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Mouse Perfusion

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This protocol is used to perfuse mice and isolate their brain for cryosectioning.

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1 Sedate mice with 120mg/kg Euthanyl then open their chest cavity to expose their heart. 2m

2 Insert the needle tip into the left ventricle and cut the right atria. Begin slowly injecting 10mL 1X phosphate buffered saline (PBS) over 5 minutes. 5m

3 Once all of the 1X PBS has been perfused, begin injecting 10mL 4% paraformaldehyde or 10% formalin over 5 minutes. 5m

4 Extract brain and incubate them in either 4% paraformaldehyde or 10% formalin for 72 hours at 3d

4°C.

5 Incubate brain in 10% sucrose for 24 hours at 4°C. 1d

6 Incubate brain in 20% sucrose for 24 hours at 4°C. 1d

7 Incubate brain in 30% sucrose for 24 hours at 4°C. 1d

8 Decant brain from sucrose solution and flash freeze in isopentane at -35 to -45°C for 1 minute. 1m

9 Cryo-section brain at 40µm. 30m

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