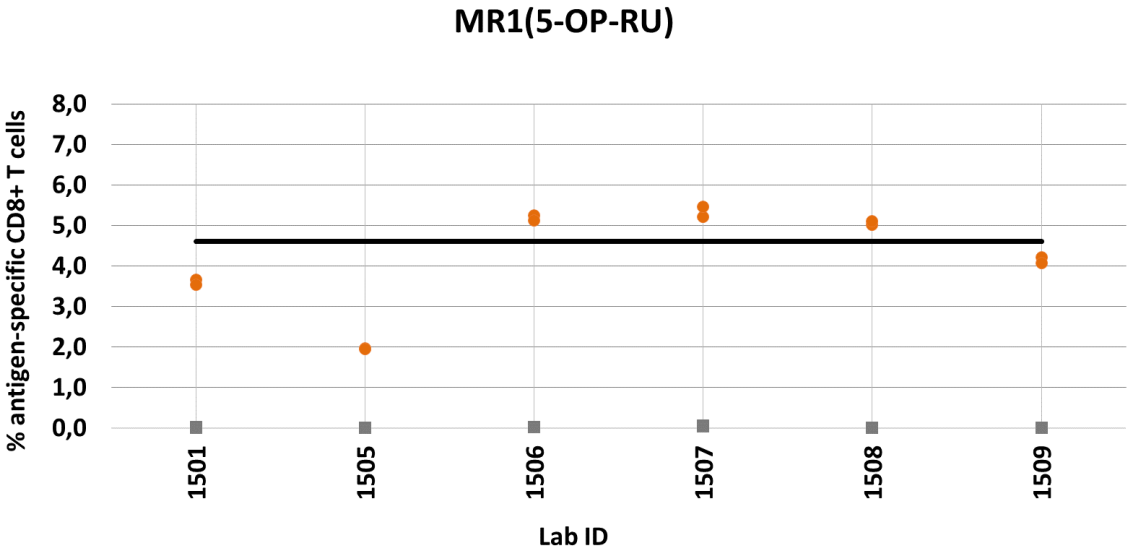

Figure 3 Results from analysis of sample PBMC HHU20181002 stained with HLA-hMR1/5-OP-RU MR1 Dextramer® (orange dots) and negative control hMR1/6-FP MR1 Dextramer® (grey squares). Median for MR1-specific CD3+ CD161+ MAIT cells is 4,6%.
NB: Lab 1502 and 1504 chose not to perform this analysis.

PERFORMANCE

We calculated relative accuracy to compare the performance of one participant with that of all participants as a group. The relative accuracy is a measure of how close each participant is to the median of data reported by all participating labs. It is defined as the mean value of duplicates for each participant measurement divided by the median of measurements of all participants. Relative accuracy scores for each laboratory are listed in Appendix 4 and an example of how the relative accuracy is calculated is shown in Appendix 5. The relative accuracy of the participating labs in the three different analyses are shown in figures 4-6.

Lab performances are divided into three groups and assigned a proficiency score according to how close their results are to the median of all participating laboratories – see table 2 and appendix 4.

Table 2 Definition of the relative accuracy:

Relative Accuracy	Corresponds to	Presented in the figure below as	Proficiency score
0.66≤RA≤1.50	Within the median range	Orange columns	3
0.50≤RA<0.66 **1.50<RA≤2.00**	Near the median range	Grey columns	2
RA<0.50 **RA>2.00**	Far from the median range	Black columns	1

