

1. Download the following files from GTEx:

- 1) GTEx_Analysis_v6_RNA-seq_RNA-SeQCv1.1.8_gene_reads.gct
- 2) GTEx_Data_V6_Annotations_SampleAttributesDS.txt
- 3) GTEx_Data_V6_Annotations_SubjectPhenotypesDS.txt

2. Converted file:

GTEx_Analysis_v6_RNA-seq_RNA-SeQCv1.1.8_gene_reads.gct to .txt

3. Get metadata (from the annotation file).

4. GET:

Samples by age (metadata).

Check if RNA-seq data is available.

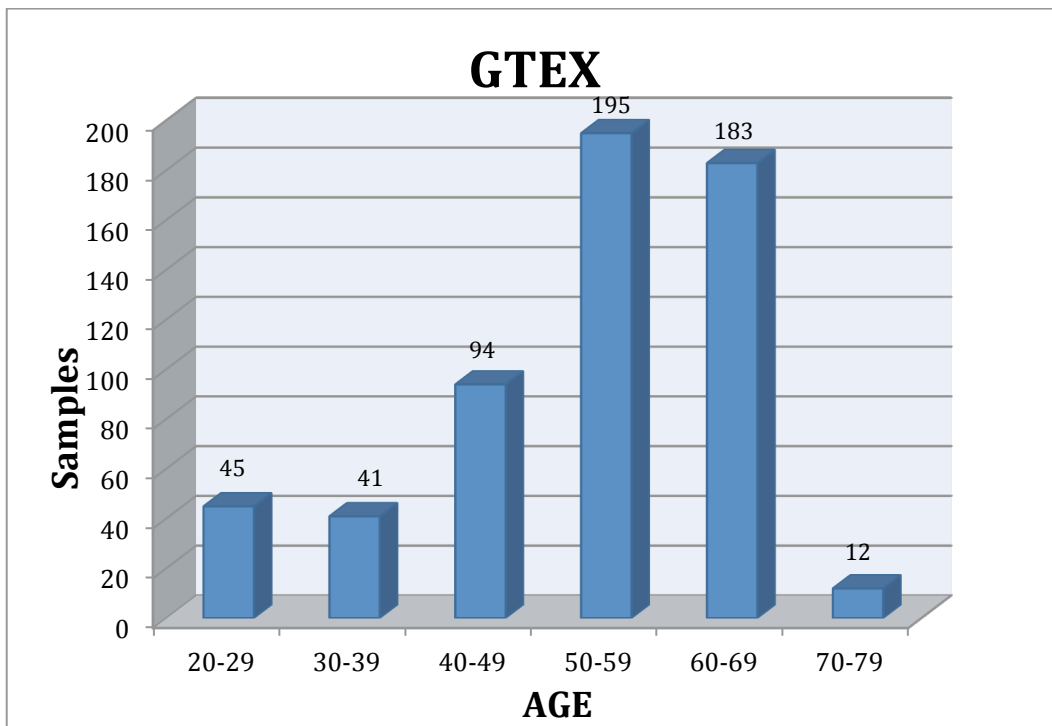
Write list -> samples of interest.

Contains: ids for 8 brain samples in the age range 20-29s and 30-39 marked as controls.

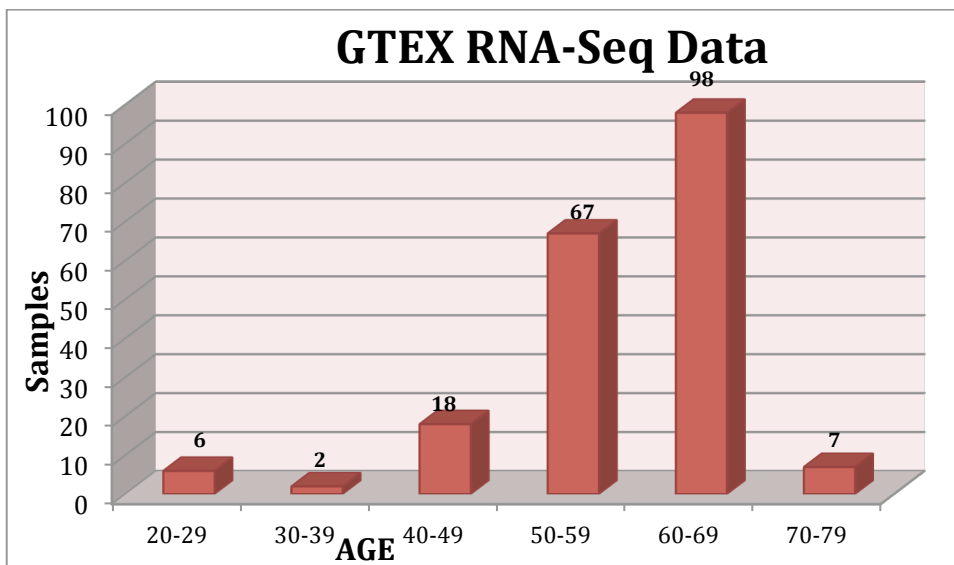
And 8 samples from age range 50-59s randomly selected.

