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Updated: 15 February 2019

Checking the Quality of your Trinity assemblage

(https://github.com/trinityrnaseq/trinityrnaseq/wiki/Transcriptome-Assembly-Quality-Assessment)

Assessing the Read Content of the Transcriptome Assembly

1. Move into the Trinity out directory folder where your newly created Trinity.fasta file has been created and run bowtie2:

```
> bowtie2-build Trinity.fasta Trinity.fasta
```

2. Make sure to change the 1.fq and 2.fq names appropriately.

```
> sudo bowtie2 -p 10 -q --no-unal -k 20 -x Trinity.fasta -1
SRR5059294_1.fastq.PwU.qtrim.fq -2 SRR5059294_2.fastq.PwU.qtrim.fq
2>align_stats.txt| samtools view -@10 -Sb -o bowtie2.bam
> cat 2>&1 align_stats.txt
```

Please note: If you have multiple reads separate with only a comma, such as:

```
> sudo bowtie2 -p 10 -q --no-unal -k 20 -x Trinity.fasta -1
SRR5666978_1.fastq.PwU.qtrim.fq,SRR5666989_1.fastq.PwU.qtrim.fq,SRR566
7091_1.fastq.PwU.qtrim.fq -2
SRR5666978_2.fastq.PwU.qtrim.fq,SRR5666989_2.fastq.PwU.qtrim.fq,SRR566
7091_2.fastq.PwU.qtrim.fq 2>align_stats.txt| samtools view -@10 -Sb -o
bowtie2.bam
```

3. A typical Trinity transcriptome assembly will have most reads mapping back to the assembly. Your output file should look like this:

```
27161346 reads; of these:
  27161346 (100.00%) were paired; of these:
    802441 (2.95%) aligned concordantly 0 times
    5258238 (19.36%) aligned concordantly exactly 1 time
    21100667 (77.69%) aligned concordantly >1 times
    ----
    802441 pairs aligned concordantly 0 times; of these:
      31198 (3.89%) aligned discordantly 1 time
    ----
    771243 pairs aligned 0 times concordantly or discordantly; of these:
      1542486 mates make up the pairs; of these:
        990222 (64.20%) aligned 0 times
        111391 (7.22%) aligned exactly 1 time
        440873 (28.58%) aligned >1 times
98.18% overall alignment rate
```