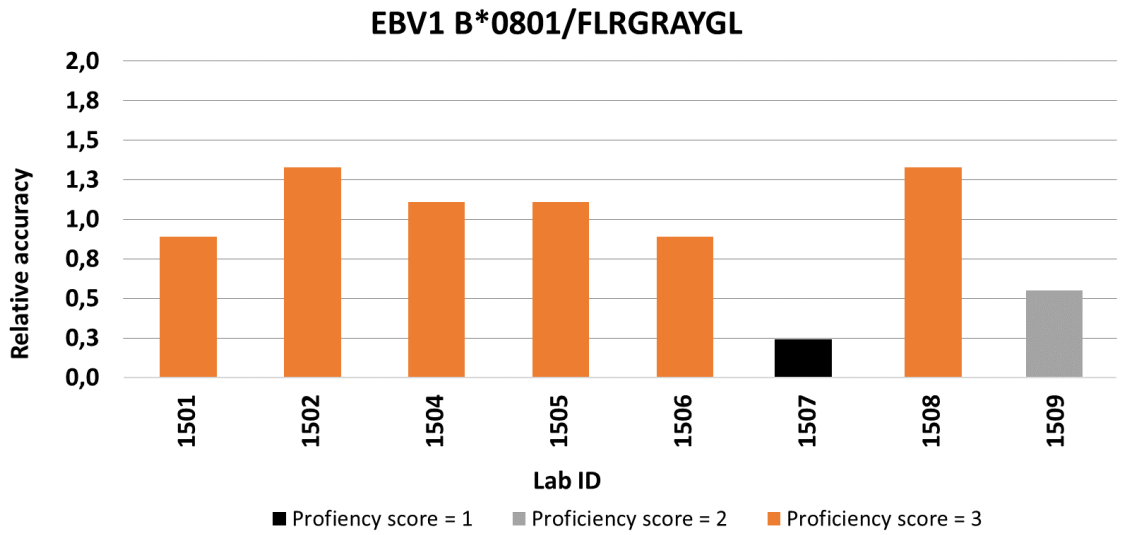

Figure 4 Relative accuracy for analysis of PBMC HHU20181002 stained with the EBV-specific HLA- B*0801/FLRGRAYGL MHC Dextramer®

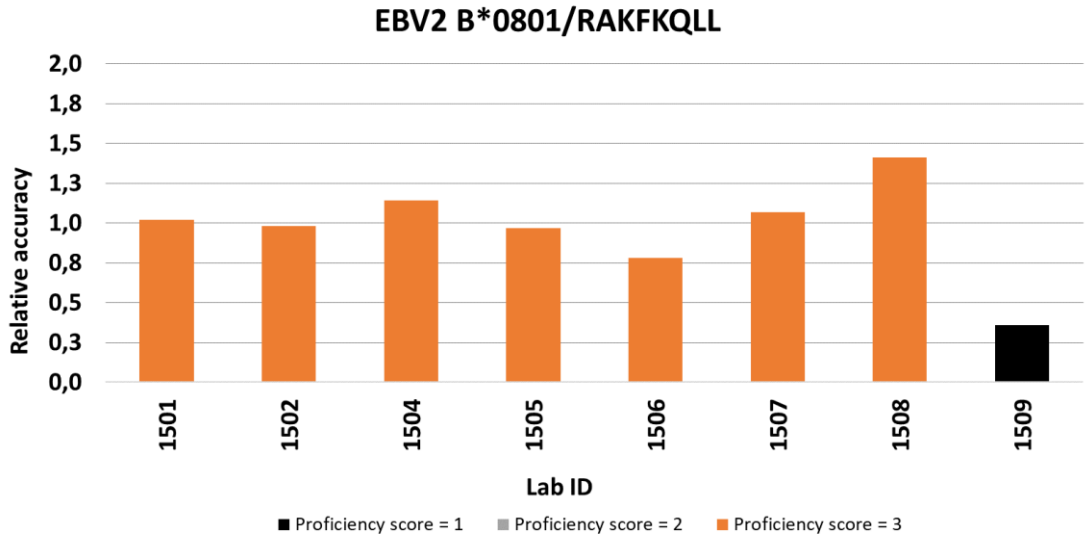

Figure 5 Relative accuracy for analysis of PBMC HHU20181002 stained with EBV specific HLA- B*0801/RAKFKQLL MHC Dextramer®

