Tissue acquisition

Following euthanasia by terminal anesthesia with isoflurane followed by decapitation, the brains were quickly removed and divided into two hemispheres. One hemisphere was dissected into distinct brain areas including cortex and snap frozen before long term storage at -80 °C. The second hemisphere was immersion fixed in 4% paraformaldehyde for at least 24 hours before being placed in 30% sucrose for 24-72 hours and then moved to 1xPBS.