

CEF Single Peptide Kit (Class I)

Catalog #: PA-CEF-005

Lot: _____

Product: CEF Single Peptide Kit (Class I)

Description: The CEF Single Peptide Kit contains 32 Class I peptides, each corresponding to a defined HLA class I-restricted T cell epitope from human Cytomegalovirus, Epstein-Barr virus and Influenza virus, packed in individual vials.

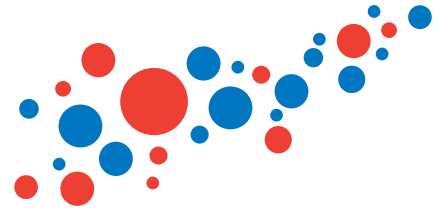
Activity: The CEF peptides stimulate corresponding peptide-specific CD8⁺ memory T cells to release IFN- γ . These peptides are restricted by HLA-A1, -A2, -A3, -A11, -A24, -A68, -B7, -B8, -B27, -B35, and -B44 molecules, and have been shown to elicit recall responses expressing these rather common MHC class I alleles – most humans have been previously exposed to these pathogens.

Recommended Use: The CEF peptides are recommended as positive controls for detecting antigen-specific CD8⁺ cells in human PBMC, for example, when performing cytokine assays for immune monitoring purposes. Such assays include IFN- γ measurements by ELISPOT and intracytoplasmic cytokine staining (ICS) – for accurate frequency measurements of the cytokine producing CD8 cells – or cytokine bead arrays (CBA), and ELISA – for a semi-quantitative readout.

Instruction for Use: *Stock solution:* Flick tube to ensure all powder is at the bottom of the tube. Add 4 μ l of tissue culture-grade DMSO followed by 16 μ l of sterile double-distilled water to peptide vial. Vortex briefly and maintain sterile handling.

Fill up to 1ml, by adding 980 μ l of tissue culture grade PBS and vortex briefly. Flick tube to ensure that all liquid is at the bottom of the tube. The stock solution (1ml, at 20 μ g/ml) is ready for use. The stock solution can be stored at 4°C for one week. CTL recommends aliquoting and storing the stock solution at -20°C to -80°C for long-term storage.

Working Solution: Prepare a 1:5 dilution (2x) of the stock in cell culture medium (e.g., CTL-Test™ Medium) as the working solution (Use peptide solutions at 2 μ g/ml of final peptide concentration that is at 1:10 dilution of the stock.) For ELISPOT assays we recommend plating 100 μ l of this 2x peptide working solution per well, directly into the ELISPOT plate, followed by the addition of 100 μ l of the PBMC for a 24h assay duration. For exact frequency measurements, we recommend testing in triplicate. Because serum is the greatest variable in assay performance, we recommend the use of serum-free media (CTL-Test™ Medium) at all steps of the assay. CTL offers such serum-free media (Cat# CTLT-010, or CTLT-005) that has been customized for low background/high signal performance with PBMC.



Storage: In LYOPHILIZED powder form store at 4°C for short term. For long-term storage, store at -20°C. The stock solution can be stored at 4°C for one week; however -20°C to -80°C is mandatory for long-term storage.

Peptides: VSDGGPNLY (Influenza A, HLA-A1), CTEKLSDY (Influenza A, HLA-A1), GILGFVFTL (Influenza A, HLA-A2), FMYSDFHFI (Influenza A, HLA-A2), CLGGLTMV (EBV, HLA-A2), GLCTLVAML (EBV, HLA-A2), NLVPMVATV (HCMV, HLA-A2), KTGGPYKR (Influenza A, HLA-A68), RVLSFIKGTK (Influenza A, HLA-A3), ILRGSVAHK (Influenza A, HLA-A3), RVRAYTYSK (EBV, HLA-A3), RLRAEAQVK (EBV, HLA-A3), SIIPSGPLK (Influenza A, HLA-A3/A11/A6), AVFDRKSDAK (EBV, HLA-A11), IVTDFSVIK (EBV, HLA-A11), ATIGTAMYK (EBV, HLA-A11), DYCYNLNKEF (EBV, HLA-A24), LPFDKTTVM (Influenza A, HLA-B7), RPPIFIRRL (EBV, HLA-B7), ELRSRYWAI (Influenza A, HLA-B8), RAKFKQLL (EBV, HLA-B8), FLRGRAYGL (EBV, HLA-B8), QAKWRLQTL (EBV, HLA-B8), SDEEEAIVAYTL (HCMV, HLA-B18), SRYWAIRTR (Influenza A, HLA-B27), ASCMGLIY (Influenza A, HLA-B27), RRIYDLIEL (EBV, HLA-B27), YPLHEQHGM (EBV, HLA-B35), IPSINVHHY (HCMV, HLA-B35), EENLLDFVRF (EBV, HLA-B44), EFFW-DANDIY (HCMV, HLA-B44), TPRVTGGGAM (HCMV, HLA-B7).

References: Currier JR, Kuta EG, Turk E, Earhart LB, Loomis-Price L, Janetzki S, Ferrari G, Birx DL, Cox JH. A panel of MHC class I restricted viral peptides for use as a quality control for vaccine trial ELISPOT assays. *J. Immunol. Methods* 260:157-172, 2002.

NIH AIDS Research & Reference Reagent Program
(www.aidsreagent.org/UploadDocs/9808_001.pdf)

Warning: For laboratory research use only, not for diagnostic use. Not for use in humans.

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