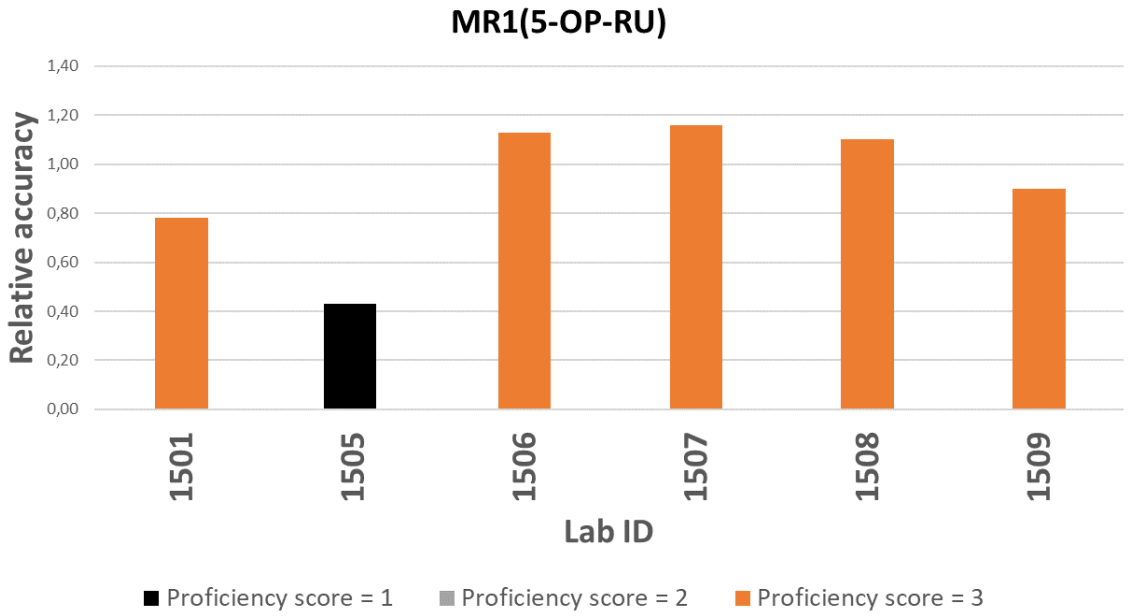

Figure 6 Relative accuracy for analysis of PBMC HHU20181002 stained with the hMR1/5-OP-RU MR1 Dextramer®.

OVERALL PROFICIENCY

The ability of each participant to identify and enumerate antigen-specific T-cells was described with an overall proficiency score. For each of the three types of analysis, the laboratories were assigned a proficiency score between 1-3, see figures 4-6, table 2 and appendix 4. The overall proficiency score was then defined by the average score obtained in the three analyses. Thus, a participant with an overall proficiency score of "3" is in the average range on all three measurements and has the highest possible score. A participant with an average score of "1" is far from average on all three measurements and has the lowest possible score. See calculation of Overall Proficiency Score in Appendix 6. Figure 7 shows the overall proficiency of all the participants.

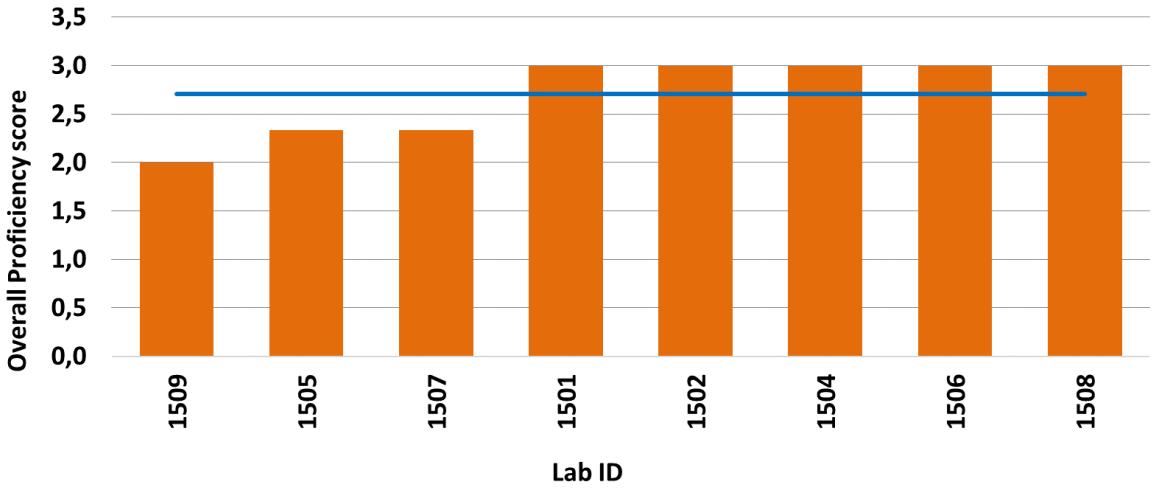

Figure 7. Overall proficiency score of the participants in the MHC Multimer Proficiency Testing 2023. The average overall proficiency score is 2,71 indicated by the blue line.

DISCUSSION

Immudex MHC Multimer Proficiency Testing provides an opportunity for laboratories worldwide to assess their proficiency in monitoring antigen-specific T-cells using flow cytometry-based MHC Multimer assay. Evaluation of laboratory performance is essential to ensure alignment and drive research and development improvements. Harmonized laboratory performance is of high importance in multicenter trials, where clinical results from different sites are compared to evaluate treatment response in immunotherapeutic research and development.

