

Materials List for

# Intravital Microscopy to Study Platelet-Leukocyte-Endothelial Interactions in the Mouse Liver

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## Citation

Courson, J.A., Langlois, K.W., Lam, F.W. Intravital Microscopy to Study Platelet-Leukocyte-Endothelial Interactions in the Mouse Liver. *J. Vis. Exp.* (188), e64239, doi:10.3791/64239 (2022).

## Date Published

October 6, 2022

## DOI

10.3791/64239

## URL

jove.com/video/64239

## Materials

Name	Company	Catalog Number	Comments
<b>Surgical Supplies</b>			
2" x 2" non-woven sponges	McKesson Med. Surg	92242000	For liver isolation
#4-0 silk braided suture with needle	SOFSILK	N/A	4-0 Softsilk coated braided black, nonabsorbable: C-1 cutting needle
#4-0 silk braided suture without needle	Ethicon	N/A	4-0 Black braided silk, nonabsorbable
21 G blunt needle (0.5 inch)	SAI Infusion Technologies	B21-50	This is used to attach to the end of the tracheostomy tube to allow for connection to the ventilator. An alternative source is Instech
23 G blunt needle (0.5 inch)	SAI Infusion Technologies	B23-50	This is used for the vascular catheter to allow for connection to a syringe. An alternative source is Instech
Dissecting Scissors (Pointed Tip)	Kent Scientific	INS600393-G	Micro Dissecting Scissors; Carbide Blades; Straight; Sharp Points; 24 mm Blade Length; 4 1/2" Overall Length
McPherson-Vannas Micro Scissors (Vannas)	Kent Scientific	INS600124	These are useful for creating the openings in the trachea and vessels
Polyethylene tubing 10	Instech	BTPE-10	This is used to make the intravascular portion of the catheter. An alternative source is BD Intramedic
Polyethylene tubing 50	Instech	BTPE-50	This is used to make the extravascular portion of the catheter. An alternative source is BD Intramedic
Polyethylene tubing 90	Instech	BTPE-90	This is used to make the tracheostomy tube. An alternative source is BD Intramedic
USP grade sterile normal saline	Covidien	8881570121	Hospira 0.0% Sodium Chloride Injection, USP
<b>Microscopy Supplies</b>			

Isoflurane delivery system and ventilator	Kent Scientific	Somnosuite	Combination rodent ventilator and volatile anesthetic delivery system
Foam spacer for warming pad during microscopy	N/A	N/A	This spacer should be cut from high quality foam, should fit around the liver microscope tray and specific height dimensions are dependent upon the microscope system
Laser scanning confocal microscope system with resonance head scanner	Olympus	FV3000	Although we describe the use of an Olympus FV3000 using a resonance head scanner, this protocol will work with most imaging systems
Liver Microscope Tray	N/A	N/A	The liver microscope tray was designed for an inverted microscope
<b>Antibodies &amp; Related Reagents</b>			
Brilliant Violet 421/anti-mouse Ly6G antibody	BioLegend	127628	3 µg/mouse. To label neutrophils
BV421/F4/80 antibody	BioLegend	123132	0.75 mg/kg. To label Kupffer cells
Dulbecco's phosphate buffered saline w/o calcium or magnesium	Gibco/ThermoFisher Scientific	14190144	Used as dialysate to remove sodium azide from antibodies
DyLight649/anti-GPIIb antibody	emfret Analytics	X649	3 µg/mouse. To label platelets
DyLight488/anti-mouse GPIIb antibody	emfret Analytics	X488	6 µg/mouse. To label platelets
Endotoxin from <i>Escherichia coli</i> serotype O111:B4	Sigma-Aldrich	L3024	5 mg/kg; Potency of endotoxin may vary from lot to lot. Therefore, the same lot should be used for a series of experiments to minimize variation due to endotoxin lot
PerCP-eFluor 710/anti-mouse P-selectin antibody	Invitrogen	46-0626-82	4 µg/mouse. To label P-selectin
Slide-a-Lyzer 7,000 MWCO cassette	Thermo Scientific	66370	Used to dialyze antibodies to remove sodium azide
Texas Red-labeled dextran	Sigma-Aldrich	T1287	~150 kDa; 250 µg/mouse
TRITC/bovine serum albumin	Sigma-Aldrich	A2289	500 µg/mouse. Dilute to a stock concentration of 50 mg/mL (5%) in normal saline. Used to label the vasculature. It may leak into the interstitial space more readily than high molecular weight dextran during inflammation