


Version 2 ▼

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Intracardial perfusion for electrophysiology in rat V.2

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1 Works for me

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ABSTRACT

This protocol is to use for clearing blood from the brain and helping to maintain viable cells for electrophysiology experiments. No PFA or any tools that have had contact with PFA should be used. This intracranial perfusion can help maintain viability of neurons in older animals and those who have undergone treatments that damage brain cells (for example: chronic methamphetamine and Parkinson's disease like treatments).

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KEYWORDS

intracranial, perfusion, electrophysiology, rat

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GUIDELINES

The rat should be unable to feel pain, but still breathing so the neurons are as healthy and in a physiological condition close to when the animal was alive when you attempt to do electrophysiology experiments later using the brain slices.

MATERIALS TEXT

anesthesia machine and isoflurane solution
forceps
sharp surgical scissors, I prefer a very small blade but use what works for you
tweezers
needles/dissection pins to secure limbs before surgery
ronguers
bone cutters
weigh spatula with long thin end to help gently remove brain from skull
4, 60 mL syringes for the artificial cerebral spinal fluid and high sucrose solution
4, 18 gauge needles
ice bucket with ice to keep solutions cold
container (beaker for example) to hold high sucrose solution and brain once removed from skull
razor in scraper handle or rat size guillotine for decapitation
styrofoam lid (concave) or dissection pan for placing rat during surgery
hood- location for anesthesia and surgery
biohazard disposal supplies (sharps box, rat body bag, and red biohazard bags and labels)
ethanol
paper towels
PPE (gloves, lab coat, even safety glasses might be a good idea)
scale if you want to weigh the animal

SAFETY WARNINGS

Sharp surgical tools and needles, be careful and get fully trained in the surgery before attempting alone

BEFORE STARTING

Gather tools and set up the area and double check before beginning any surgical procedure.

Anesthesia

- 1 anesthetize rat with isoflurane anesthesia machine.

- 2 when rat is lying down immobile, doesn't right it's position when moved, and is breathing slowly put isoflurane nose cone on rat and test for pain with forceps toe pinch of both feet

Surgery and intracranial perfusion

- 3 open rat thoracic cavity under sternum
- 4 hold sternum back with forceps
- 5 cut diaphragm
- 6 cut sides of rib cage
- 7 small cut to open right atria
- 8 inject into heart ventricle ~ 120 mL ice cold artificial cerebral spinal fluid, then ~ 120 mL ice cold high sucrose solution

Brain extraction

- 9 decapitate rat
- 10 carefully remove skull
- 11 gently free brain from skull, cut cranial nerves when necessary

12 let brain gently release from skull into ice cold high sucrose solution

Cleanup

13 clean surgical tools carefully and let air dry

14 sharps disposed of in sharps biohazard red box

15 let blood dissolve in bleach in flask in hood to decontaminate

16 dispose of other biohazard material in a double bagged biohazard red bag .add sticker label.
dispose of in biohazard waste bins.

17 clean surfaces in hood with ethanol and paper towels

18 bring rat body in body bag with labeled tag to BSB 099 cold room

19 bring cage and accessories with it to BSB dirty cage area for washing 0 μ L