In the MHC Multimer Proficiency Testing 2023, participants used their own laboratory-specific assay protocol to enumerate antigen-specific CD8+ T cells using MHC Dextramer® reagents in flow cytometry-based assays. In this report, each participant can see how well their obtained results align with the rest of the participants. This critical knowledge provides each participant with the opportunity to evaluate their assay protocol, to ensure and sustain their ability to enumerate T-cells accurately, reproducibly, and in alignment with other researchers across sites, or to identify necessary protocol optimizations. To facilitate inter-lab comparisons, we have performed simple statistical data analysis and calculated an overall proficiency score according to criteria chosen by Immudex. However, this is not an exact science and is only meant as a help to get an overview of the results. Different choices of analysis would be equally valid and might have given a slightly different outcome. Simple visual inspection of the distribution of the results (fig 1-3) is also a good way of assessing overall lab performance.

The assays provided reproducible results in all labs with only small variations in the duplicate measurements. In addition, variation between labs was modest with CVs between 30-40% (see appendix 3).

As a novelty this year participants had the opportunity to monitor MAIT cells using Immudex MR1 Dextramer® reagent, which 6 participants accepted. The reported % MAIT cells was detected with same low variation as detection of the viral EBV MHC Dextramer® reagents

To compare the performance of the participants, relative accuracies were calculated and 82% of all reported measurements were found to be in the average range (defined as a relative accuracy of $0.66 \leq RA \leq 1.50$) or in the near average range (defined as $0.50 \leq RA < 0.66$ & $1.50 < RA \leq 2.00$).

We also calculated proficiency scores for the three different analyses based on the relative accuracies (see appendix 4) and an overall proficiency score for all three analyses together (see appendix 6). All 8 participants got a proficiency score of ≥ 2.0 corresponding to being in the average or near average range. This result is similar to what was observed in the last four MHC Multimer Proficiency Testing from 2018-2021, where 89%, 89%, 80% and 90%, , obtained a proficiency score of ≥ 2,0. This is a testimony to the robustness of immune monitoring with MHC Dextramer® reagents.

Conclusively, this Multimer Proficiency Testing shows that MHC Multimer assays are:

- well harmonized across different laboratories
- equally harmonized when looking at virus specific (EBV) antigen specific CD8+T-cells.
- Detection of CD3+, CD161+ MAIT cells using MR1-Dextramer® reagents showed the same low variability as detection of the viral antigen specific T cells
- a useful tool for evaluating treatment response in immunotherapeutic research and development.

ABOUT IMMUDEX

Based in Virum, Denmark, with North American operations based in Fairfax, Virginia, Immudex manufactures MHC Dextramer® and other Dextramer®-based products for the detection of immune cells.

Immudex' MHC Dextramer® products are utilized for the quantification or sorting of antigen-specific T cells in life science research, in-vitro diagnostics, as well as the development of immunotherapeutics and vaccines. The primary focus is research-use-only products for the immune monitoring of immunotherapy development. But we also offer MHC I Dextramer® produced according to current good manufacturing practices (cGMP) Immudex is ISO 13485:2016 certified, registered with the FDA and audited regularly, which guarantees that MHC I Dextramer® (GMP) are produced in compliance with strict international cGMP standards for medical devices regarding quality control and product traceability. We have developed a kit for monitoring of CMV cellular immunity in transplant and other immune-deficient patients. In Europe, the CE-marked Dextramer® CMV Kit is approved for in vitro diagnostic use to quantify CMV-specific T cells. USA FDA 510(k) clearance for the CMV kit was granted in March 2017.

Our state-of-the-art dCODE Dextramer® reagents enable massive multiplexing of antigen-specific T-cell detection. Detection of over 1000 CD8+ T-cell specificities from a single blood sample has been achieved.

