

STEMprep™ Mouse Lung Dissociation Kit



For processing 50 lungs using the STEMprep™ Tissue Dissociation System

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Catalog #100-2135

50 Preparations

TOLL FREE PHONE 1 800 667 0322 • PHONE +1 604 877 0713

INFO@STEMCELL.COM • TECHSUPPORT@STEMCELL.COM

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Product Description

STEMprep™ Mouse Lung Dissociation Kit provides an optimized enzymatic dissociation cocktail for processing lung tissues into single-cell suspensions. When used with the STEMprep™ Tissue Dissociator (Catalog #100-2112), mechanical dissociation is combined with enzymatic degradation of the extracellular matrix, efficiently breaking down tissue structure while preserving cellular integrity. This approach yields a high number of leukocytes and endothelial cells. The resulting single cells are immediately suitable for downstream applications, such as cell separation, culture, or various analyses.

For best results, use with STEMprep™ Tissue Dissociator and STEMprep™ Sample Tubes (Catalog #200-0800).

Product Information

The following components are sold as part of STEMprep™ Mouse Lung Dissociation Kit (Catalog #100-2135) and are not available for individual sale.

COMPONENT NAME	COMPONENT #	SIZE	STORAGE	SHELF LIFE
STEMprep™ Enzyme A	100-2131	1 x 2.5 mL	Store at -20°C.	Stable until expiry date (EXP) on label.
STEMprep™ Enzyme B	100-2149	2 x 1.25 mL	Store at -20°C.	Stable until expiry date (EXP) on label.
STEMprep™ Enzyme Diluent Z	100-2134	1 x 125 mL	Store at 2 - 8°C. Do not freeze.	Stable until expiry date (EXP) on label.

Materials Required but Not Included

PRODUCT NAME	CATALOG #
50 mL Conical Tubes	e.g. 38010
70 µm Cell Strainer	e.g. 27260
Culture Dish, Non-Treated	e.g. 38045
D-PBS (Without Ca++ and Mg++)	37350
EasySep™ Buffer OR PBS containing 2% FBS and 1 mM EDTA	20144
STEMprep™ Sample Tubes	200-0800
STEMprep™ Tissue Dissociator	100-2112

Preparation of Reagents and Materials

Thaw STEMprep™ Enzyme A and Enzyme B at room temperature (15 - 25°C) for immediate use or overnight at 2 - 8°C. Do not thaw at 37°C.

NOTE: Once thawed, use immediately or aliquot and store at -20°C until the expiry date as indicated on the label. After thawing the aliquots, use immediately. Do not re-freeze.

LUNG DISSOCIATION ENZYME COCKTAIL

Prepare lung dissociation enzyme cocktail fresh before use.

For one lung, prepare 2.5 mL of lung dissociation enzyme cocktail by combining the following in a STEMprep™ Sample Tube:

- 2.4 mL of Enzyme Diluent Z
- 50 µL of Enzyme A
- 50 µL of Enzyme B

Sample Preparation

Dissect mouse lung into a non-treated culture dish containing cold PBS. Remove thymus, heart, and trachea. Trim blood vessels and connective tissue from the lung tissue. Using scissors, carefully dissect the lung into its five individual lobes.

Directions for Use

1. Transfer all lung lobes from one mouse (250 - 495 mg) into the STEMprep™ Sample Tube containing 2.5 mL of lung dissociation enzyme cocktail.
NOTE: Plastic Rack for Centrifuge Tubes, 50 mL (Catalog #200-0651) is provided with each STEMprep™ Tissue Dissociator System to manage STEMprep™ Sample Tubes while loading or removing tissues.
2. Tightly close the STEMprep™ Sample Tube lid (ensuring proper closure) and insert it into a slot on the STEMprep™ Tissue Dissociator. For complete instructions on using the instrument, refer to the Technical Manual: STEMprep™ Tissue Dissociator (Document #10000030598), available at www.stemcell.com, or contact us to request a copy.
NOTE: For best results, avoid pinning the tissue with the rotor in the center of the STEMprep™ Sample Tube. Ensure total sample volume containing tissue does not exceed the "MAX" fill line on the tube.
3. Select and run the "Mouse Lung" dissociation protocol on the STEMprep™ Tissue Dissociator.
NOTE: If the tube will not be removed immediately after protocol completion, add a 4°C hold to preserve sample integrity.
4. After the protocol is complete, remove the STEMprep™ Sample Tube from the slot. Place a 70 µm cell strainer in a 50 mL conical tube. Pre-wet the strainer with 5 mL of EasySep™ Buffer. Carefully pour the sample over the strainer.
5. Rinse the STEMprep™ Sample Tube with 10 mL of cold EasySep™ Buffer and pour the wash over the strainer. Repeat this wash step once more. Top up the conical tube to 50 mL with EasySep™ Buffer and screw on the cap. Discard the strainer and STEMprep™ Sample Tube.
NOTE: Small tissue fragments may remain in the filter after dissociation; these typically do not affect yield. Larger tissue pieces can be gently pushed through the strainer using the rubber end of a syringe plunger.
6. Centrifuge the conical tube at 300 x g for 10 minutes at room temperature with the brake on low. After centrifugation, carefully aspirate the supernatant.
7. OPTIONAL: If desired, perform red blood cell lysis by adding 5 mL of Ammonium Chloride Solution (Catalog #07850) to the cell pellet. Thoroughly mix the cell suspension by pipetting up and down. Incubate on ice for 10 minutes. Top up sample to 50 mL with EasySep™ Buffer. Centrifuge at 300 x g for 10 minutes at room temperature with the brake on low. After centrifugation, carefully aspirate the supernatant.
NOTE: If proceeding to EasySep™, perform red blood cell lysis.
8. Resuspend cell pellet in the desired volume of EasySep™ Buffer or medium of your choice. Cells are ready for downstream use.

Data

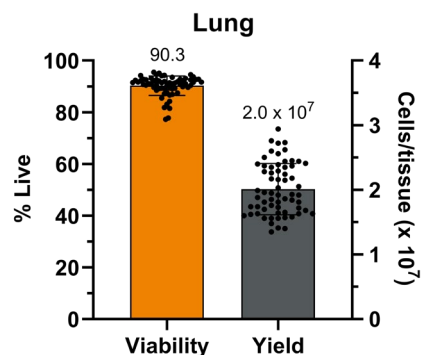


Figure 1. Cell Viability and Yield Obtained Using the STEMprep™ Mouse Lung Dissociation Kit

Lungs from healthy C57BL/6 mice were dissociated into single-cell suspensions using the STEMprep™ Mouse Lung Dissociation Kit on the STEMprep™ Tissue Dissociator. Following STEMprep™ processing, viability and yield of single-cell suspensions were assessed by flow cytometry. Data are presented as mean \pm SD.

Related Products

For more information about STEMprep™ kits and protocols, visit www.stemcell.com/stemprep, contact us at techsupport@stemcell.com.

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