**1.7514-OGS2.0\_allcd (OGS 2.0 annotation)**

export PATH=$HOME/bowtie2-2.2.1:$HOME/tophat-2.0.11.Linux\_x86\_64:$HOME/samtools-0.1.19:$HOME/cufflinks-2.2.0.Linux\_x86\_64:$PATH

echo$PATH

cd /mnt/lustre/jlynch42

cuffdiff -p 16 -o NvAp/**7514-OGS2.0\_allcd** -L A,P -u NvAp/7414all3ann20cm/merged.gtf NvAp/7414\_1A\_Annot20th/accepted\_hits.bam,NvAp/7414\_2A\_Annot20ssth/accepted\_hits.bam,NvAp/7414\_3A\_Annot20ssth/accepted\_hits.bam NvAp/7414\_1p\_Annot20ssth/accepted\_hits.bam,NvAp/7414\_2P\_Annot20ssth/accepted\_hits.bam,NvAp/7414\_3p\_Annot20ssth/accepted\_hits.bam

cuffmerge -p 16 -o NvAp/7414all3ann20cm -g nv2clean.gff -s assnv1.fa NvAp/assembliesall3anno20.txt

./NvAp/7414\_1A\_Annot20sscl/transcripts.gtf

./NvAp/7414\_3A\_Annot20sscl/transcripts.gtf

./NvAp/7414\_2A\_Annot20sscl/transcripts.gtf

./NvAp/7414\_1p\_Annot20sscl/transcripts.gtf

./NvAp/7414\_2P\_Annot20sscl/transcripts.gtf

./NvAp/7414\_3p\_Annot20sscl/transcripts.gtf

cufflinks -p 16 -o NvAp/7414\_3p\_Annot20sscl -G nv2clean.gff NvAp/7414\_3p\_Annot20ssth/accepted\_hits.bam

cufflinks -p 16 -o NvAp/7414\_3A\_Annot20sscl -G nv2clean.gff NvAp/7414\_3A\_Annot20ssth/accepted\_hits.bam

cufflinks -p 16 -o NvAp/7414\_2P\_Annot20sscl -G nv2clean.gff NvAp/7414\_2P\_Annot20ssth/accepted\_hits.bam

cufflinks -p 16 -o NvAp/7414\_2A\_Annot20sscl -G nv2clean.gff NvAp/7414\_2A\_Annot20ssth/accepted\_hits.bam

cufflinks -p 16 -o NvAp/7414\_1p\_Annot20sscl -G nv2clean.gff NvAp/7414\_1p\_Annot20ssth/accepted\_hits.bam

cufflinks -p 16 -o NvAp/7414\_1A\_Annot20sscl -G nv2clean.gff NvAp/7414\_1A\_Annot20th/accepted\_hits.bam

tophat2 -p 16 -r 50 --library-type fr-secondstrand -I 10000 -o NvAp/7414\_3p\_Annot20ssth -G nv2clean.gff ass1nvbowtie2 NvAp/3P\_GCCAAT\_L001\_R1\_001.fastq NvAp/3P\_GCCAAT\_L001\_R2\_001.fastq

tophat2 -p 16 -r 50 --library-type fr-secondstrand -I 10000 -o NvAp/7414\_3A\_Annot20ssth -G nv2clean.gff ass1nvbowtie2 NvAp/3A\_ACAGTG\_L001\_R1\_001.fastq NvAp/3A\_ACAGTG\_L001\_R2\_001.fastq

tophat2 -p 16 -r 50 -I 10000 --library-type fr-secondstrand -o NvAp/7414\_2P\_Annot20ssth -G nv2clean.gff ass1nvbowtie2 NvAp/2P\_TGACCA\_L001\_R1\_001.fastq NvAp/2P\_TGACCA\_L001\_R2\_001.fastq

tophat2 -p 16 -r 50 --library-type fr-secondstrand -I 10000 -o NvAp/7414\_2A\_Annot20ssth -G nv2clean.gff ass1nvbowtie2 NvAp/2A\_TTAGGC\_L001\_R1\_001.fastq NvAp/2A\_TTAGGC\_L001\_R2\_001.fastq

tophat2 -p 16 -r 50 -I 10000 --library-type fr-secondstrand -o NvAp/7414\_1p\_Annot20ssth -G nv2clean.gff ass1nvbowtie2 NvAp/1P\_CGATGT\_L001\_R1\_001.fastq NvAp/1P\_CGATGT\_L001\_R2\_001.fastq

tophat2 -p 16 -r 50 --library-type fr-secondstrand -I 10000 -o NvAp/7414\_1A\_Annot20th -G nv2clean.gff ass1nvbowtie2 NvAp/1A\_ATCACG\_L001\_R1\_001.fastq NvAp/1A\_ATCACG\_L001\_R2\_001.fastq