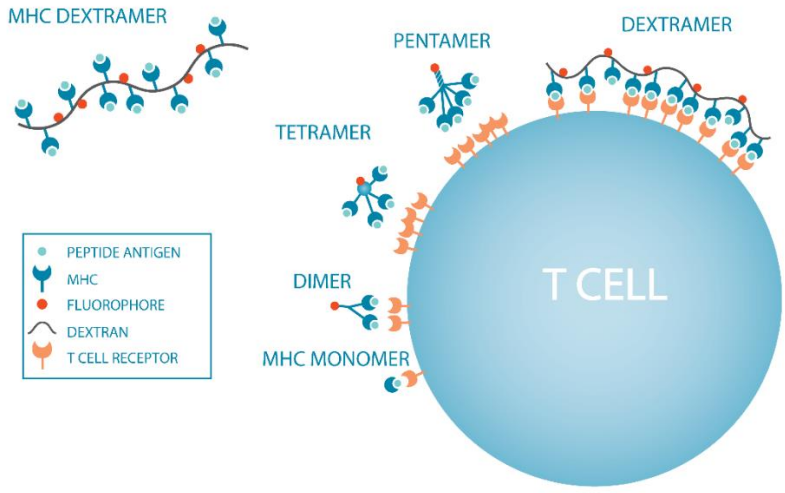

Figure 10 Schematic drawing of MHC Dextramer® and conventional MHC multimers binding to T-cell receptors (TCRs) on the surface of a T cell. MHC Dextramer® reagents are fluorescently-labeled MHC multimers that can bind simultaneously to multiple TCRs on a single T cell with exceptional avidity. This enables sensitive detection and isolation of antigen-specific T cell populations with a broad range of TCR affinities.

RESOURCES FROM IMMUDEX

We are committed to building a global community of proficiency in immune monitoring. Reach to us if you have questions or want to know more about the Immudex Proficiency Testing.

Proficiency Testing

Access the Immudex Proficiency Testing site, where you will find information about MHC Multimer and ELISpot Proficiency Testing.

Read more

Contact the Proficiency Testing Coordinator

We are here to support you through all the process. From the proficiency testing to answering questions regarding deadlines, PBMC samples, data analysis. We want to ensure the process is easy for you.

proficiencypanel@immudex.com

Performance Reports

Curious about previous year's results? Find out more for MHC Multimer and ELISpot Proficiency Testing.

MHC Multimer Proficiency Testing reports

ELISpot Proficiency Testing Reports

Technical Support

Let us know if you experience difficulties or have questions. Immudex will help you get the most out of your Dextramer® products.

APPENDIX 1: INSTRUCTIONS FOR PROFICIENCY TESTING

PLEASE READ INSTRUCTIONS CAREFULLY BEFORE THAWING AND STAINING THE CELLS

Introduction

Making accurate, reproducible, and state-of-the-art T-cell immune monitoring analysis is becoming increasingly important in immunotherapeutic research and development. By participating in this Proficiency Testing, you get the chance to assess how well you detect antigen-specific CD8+ T cells, and unconventional T cells such as the MAIT cells, using predefined MHC Multimer reagents.

Please analyze the provided PBMC sample according to these instructions and report your results back to Immudex. Results and performance from all participants are presented in a final report, where participant's names and affiliation are kept anonymous. In the report. you will be able to see how accurately you enumerated different antigen-specific T cell populations compared to the other participating laboratories, including Immudex. This will allow you to assess your MHC Multimer assay protocol – to confirm your ability to accurately identify antigen-specific T-cell responses, or perhaps discover a need for protocol optimizations.

We recommend you have a look at the Multimer Harmonization Guidelines (Appendix 2).

Deadlines and Immudex contact

Data submission: September 22, 2023

Report from Immudex: November 10, 2023

If you have any questions, please contact the organizer:

Niels Montano Frandsen, Global Product Manager
proficiencypanel@immudex.com

or customer support:
customer@immudex.com

PBMC samples

You will receive one vial of pre-tested PBMC samples:
PBMC HHU20181002 (the vial contains ≥ 10 million cells in 1.5ml).
Please store PBMCs at ≤ -150°C upon arrival.

MHC Multimer reagents

The following MHC Multimer reagents are needed for the experiments:

- EBV1-specificity: HLA-B*0801/FLRGRAYGL MHC Multimer
- EBV2-specificity: HLA-B*0801/RAKFKQLL MHC Multimer
- Negative Control MHC Multimer
- hMR1 / 5-OP-RU multimer
- hMR1 / 6-FPmultimer (negative control)

If you have requested MHC Dextramer® reagents, you will receive these five PE-labeled reagents:

• WI2147-PE 10	EBV1: HLA-B*0801/FLRGRAYGL MHC Dextramer®	10 tests
• WI2148-PE 10	EBV2: HLA-B*0801/RAKFKQLL MHC Dextramer®	10 tests
• WI03233-PE 10	HLA-B*0801/Neg. Control MHC Dextramer®	10 tests
• ZA08004-PE 10	MR1 Dextramer®, hMR1/5-OP-RU	10 tests
• ZA08003-PE 10	MR1 Dextramer®, hMR1/6-FP (Neg. Control)	10 tests

MHC Dextramer® reagents must be stored at 2-8°C protected from light.

