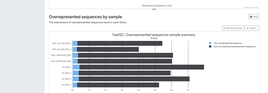
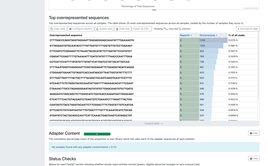
multiqc
v1.33
General Stats
Duplicodemers Metrics
Bicondemers: reference summary
Bicondemers: read composition
FastQC
FastQC (raw)
Sequence Counts
Sequence Quality Histograms
Per Sequence Quality Scores
Per Base Sequence Content
Per Sequence GC Content
Per Base N Content
Sequence Length Distribution
Sequence Duplication Levels
Overrepresented sequences by sample
Adapter Content
Status Checks
Cutadapt
Filtered Reads
Trimmed Sequence Lengths
FastQC (trimmed)
Sequence Counts
Sequence Quality Histograms
Per Sequence Quality Scores
Per Base Sequence Content
Per Sequence GC Content
Per Base N Content
Sequence Length Distribution
Sequence Duplication Levels
Overrepresented sequences by sample
Top overrepresented sequences
Adapter Content
Status Checks
Software Versions
Workflow Parameters
FastQC (filtered)
Sequence Counts
Sequence Quality Histograms
Per Sequence Quality Scores
Per Base Sequence Content
Per Sequence GC Content
Per Base N Content
Sequence Length Distribution
Sequence Duplication Levels
Overrepresented sequences by sample
Top overrepresented sequences
Adapter Content
Status Checks
DuDeMux
Picard
QualiMap
Depends Logic of reads
Mean Coverage Profile
Bases
Read Coverage
Insert Sizes
Read Duplication
Junction Saturation
Mapped Reads NGI
GC distribution
Mismatches
Species
Species Confirmation
Mapped Reads
Organism Percentage of Host
Organism Percentage of Host
Cutadapt
Summary Statistics
Adapters Counts
Ophidia
Ophidia Confirmation
Ophidia Confirmation
Ophidia Confirmation



Overrepresented sequences by sample

The total amount of overrepresented sequences found in each library.



Top overrepresented sequences

The overrepresented sequences across all samples. The table shows (i) most overrepresented sequences across all samples, ordered by the number of samples they occur in.

Overrepresented sequence	Sample	Percentage	% of all reads
GTTTAGCCAGCTAGATTGAGGCTTGAGAGGGAGCAAGTTTTAAAAATG	S1	1.048	0.007%
ATTAGGAATACTACAGCAGCTTTGTTAGTTTTTTTGTTGTTTTTTTT	S1	1.048	0.006%
GTTGAATTCCGAGGGTTTCTCAAGCAGCAGCTGTTTAGTGTGTCGTT	S1	843	0.005%
GTTTAGCCAGCTAGATTGAGGCTTGAGAGGGAGCAAGTTTTAAAAAT	S1	843	0.005%
GTTGTTGGTTTGTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTT	S1	843	0.005%
GTTTAATTAAAATCCAAGAAAAAT TCAATTAAAAAAAAAAAAAAAAA	S1	843	0.005%
GTTTAGCCAGCTAGATTGAGGCTTGAGAGGGAGCAAGTTTTAAAAATGAAGG	S1	843	0.005%
GTTTAGCCAGCTAGATTGAGGCTTGAGAGGGAGCAAGTTTTAAAAATGAAGGG	S1	843	0.005%
GTTTAGCCAGCTAGATTGAGGCTTGAGAGGGAGCAAGTTTTAAAAATGAAGGGG	S1	843	0.005%
ATAAAATCTTGTAGGGAGGGAGTTTTGGGGGTTTTTGGGGTTTTTGATGTG	S1	418	0.002%
ATAAAATCTTGTAGGGAGGGAGTTTTGGGGGTTTTTGGGGTTTTTGATGTGG	S1	418	0.002%
ATAAAATCTTGTAGGGAGGGAGTTTTGGGGGTTTTTGGGGTTTTTGATGTGGT	S1	418	0.002%
GTTTAGCCAGCTAGATTGAGGCTTGAGAGGGAGCAAGTTTTAAAAATGAAGGGGTC	S1	418	0.002%
GTTTAGCCAGCTAGATTGAGGCTTGAGAGGGAGCAAGTTTTAAAAATGAAGGGGTCG	S1	418	0.002%
GTTTAGCCAGCTAGATTGAGGCTTGAGAGGGAGCAAGTTTTAAAAATGAAGGGGTCGG	S1	418	0.002%

Showing 1 to 20 of 20 entries

Adapter Content

The cumulative percentage count of the proportion of your library which has seen each of the adapter sequences at each position.

