



Aug 07,  
2019

## Vascular perfusion of mice [↗](#)

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**1** Works for me [dx.doi.org/10.17504/protocols.io.36dgra6](https://doi.org/10.17504/protocols.io.36dgra6)

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### ABSTRACT

#### Summary:

This protocol describes the procedure for perfusion of mice prior to organ or tissue harvesting .

#### Diabetic Complications:



Cardiovascular



Retinopathy



Neuropathy



Nephropathy



Uropathy

### EXTERNAL LINK

<https://diacomp.org/shared/document.aspx?id=24&docType=Protocol>

### MATERIALS TEXT

Reagents and Materials	Quantity Required
Perfusion Apparatus	1
Perfusion Bottles	2
Small diameter tubing (< 1cm)	
butterfly 23 gauge $\frac{3}{4}$	1
Ketamine	3 ml
Xylazine	1 ml
Plastic Tray	1
Phosphate Buffered Saline, pH 7.4	
PFA	
Sucrose	
70% alcohol	
Scissors	
Cotton swabs	

#### 1 Preparation (see figure below for setup)

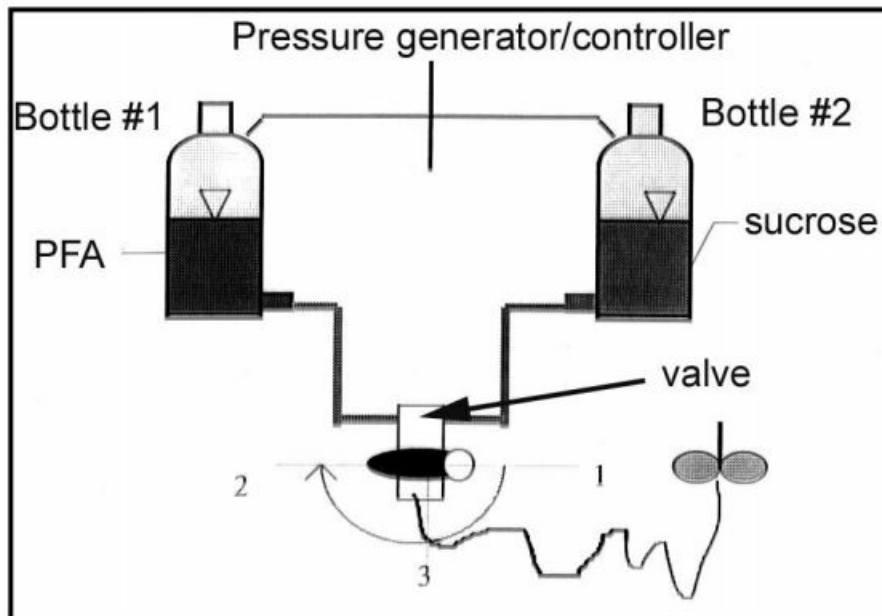
- ◆ Fix tubing (small diameter tubes, below 1 cm) to the bottles: 1 bottle for PFA, 1 bottle for sucrose
- ◆ Fill perfusion solutions in the bottles: solutions must be freshly prepared and filtered (using 0.22  $\mu$ m filter)

- ◆ Adjust perfusion pressure to 150-160 mm Hg
- ◆ Fix needle at the end of tubing after valve: butterfly 23 gauge 3/4
- ◆ **Remove any air from all tubing!!**
- ◆ Anesthetic: ketamine-xylazine mix (150/10 mg/kg=0.1 ml/20 g BW)
- ◆ Mix 3 ml ketamine 100mg/ml + 1 ml xylazine 20mg/ml + 6 ml sterile saline
- ◆ Fix the anesthetized animal in a plastic tray
- ◆ Solution #1: 4 % PFA in PBS, pH 7.4
- ◆ Solution #2: 18% Sucrose in PBS, pH 7.4

## 2 Perfusion:

- ◆ Swab mouse with 70% alcohol to wet the fur, cut open abdominal skin by a longitudinal incision
- ◆ Remove skin and gut to expose abdominal aorta and Vena cava
- ◆ Clean the aort abdominalis and vena cava from connective tissue and fat with cotton swabs
- ◆ Clamp aorta abdominalis and vena cava **in the area of the iliac arteries**
- ◆ Insert the **butterfly 23 gauge 3/4** needle in the Aorta below the renal arteries
- ◆ Immediately cut open the Vena cava with small and sharp scissors: this should allow a rapid drain of the perfusion solutions!
- ◆ Watch the needle carefully during the entire perfusion!!!
- ◆ Start perfusion for 3 minutes with PFA, pH 7.4 at 37°C
- ◆ Switch valve to Solution #2:
- ◆ Without any interruption of pressure/flow perfuse for 5 minutes with sucrose, 7.4 at 37°C
- ◆ Stop perfusion, remove butterfly
- ◆ Cut out kidneys at renal hilum for further processing
- ◆ Before starting to perfuse the next animal rinse the tubing and needle extensively

## 3 Schematic of custom-made perfusion system



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