Additional reagents needed for analysis

We recommend you use your own choice of materials (multimer, antibodies, viability marker etc.) and protocol (staining tubes/plates, washing buffer etc.) for the Proficiency Test to make it reflect routine sample analysis being conducted in your laboratory. However, in addition to MHC Multimer it is necessary to include anti-CD8 antibody for staining of EBV specific T-cells and both anti-CD3 and anti-CD161 antibodies must be included to identify hMR1 Multimer stained MAIT cells.

Experimental setup

Analyze the two PBMC samples as listed below in duplicates (summarized in Table 1). Please note, that the indicated staining ID's must be used when naming the FCS files.

Stain PBMC HHU20181002 with:

- Negative Control MHC Multimer
- HLA-B*0801/FLRGRAYGL MHC Multimer
- HLA-B*0801/RAKFKQLL MHC Multimer
- hMR1 / 5-OP-RU Multimer
- hMR1 / 6-FP (Neg. Control)

Table 3 Required analysis for the MHC Multimer Proficiency Testing 2023

Staining ID	PBMC Donor	MHC Multimer specificity
R1	PBMC HHU20181002	Negative control
R2	PBMC HHU20181002	Negative control
R3	PBMC HHU20181002	B*0801/FLRGRAYGL
R4	PBMC HHU20181002	B*0801/FLRGRAYGL
R5	PBMC HHU20181002	B*0801/RAKFKQLL
R6	PBMC HHU20181002	B*0801/RAKFKQLL
R7	PBMC HHU20181002	hMR1 / 6-FP (Neg.
R8	PBMC HHU20181002	hMR1 / 6-FP (Neg.
R9	PBMC HHU20181002	hMR1 / 5-OP-RU
R10	PBMC HHU20181002	hMR1 / 5-OP-RU

Instructions for cell preparation and sample analysis

Thaw PBMCs and count the cells.

Please record:

- Total cell numbers
- Viability of the cells (% viable cells)

Staining and gating

Use your own protocol for staining and subsequent gating of the MHC Multimer-specific T cells and MR1 Multimer-specific MAIT cells, but we recommend that you have a look at the gating strategies outlined in the accompanying PowerPoint slides (slides 1 and 3).

Due to the great difference in abundance of EBV and the MR1 positive cells, we recommend that you use ¾ of the cells for the EBV specificities, and the remaining ¼ of the cells for the MR1 specificity.

Due to the frequency of the EBV specific T cells in this HPBMC sample we deviate from the "Assay harmonization guidelines" (see page 6) for acquiring 100.000 CD8+ T cells, we recommend acquiring at least 20.000 CD8+ T cells, if possible.

NB: If you use MHC Dextramer® reagents, please read the staining protocol provided with the reagents (also available on our website (https://www.immudex.com/resources/protocols/ - MHC Dextramer® Staining Protocol).

Data recording

For the EBV-specific MHC Multimers you will need to:

- Record the number of CD8+ T cells (e.g., number of events in the plots shown in Figure 1A).
- Record the number of MHC multimer+ CD8+ T cells (e.g., number of events in gate R1 to R6 in Fig 1A).
- Calculate the percentage of MHC multimer+ CD8+ T cells out of total CD8+ T cells (ex: (R3/total CD8+) x 100 %). Please record all results with two decimals.

For the MR1 specific Multimer you will need to:

- Record the number of CD3+ CD161+ cells (e.g., number of events in plots shown below in Figure 1B).
- Record the number of MR1 multimer+, CD3+ CD161+ MAIT cells (e.g., number of events in gate R7- R10 in Fig 1B).
- Calculate the percentage of MR1 multimer+, CD3+ CD161+ MAIT cells out of total CD3+ CD161+ cells (ex: (R9/total CD3+ CD161+) x100 %). Please record all results with two decimals.