No samples found with any adapter contamination > 0.1%

Status Checks

Status for each FastQC section showing whether results seem entirely normal (green), slightly abnormal (orange) or very unusual (red).

BBTools

Kmer counting, assembly, alignment, and statistics tools for DNA/RNA sequencing reads. See http://jgi.doe.gov/data-and-tools/bbtools/.

BBSplit alignment statistics

Statistics on how many reads mapped to which reference genome.

BBSplit Statistics

Statistics on how many reads mapped to which reference genome.

Sample Name	human % mapping	human % binning	primary % binning
MAP1_AN_DNA_MAP1	0.0%	100%	75%
MAP1_UNMAPPED_MAP1		0%	25%
MAP1_UNMAPPED_MAP1	0.0%	87%	75%
MT_MAP1		13%	25%
MT_MAP1	0.0%	100%	100%

FastQC (filtered)

This section of the report shows FastQC results after a preprocessor removal BBDuk2 and/or Cutadapt step. See Processing-FastQ-data-prior-to-processing-Reads.

Sequence Counts

Sequence counts for each sample. Duplicate read counts are an estimate only.

Sequence Quality Histograms

The mean quality value across each base position in the read.

Per Sequence Quality Scores

The number of reads with average quality scores. Shows if a subset of reads has poor quality.

Per Base Sequence Content

The proportion of each base position for an average sample.

Click a sample name to see a line plot for that dataset.

Rollover for sample name

Per Sequence GC Content

The average GC content of reads. Normal random library typically have a roughly normal distribution of GC content.

Per Base N Content

The percentage of base calls at each position for which an N was called.

Sequence Length Distribution

The distribution of fragment sizes (read lengths) found. See the FastQC help.

Sequence Duplication Levels

The relative level of duplication found for every sequence.

Overrepresented sequences by sample

The total amount of overrepresented sequences found in each library.

Top overrepresented sequences

The overrepresented sequences across all samples. The table shows (i) most overrepresented sequences across all samples, ordered by the number of samples they occur in.

Overrepresented sequence	Sample	Percentage	% of all reads
GTTTAGCCAGCTAGATTGAGGCTTGAGAGGGAGCAAGTTTTAAAAATG	S1	1.048	0.007%
ATTAGGAATACTACAGCAGCTTTGTTAGTTTTTTTGTTGTTTTTTTT	S1	1.048	0.006%
GTTGAATTCCGAGGGTTTCTCAAGCAGCAGCTGTTTAGTGTGTCGTT	S1	843	0.005%
GTTTAGCCAGCTAGATTGAGGCTTGAGAGGGAGCAAGTTTTAAAAAT	S1	843	0.005%
GTTGTTGGTTTGTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTT	S1	843	0.005%
GTTTAATTAAAATCCAAGAAAAAT TCAATTAAAAAAAAAAAAAAAAA	S1	843	0.005%
GTTTAGCCAGCTAGATTGAGGCTTGAGAGGGAGCAAGTTTTAAAAATGAAGG	S1	843	0.005%
GTTTAGCCAGCTAGATTGAGGCTTGAGAGGGAGCAAGTTTTAAAAATGAAGGG	S1	843	0.005%
GTTTAGCCAGCTAGATTGAGGCTTGAGAGGGAGCAAGTTTTAAAAATGAAGGGGTC	S1	418	0.002%
GTTTAGCCAGCTAGATTGAGGCTTGAGAGGGAGCAAGTTTTAAAAATGAAGGGGTCG	S1	418	0.002%
GTTTAGCCAGCTAGATTGAGGCTTGAGAGGGAGCAAGTTTTAAAAATGAAGGGGTCGG	S1	418	0.002%

Showing 1 to 20 of 20 entries

Adapter Content

The cumulative percentage count of the proportion of your library which has seen each of the adapter sequences at each position.

No samples found with any adapter contamination > 0.1%

Status Checks

Status for each FastQC section showing whether results seem entirely normal (green), slightly abnormal (orange) or very unusual (red).

DupRadar

DupRadar provides duplication rate quality control for NGS libraries. Highly expressed genes can be expected to have a lot of duplicate reads, but high numbers of duplicates at low read counts may indicate problems with library preparation or low sequencing depth. The plot shows the duplication rate (= 1 - number of unique reads/number of reads) as a function of the gene expression level (= number of reads mapped to the gene/gene length). See http://bioconductor.org/packages/release/bioc/html/dupRadar.html.

