

Background: Immune Epitopes

Epitopes are parts of antigens interacting with receptors of the immune system, such as T cell receptors (TCR) and antibodies. Epitope recognition by these receptors mediates the differentiation between self and non-self by the immune system. Knowledge about epitope structures and how they affect the immune response is required to continue development of techniques that detect, monitor, and fight diseases.

Use Case: T cell Epitope Mapping

Objective

An investigator wants to identify which parts of the M2 protein of the influenza H5N1 strain (bird flu) are recognized by T-cells following infection in mice.

Experimental Protocol

Experimental Infection

Five healthy mice of the strain C57BL/6 at age 8 weeks are infected intraperitoneal (i.p.) with live influenza H5N1 organisms in PBS.

Ex Vivo Sample Collection

14 days post infection, the mice are sacrificed, and their spleens removed. A specific subset of T-cells (CD8+ cells) is extracted from the spleen cells using magnetic beads.

T cell Response Assay

An immortalized murine lymphoblast cell line expressing a single MHC class I molecule (H-2 K^b) is pulsed with a peptide from the influenza H5N1 M2 protein. The peptide pulsed cells (antigen presenting cells) are incubated together with the CD8+ cells (effector cells) on an ELISPOT plate, and the number of IFN-gamma producing cells per million effector cells is recorded. The same assay is repeated for a number of peptides which span the H5N1 M2 protein. As a negative control, the assay is repeated with CD8+ cells from naïve mice.

Data Analysis

The number of spots is considered to be significant above background if it is twice the number of spots found in the negative control.

Conclusion about Biological Reality

Peptide X, which is contained in the M2 protein of H5N1 is / isn't an H-2 K^b restricted T-cell epitope, recognized in C57BL/6 mice following infection with H5N1