Generate a random sequence of 8 numbers ranging from 1 to 67 and used those 8 samples.(matlab)

SAMPID	C/P	Age	Gender	SMCENTER	SMTS	SMTSD	SMTSTPTREF	SMNABTCH	SMNABTCHT
GTEx-11ZUS-0011-R10b-SM-5BC79	Control	30-39	Male	C1, A1	Brain	Brain - Frontal Cortex (BA9)	Actual Death	BP-43234	RNA isolation_PAXgene Tissue miRNA
GTEx-12126-0011-R10b-SM-5BC6T	Control	20-29	Male	C1, A1	Brain	Brain - Frontal Cortex (BA9)	Actual Death	BP-43234	RNA isolation_PAXgene Tissue miRNA
GTEx-12WSE-0011-R10b-SM-5P9JV	Control	20-29	Male	C1, A1	Brain	Brain - Frontal Cortex (BA9)	Actual Death	BP-43982	RNA isolation_PAXgene Tissue miRNA
GTEx-QDT8-0011-R10A-SM-32PKG	Control	30-39	Female	C1, A1	Brain	Brain - Frontal Cortex (BA9)	Actual Death	BP-20394	RNA isolation_QIAGEN miRNeasy
GTEx-R55E-0011-R11A-SM-2TC6I	Control	20-29	Male	C1, A1	Brain	Brain - Cerebellar Hemisphere	Actual Death	BP-21968	RNA isolation_Trizol Manual (Cell Pellet)
GTEx-T2IS-0011-R11A-SM-32QPC	Control	20-29	Female	C1, A1	Brain	Brain - Cerebellar Hemisphere	Actual Death	BP-24010	RNA isolation_QIAGEN miRNeasy
GTEx-T5JC-0011-R10A-SM-32PM2	Control	20-29	Male	C1, A1	Brain	Brain - Frontal Cortex (BA9)	Actual Death	BP-24011	RNA isolation_QIAGEN miRNeasy
GTEx-WHSE-0011-R11A-SM-3P5YY	Control	20-29	Female	C1, A1	Brain	Brain - Cerebellar Hemisphere	Actual Death	BP-28281	RNA isolation_PAXgene Tissue miRNA
GTEx-13VXU-0011-R11b-SM-5O9DJ	Perturbation	50-59	Male	B1, A1	Brain	Brain - Hemisphere	Actual Death	BP-47673	RNA isolation_PAXgene Tissue miRNA
GTEx-14A5I-2826-SM-5SIBF	Perturbation	50-59	Female	C1	Brain	Brain - Cerebellum	Actual Death	BP-47656	RNA Extraction from Paxgene-derived Lysate Plate Based
GTEx-13OVJ-0011-R10b-SM-5L3HT	Perturbation	50-59	Female	B1, A1	Brain	Brain - Frontal Cortex (BA9)	Actual Death	BP-47594	RNA isolation_PAXgene Tissue miRNA
GTEx-12WSC-0011-R10a-SM-5GU57	Perturbation	50-59	Male	C1, A1	Brain	Brain - Frontal Cortex (BA9)	Actual Death	BP-43747	RNA isolation_PAXgene Tissue miRNA
GTEx-13FHP-3026-SM-5IJB5	Perturbation	50-59	Male	C1	Brain	Brain - Cortex	Actual Death	BP-46978	RNA Extraction from Paxgene-derived Lysate Plate Based
GTEx-ZE7O-0011-R10a-SM-57WAZ	Perturbation	50-59	Female	B1, A1	Brain	Brain - Frontal Cortex (BA9)	Presumed Death	BP-41252	RNA isolation_PAXgene Tissue miRNA
GTEx-131XW-0011-R10a-SM-5DUVA	Perturbation	50-59	Female	C1, A1	Brain	Brain - Frontal Cortex (BA9)	Actual Death	BP-44932	RNA isolation_PAXgene Tissue miRNA



Graph 1: Shows the age range distribution for all the 570 GTEX donors



Graph 2: Shows the number of unique Brain donor samples with RNA-Seq data.

5. Get RNA-Seq data

Script: GTEXRseq.py

6. Clean and format list - *Characteristic Direction example.*

File: RNAseqbrain-out.txt

7. Characteristic direction.

Script: Gtex-ChrDir.py

8. Results and files:

20vs50.txt : List of the 16 samples ids for analyzes.

RNAseqbrain-out.txt : Formatted ChrDir input file.

Gtex_Brain.xlsx : Tables, graphs, metadata for Gtex Brain samples.

GtexBrain-20vs50-chdir-full.txt: Complete ChrDir result gene list.

GtexBrain-20-30svs50s-chdir_DOWN.txt: All down genes.

GtexBrain-20-30svs50s-chdir_UP.txt : All up genes.

GtexBrain-20-30svs50s-chdir_NoChange.txt : genes with no change in expression.

GtexBrain-20-30svs50s-chdir_5000Down.txt: First 5000 down genes.

GtexBrain-20-30svs50s-chdir_5000Up.txt: First 5000 up genes.