

File S1

Code used for RNAseq analysis.

```
# Sickle (version 1.200)
# Run on each of the four FASTQ files: A3-control (A3C), A3-nicotine (A3N), A4-control (A4C), and A4-nicotine (A4N)

sickle se -f A3C.fastq.gz -t sanger -o A3C.sickle.fastq -q 30 -l 30 -n
gzip A3C.sickle.fastq

# TopHat (version 2.0.9)
# Run on each of the four trimmed FASTQ files

tophat2 -p 12 -G genes.gtf -o ./Assembly_A3C/ --no-novel-juncs --library-type
fr-unstranded genome A3C.sickle.fastq.gz

# Cufflinks (version 2.1.1)
# Run on all four TopHat assemblies simultaneously

cuffdiff -N -o ./CuffDiff_Output/ -b genome.fa -p 12 -u genes.gtf
-L A3C,A3N,A4C,A4N ./Assembly_A3C/accepted_hits.bam ./Assembly_A3N/accepted_hits.bam
./Assembly_A4C/accepted_hits.bam ./Assembly_A4N/accepted_hits.bam
```