Additional details about collection, processing, and analysis of brain samples in the Genotype-Tissue Expression (GTEx) project

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Brain tissues obtained at autopsy are processed immediately according to standardized protocols (http://biospecimens.cancer.gov/resources/sops/GTEx\_SOPs/II/A/GTEx%20Tissue%20Procurement%20SOP%20(PR-0004%20v1.2.1).pdf). The brain and upper spinal cord segment are removed from the calvarium at the Biospecimen Source Site (BSS). The cerebral cortex and cerebellum are sampled at the BSS, and the remaining whole brain is packed in wet ice for immediate shipment. Upon receipt at the University of Miami Brain Bank, the brain is weighed and photographed to record structural landmarks and any surface abnormalities. The brain is sectioned coronally into 1.5 cm slices using a plexiglass guide plane. Once sampled as described below, both hemispheres of the brain are banked frozen for future anatomic sectioning and region-of-interest dissections.

Any attempt to define the patterns of gene expression within the adult human brain requires precise sampling of multiple brain areas with precision and reproducibility to minimize sampling error. Select regions of the brain, including the neocortex (frontal, cingulate, temporal, motor, parietal and occipital), the hippocampus including the entorhinal cortex, caudate, thalamus, midbrain at the level of the substantia nigra, periventricular white matter, the cerebellar hemisphere, pons and the medulla are sampled, transferred to pathology cassettes, and fixed in buffered formalin for paraffin embedding and neuropathological review. The remaining coronal slices are rapidly frozen by immersion in 2-methylbutane that is pre-cooled at -40o C. Frozen coronal slices are placed in air-tight freezer storage bags, heat sealed, barcoded, and stored at -80o C.

For the GTEx project, in addition to resampling the cerebral cortex and cerebellum from the frozen brain, an additional nine anatomic regions are dissected and shipped frozen to the GTEx Laboratory, Data Analysis, and Coordinating Center. These regions include the amygdala, anterior cingulate cortex (BA24), caudate (basal ganglia), hippocampus, hypothalamus, nucleus accumbens (basal ganglia), putamen (basal ganglia), C-1 spinal cord, and pituitary gland. Those of sufficient quality undergo RNA-Seq analysis. In the future, select brain regions can be sampled from cryopreserved frozen slices in standard 200 mg punches or block specimens and shipped to NIH-approved end-users.