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Mechanical liver dissociation



In 1 collection

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ABSTRACT

This protocol details the mechanical dissociation of liver.

MATERIALS

Reagents:

- RPMI1640 medium with L-glutamine (Lonza, BE12-702F)
- **⊠** Ethanol ≥70% (v/v) TechniSolv® **VWR International Catalog #**83801.360

Equipment:

- gentleMACS Octo Dissociator with Heaters Miltenyi Biotec Catalog # 130-096-427
- Sharps container 1.5L (BD, 305624)
- 23G needle HSW HENKE-JECT (HenkeSass Wolf, 4710006025)
- GentleMACS

 C Tube Miltenyi Biotec Catalog #130-096-334

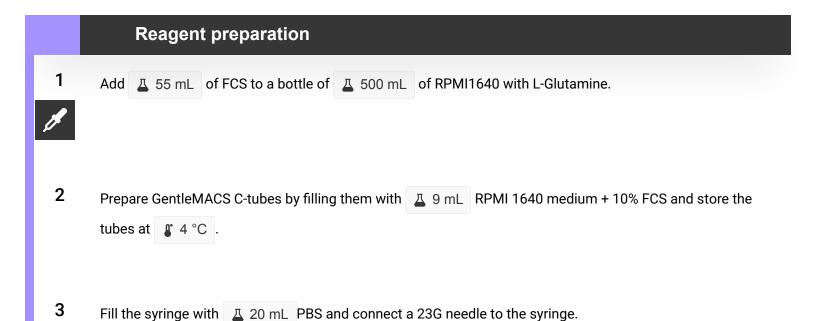
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Procedure

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Note

IMPORTANT! Proceed with liver perfusion shortly after euthanasia to prevent clotting of the blood.

Euthanize the mice by decapitation.

5 Position mouse in dorsal recumbency and pin down. Spray the abdominal and thoracic region with 70% EtOH.

6	Make a parallel incision from the caudal to cranial side starting from the mouse's abdomen until the thoracic region.
7	Gently move the intestines to the right side to get access to the liver using forceps and carefully flip the median lobe and left lobe to the cranial side to visualize the portal vein.
8	Insert the tip of the needle in the hepatic portal vein. Slowly push 4 20 mL PBS through.
	Note
	The liver will become tense and pale if the perfusion is correctly performed.
9	Carefully dissect the perfused liver and transfer into the C-tube containing the medium.
	Note
	Remove gallbladder during dissection of the mouse liver.
10	Tightly close C-tube and attach it upside down onto the GentleMACS Octo Dissociator.
11	Run program m_spleen_04 twice.
12	After termination of the program, detach C-tube from the GentleMACS Octo Dissociator and keep the tubes on ice until further processing.

13 Dispose the needle in an approved sharps container.

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