

Cufflinks and RSEM command line

Cufflinks 2.1.1[1] was applied to generate isoform expression as one of the baseline to compare with Net-RSTQ using the following command:

```
./cufflinks -p 4 -F 0 -G hg19RefSeq.gtf -o x.bam.
```

RSEM1.2.20[2] was applied to generate isoform expression as another baseline to compare with Net-RSTQ in simulation and qRT-PCR studies using the following command:

```
./rsem-calculate-expression --paired-end --bowtie2 --bowtie2-path bowtie2/ -p 2 x1.fastq x2.fastq hg19RefSeq x.
```

References

1. Trapnell C, Williams BA, Pertea G, Mortazavi A, Kwan G, et al. (2010) Transcript assembly and quantification by RNA-Seq reveals unannotated transcripts and isoform switching during cell differentiation. *Nat Biotech* 28: 511-515.
2. Li B, Dewey CN (2011) RSEM: accurate transcript quantification from RNA-Seq data with or without a reference genome. *BMC Bioinformatics* 12: 323.