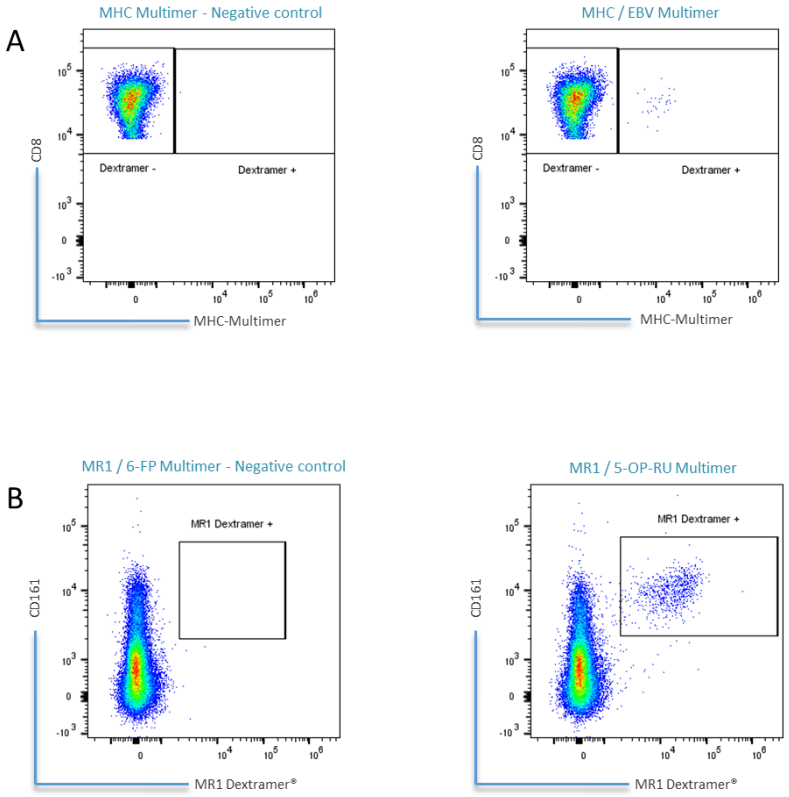

Figure 4. Example of Multimer gating for A) EBV and B) MR1 multimers.
Please see "PowerPoint Dot plot" (provided by email) for gating example.

Reporting data

1. Fill in the "PowerPoint Dot plot" slide (provided by email) with your gating strategy and dot plots. The dot plots must show CD8 staining on the y-axis and MHC Multimer staining on the x-axis as illustrated on the slides 1-2 and CD161 on the y-axis and MR1 Multimer staining on the x-axis as illustrated on slides 3-4.

2. Create a Zip file, name it with your Lab ID (provided by email) and include the following files:
    a. The filled-in "PowerPoint Dot plot".
    b. The 10 FCS files, named exactly as described in Table 1.
    c. If acquired, include your single-color compensation files.

3. Data reporting
    a. Upload data zip file here: https://immudex.sharefile.com/i/i24a3625aace41a3b
    b. Report data and results obtained from sample analysis in this survey: https://immudex.wufoo.com/forms/r1fmm8j804trpoi/

APPENDIX 2: ASSAY HARMONIZATION GUIDELINES

Multimer Harmonization Guidelines to Optimize Assay Performance

A. Establish lab SOP for MHC peptide multimer staining:
A1. Count at least 100,000 CD8+ T cells per staining. A2. Establish adequate measures to quantify non-specific binding of Multimer to CD8+ cells (e.g. irrelevant Multimer or autofluorescence). A3. Establish adequate measures to reduce the amount of non-specific binding of Multimer in the CD8+ population to allow accurate quantification (e.g. DUMP channel or DEAD cell dyes).

B. Establish SOP for software analyses of stained samples, including:
B1. Gating strategy.
B2. Rules to set the gates.

C. Establish a human auditing process of all results:
C1. Are all dot plots correctly compensated?
C2. Have the gates been set correctly?
C3. Are the reported frequencies of multimer-positive cells plausible?

D. Lab environment
D1. Only let experienced personnel (per lab SOP) conduct assay.

APPENDIX 3: PARTICIPANT DATA

Donor lot.	HHU20181002	% Multimer positive cells of the CD8+ T cells								
Multimer Reagent		Neg.ctrl			EBV 1: B*0801/FLRGRAYGL			EBV 2: B*0801/RAKFKQLL		
Lab ID number	% Viability	R1	R2	Average	R3	R4	Average	R5	R6	Average
1501	96	0,00	0,00	0,00	0,19	0,18	0,19	1,25	1,09	1,17
1502	89	0,02	0,01	0,02	0,27	0,28	0,28	1,11	1,14	1,13
1504	82	0,00	0,00	0,00	0,23	0,23	0,23	1,31	1,30	1,31
1505	90	0,01	0,01	0,01	0,22	0,24	0,23	1,07	1,16	1,12
1506	94	0,02	0,01	0,02	0,18	0,19	0,19	0,89	0,89	0,89
1507	84	0,02	0,04	0,03	0,07	0,03	0,05	1,27	1,18	1,23
1508	46	0,01	0,01	0,01	0,27	0,28	0,28	1,67	1,57	1,62
1509	68	0,00	0,00	0,00	0,11	0,12	0,12	0,38	0,45	0,42
all labs	**Median**			**0,01**			**0,21**			**1,15**
						SD	0,07			0,33
						Mean	0,19			1,11
						CV	38%			29%

