Sample Collection and Processing for RNA analysis

In the BRAINcode project, 190 high-quality, frozen postmortem human brain samples identified from Banner Sun Health Institute, Brain Tissue Center at Massachusetts General Hospital, Harvard Brain Tissue Resource Center at McLean Hospital, University of Kentucky ADC Tissue Bank, the University of Maryland Brain and Tissue Bank, Pacific Northwest Dementia and Aging Neuropathology Group (PANDA) at University of Washington Medicine Center, and Neurological Foundation of New Zealand Human Brain Bank, were collected. A subset of these samples were previously analyzed in Dong et al., 2018, "Enhancers active in dopamine neurons are a primary link between genetic variation and neuropsychiatric disease" published in Nature Neuroscience. Detailed quality measures and demographic characteristics of these high-quality, frozen postmortem samples were obtained. Median RNA integrity numbers (RIN) were 7.7, 7.4, and 7.3 for substantia nigra samples (used to laser-capture dopamine neurons), temporal cortex (used to laser-capture temporal cortex pyramidal neurons), and motor cortex samples (used to laser-capture Betz cells) indicating high RNA quality. Median post-mortem intervals were exceptionally short with 3 hours for substantia nigra, 3 hours for temporal cortex, and 13 hours for motor cortex samples further consistent with the highest sample quality. The 190 brain samples are composed of 102 samples from healthy control ("HC") subjects, 27 from incidental Lewy body cases ("ILB"), 18 from Parkinson's disease cases ("PD"), and 43 from Alzheimer's disease cases ("AD"). Healthy control subjects are defined with the following stringent inclusion and exclusion criteria. Inclusion criteria: (1) absence of clinical or neuropathological diagnosis of a neurodegenerative disease e.g., Parkinson's disease according to the UKPDBB criteria66, Alzheimer's disease according to NIA-Reagan criteria67, dementia with Lewy bodies by revised consensus criteria68. (2) PMI $\leq$ 48 hours; (3) RIN69 $\geq$ 6.0 by Agilent Bioanalyzer (good RNA integrity); (4) visible ribosomal peaks on the electropherogram. Exclusion criteria were: (1) a primary intracerebral event as the cause of death; (2) brain tumor (except incidental meningiomas); (3) systemic disorders likely to cause chronic brain damage. Incidental Lewy body cases are those not meeting clinical diagnostic criteria for PD or other neurodegenerative diseases but found with Lewy body inclusion at autopsy. ILB is widely considered a preclinical stage of PD70, providing a unique opportunity to investigate preclinical molecular changes of PD. Seven non-brain tissue samples were included as controls, including four samples of peripheral blood mononuclear cell (PBMC) and three fibroblasts (FB), provided by Harvard Biomarker Study and Coriell Institute.