NIH TETRAMER CORE FACILITY

Human MR1 tetramer staining

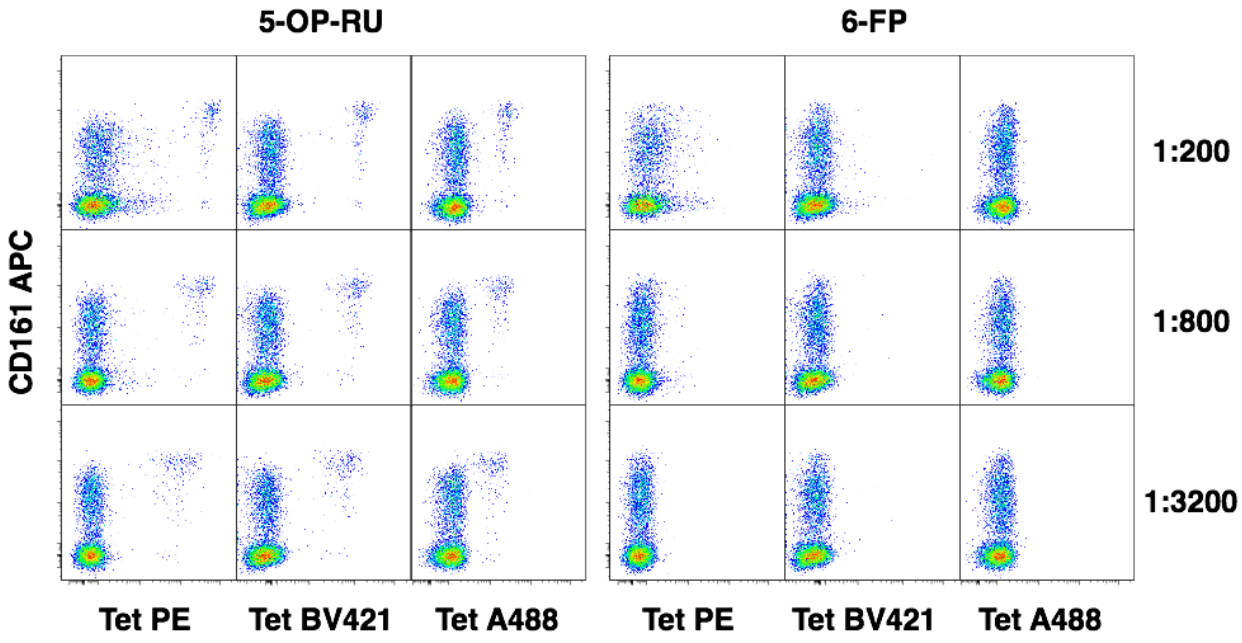
Identification of MAIT cells with MR1 tetramers was described in "T-cell activation by transitory neo-antigens derived from distinct microbial pathways." Corbett AJ, Eckle SB, Birkinshaw RW, Liu L, Patel O, Mahony J, Chen Z, Reantragoon R, Meehan B, Cao H, Williamson NA, Strugnell RA, Van Sinderen D, Mak JY, Fairlie DP, Kjer-Nielsen L, Rossjohn J, McCluskey J. (2014). Nature. 509, 361-5. DOI: http://dx.doi.org/10.1038/nature13160 PMID: 24695216. Please cite this paper if you publish using these tetramers.

Human MR1 tetramers are loaded with 5-A-RU in the presence of methylglyoxal, resulting in the activating ligand 5-OP-RU. These tetramers stain brightly with a number of fluorochromes as shown in the figure below. You should expect 1-5% of human peripheral blood T cells to stain with the 5-OP-RU tetramer. We are providing 6-FP loaded MR1 tetramers as a negative control.

Clients should titrate the reagents in their assays, however a good starting point would be:

- 1:1000 for bright fluorochromes (PE, APC, BV421)
- 1:500 for other fluorochromes (Alexa 488, etc.)
- Note that using higher concentrations of tetramer is wasteful and will increase background staining, see the top left panel in the figure below.

Please contact Rick Willis, Technical Director (richard.willis@emory.edu, 404-727-7215), with any questions about the reagents.



Human peripheral blood was stained with hMR1 tetramers at the dilutions shown for 40 min at room temperature followed by other surface stains for 20 min. Cells were fixed and RBCs were lysed with BD FACS lysing solution. Plots above were gated on $CD3^{+}CD19^{-}$ lymphs. We expect a wide range of staining conditions will work well for human MR1 tetramers.