Donor lot.	HHU20181002	% MR1 Multimer positive cells					
Multimer Reagent		MR1(6-FP) Neg.ctrl			MR1(5-OP-RU)		
Lab ID number	% Viability	R7	R8	Average	R9	R10	Average
1501	96	0,02	0,00	0,01	3,67	3,54	3,61
1502	89	n.a	n.a	n.a	n.a	n.a	n.a
1504	82	n.a	n.a	n.a	n.a	n.a	n.a
1505	90	0,00	0,00	0,00	1,97	1,95	1,96
1506	94	0,03	0,03	0,03	5,24	5,12	5,18
1507	84	0,06	0,05	0,06	5,22	5,46	5,34
1508	46	0,00	0,00	0,00	5,01	5,11	5,06
1509	68	0,01	0,01	0,01	4,22	4,07	4,15
all labs	**Median**			**0,01**			**4,60**
						SD	1,18
						Mean	4,22
						CV	28%

APPENDIX 4: CALCULATIONS OF PROFICIENCY SCORES

HHU20181002	EBV 1: B*0801/FLRGRAYGL				
Lab ID	Average % Dextramer+ cells	0.66≤RA≤1.50	0.50≤RA<0.66 & 1.50<RA≤2.00	RA<0.50 & RA>2.00	Proficiency score
1501	0,19	0,89			3
1502	0,28	1,33			3
1504	0,23	1,11			3
1505	0,23	1,11			3
1506	0,19	0,89			3
1507	0,05			0,24	1
1508	0,28	1,33			3
1509	0,12		0,55		2
Median	**0,21**				

HHU20181002	EBV 2: B*0801/RAKFKQLL				
Lab ID	Average % Dextramer+ cells	0.66≤RA≤1.50	0.50≤RA<0.66 & 1.50<RA≤2.00	RA<0.50 & RA>2.00	Proficiency score
1501	1,17	1,02			3
1502	1,13	0,98			3
1504	1,31	1,14			3
1505	1,12	0,97			3
1506	0,89	0,78			3
1507	1,23	1,07			3
1508	1,62	1,41			3
1509	0,42			0,36	1
Median	**1,15**				

HHU20181002	MR1(5-OP-RU)				
Lab ID	Average % Dextramer+ cells	0.66≤RA≤1.50	0.50≤RA<0.66 & 1.50<RA≤2.00	RA<0.50 & RA>2.00	Proficiency score
1501	3,61	0,78			3
1502	n.a	n.a	n.a	n.a	n.a.
1504	n.a	n.a	n.a	n.a	n.a.
1505	1,96			0,43	1
1506	5,18	1,13			3
1507	5,34	1,16			3
1508	5,06	1,1			3
1509	4,15	0,9			3
Median	**4,60**				

APPENDIX 5: CALCULATION OF THE RELATIVE ACCURACY

Table 4 Example of relative accuracy calculation on PBMC donor HHU20181002 stained with the HLA-B*0801/FLRGRAYGL.

HHU20181002	% MR1 Multimer positive cells						
Multimer Reagent	MR1(6-FP) Neg.ctrl			MR1(5-OP-RU)			
Lab ID number	R7	R8	Average	R9	R10	Average	Relative accuracy
1501	0,02	0,00	0,01	3,67	3,54	3,61	3,61/4,60= 0,78
Median (all labs)							4,60

APPENDIX 6: CALCULATION OF THE OVERALL PROFICIENCY SCORE

	Proficiency score			Overall proficiency score
Lab ID no.	B*0801/FLRGRAYGL	B*0801/RAKFKQLL	MR1(5-OP-RU)	(mean)
1501	3	3	3	3,0
1502	3	3	n.a.	3,0
1504	3	3	n.a.	3,0
1505	3	3	1	2,3
1506	3	3	3	3,0
1507	1	3	3	2,3
1508	3	3	3	3,0
1509	2	1	3	2,0