**2. 7514-OGS2.0\_2+3cd (OGS 2.0 annotation)**

cuffdiff -p 16 -o NvAp/**7514-OGS2.0\_2+3cd** -L A,P -u NvAp/7414all3ann20cm/merged.gtf NvAp/7414\_2A\_Annot20ssth/accepted\_hits.bam,NvAp/7414\_3A\_Annot20ssth/accepted\_hits.bam NvAp/7414\_2P\_Annot20ssth/accepted\_hits.bam,NvAp/7414\_3p\_Annot20ssth/accepted\_hits.bam

cuffmerge -p 16 -o NvAp/7414all3ann20cm -g nv2clean.gff -s assnv1.fa NvAp/assembliesall3anno20.txt

./NvAp/7414\_1A\_Annot20sscl/transcripts.gtf

./NvAp/7414\_3A\_Annot20sscl/transcripts.gtf

./NvAp/7414\_2A\_Annot20sscl/transcripts.gtf

./NvAp/7414\_1p\_Annot20sscl/transcripts.gtf

./NvAp/7414\_2P\_Annot20sscl/transcripts.gtf

./NvAp/7414\_3p\_Annot20sscl/transcripts.gtf

cufflinks -p 16 -o NvAp/7414\_3p\_Annot20sscl -G nv2clean.gff NvAp/7414\_3p\_Annot20ssth/accepted\_hits.bam

cufflinks -p 16 -o NvAp/7414\_3A\_Annot20sscl -G nv2clean.gff NvAp/7414\_3A\_Annot20ssth/accepted\_hits.bam

cufflinks -p 16 -o NvAp/7414\_2P\_Annot20sscl -G nv2clean.gff NvAp/7414\_2P\_Annot20ssth/accepted\_hits.bam

cufflinks -p 16 -o NvAp/7414\_2A\_Annot20sscl -G nv2clean.gff NvAp/7414\_2A\_Annot20ssth/accepted\_hits.bam

cufflinks -p 16 -o NvAp/7414\_1p\_Annot20sscl -G nv2clean.gff NvAp/7414\_1p\_Annot20ssth/accepted\_hits.bam

cufflinks -p 16 -o NvAp/7414\_1A\_Annot20sscl -G nv2clean.gff NvAp/7414\_1A\_Annot20th/accepted\_hits.bam

tophat2 -p 16 -r 50 --library-type fr-secondstrand -I 10000 -o NvAp/7414\_3p\_Annot20ssth -G nv2clean.gff ass1nvbowtie2 NvAp/3P\_GCCAAT\_L001\_R1\_001.fastq NvAp/3P\_GCCAAT\_L001\_R2\_001.fastq

tophat2 -p 16 -r 50 --library-type fr-secondstrand -I 10000 -o NvAp/7414\_3A\_Annot20ssth -G nv2clean.gff ass1nvbowtie2 NvAp/3A\_ACAGTG\_L001\_R1\_001.fastq NvAp/3A\_ACAGTG\_L001\_R2\_001.fastq

tophat2 -p 16 -r 50 -I 10000 --library-type fr-secondstrand -o NvAp/7414\_2P\_Annot20ssth -G nv2clean.gff ass1nvbowtie2 NvAp/2P\_TGACCA\_L001\_R1\_001.fastq NvAp/2P\_TGACCA\_L001\_R2\_001.fastq

tophat2 -p 16 -r 50 --library-type fr-secondstrand -I 10000 -o NvAp/7414\_2A\_Annot20ssth -G nv2clean.gff ass1nvbowtie2 NvAp/2A\_TTAGGC\_L001\_R1\_001.fastq NvAp/2A\_TTAGGC\_L001\_R2\_001.fastq

tophat2 -p 16 -r 50 -I 10000 --library-type fr-secondstrand -o NvAp/7414\_1p\_Annot20ssth -G nv2clean.gff ass1nvbowtie2 NvAp/1P\_CGATGT\_L001\_R1\_001.fastq NvAp/1P\_CGATGT\_L001\_R2\_001.fastq

tophat2 -p 16 -r 50 --library-type fr-secondstrand -I 10000 -o NvAp/7414\_1A\_Annot20th -G nv2clean.gff ass1nvbowtie2 NvAp/1A\_ATCACG\_L001\_R1\_001.fastq NvAp/1A\_ATCACG\_L001\_R2\_001.fastq