

In Vivo Lymphocyte Homing in Mice IACUC Standard Procedure Effective Date: August 2025

Description of procedure:

Donor mice will be euthanized per the <u>UCSF Euthanasia Guidelines</u>, and lymphoid organs will be harvested to prepare leukocytes for injection into recipient mice of the same genetic background. These leukocytes may be labeled with a non-toxic fluorescent dye for tracking. Recipient mice will be kept warm to facilitate vasodilation and restrained briefly in a mouse restrainer. Optionally, at time 0, antibody or other test compounds in sterile saline may be injected IV into the tail vein per the <u>UCSF Tail Vein Injection procedure</u>. After a defined period (e.g. 1 hour to 1 day), the mouse is injected by IV into the tail vein with the desired number of leukocytes (typically between 10^6 and 10^7) from the donor mice in sterile saline. Alternatively, recipient mice are anesthetized and cells are transferred by <u>Retro Orbital Injection</u>. The mouse is returned to the cage. Endpoints for experiments are varied (minutes to days), and the timeline will be specified in the individual protocol.

Agents:

This procedure requires fluorescent dye, test compound, and sterile saline. All non-pharmaceutical agent use must be justified and all agents administered to animals should be listed in the "Agents" section of the RIO IACUC protocol.

Adverse Effects:

Adverse effects should be listed in the "Adverse Effects" section of the RIO IACUC protocol.

Examples of potential adverse effects include: Reaction to agent including lethargy and labored breathing

Alternatives Considerations Search:

Date of	Key Words	Search	<u>Years</u>
Search		Site	<u>Covered</u>
9/5/2025	Lymphocyte, homing, allogenic, transplant, technique, study, measure, alternative, refinement, distress, pain, mouse	PubMed	1965-2025

References:

Butcher, E.C. and Ford, W.L. (1986) Following cellular traffic: methods of labelling lymphocytes and other cells to tracetheir migration in vivo. In: D.M. Weir and L.A. Herzenberg (Eds.), Handbook of Experimental Immunology, 4th edn. Blackwell, Oxford, p. 57.

Loeffler D, Ratner S. In vivo localization of lymphocytes labelled with low concentrations of Hoechst 33342. J Immunol Methods. 1989 Apr 21;119(1):95-101. doi: 10.1016/0022-1759(89)90385-2. PMID: 2708828.



Weston S, Parish C. New fluorescent dyes for lymphocyte migration studies. Analysis by flow cytometry and fluorescence microscopyJournal of Immunological Methods, 133 (1990) 87-97. doi: 10.1016/0022-1759(90)90322-M