Picard

Picard is a collection of Java tools for analysis of sequencing data. See http://broadinstitute.github.io/picard/.

Mark Duplicates

Picard's MarkDuplicates categorizes by duplication type. Note: reads are identified - not the fate for the reads.

QualiMap

QualiMap examines sequencing alignment data and the derivatives like feature counts. See https://bitbucket.org/kokonech/qualimap. See: 10.1093/bioinformatics/btu170 10.1093/bioinformatics/btv566

Genomic origin of reads

QualiMap 2 mapped reads are originating in exonic, intronic or intergenic regions. These can be displayed as either the number or percentage of mapped reads.

Gene Coverage Profile

Gene coverage profile shows the mean coverage across the length of all mapped transcripts.

RSeQC

A modular tool kit for evaluating RNA-seq data. See http://rseqc.sourceforge.net/ DOI: 10.2202/1544-6115.1621

Read Distribution

Read Distribution calculates how mapped reads are distributed over genomic features.

Inner Distance

Inner Distance calculates the inner distance (or insert size) between two paired RNA reads. Note that this can be negative if fragments overlap.

Read Duplication

read_duplication.py calculates how many alignment positions have a certain number of reads (duplicates). Both - plot truncated at 500 occurrences and Overall.

Junction Annotation

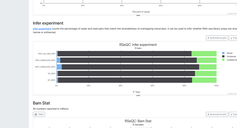
junction_annotation.py compares detected splice junctions to a reference gene model. An RNA read can be spliced ('cut') one more, each time is called a splicing event.

Junction Saturation

junction_saturation.py checks if a sufficient number of known splicing junctions that are detected in each dataset. If sequencing depth is sufficient, all (annotated) splice junctions should be

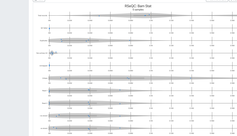
RSeQC Junction Saturation

Junction Saturation. Status: Pass



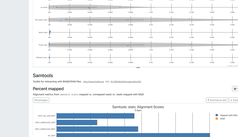
Infer experiment

Infer experiment counts the percentage of reads and read pairs that match the strandedness of overlapping transcripts. It can be used to infer whether RNA-seq library preps are strand-specific or unstranded.



Bam Stat

All mapped read statistics.

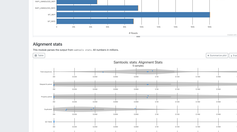


Samtools

Counts for reads mapping with BWA/SSW/HMA. See https://github.com/... for SAMtools/bamtools/BWA/SSW/HMA.

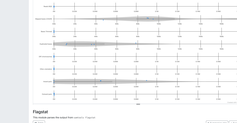
Percent mapped

Percent mapped (after filtering). Counts mapped vs. unmapped reads vs. reads mapped with MQ0



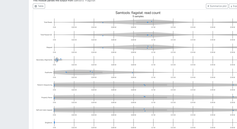
Alignment stats

The number passed into the subset flash samtools_stats: 20 reads in millions.



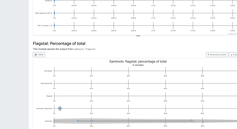
Alignment stats

The number passed into the subset flash samtools_stats: 20 reads in millions.



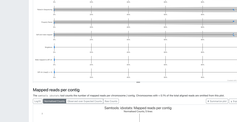
Flagstat

The number passed into the subset flash samtools_flagstat.



Flagstat: Percentage of total

The number passed into the subset flash samtools_flagstat.



Summary Statistics

Summary statistics from the STAR alignments.

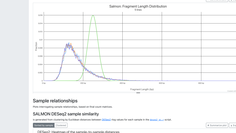
Sample Name	Total reads	Aligned	Uniq aligned	Avg. mapped len	Deletion/kb	Mismatch rate	Del rate	Ins rate	Ins len
SRR_LIB_SRR_	20.0M	99.0%	98.0%	150.0 bp	0.01	0.6%	0.0%	0.0%	1.0
SRR_LIBRARIES_SRR_	20.0M	99.0%	98.0%	150.0 bp	0.01	0.6%	0.0%	0.0%	1.0
SRP_LIBRARIES_SRR_	20.0M	99.0%	98.0%	150.0 bp	0.01	0.6%	0.0%	0.0%	1.0
MY_MAPS	20.0M	99.0%	98.0%	150.0 bp	0.01	0.6%	0.0%	0.0%	1.0
MY_MAPS	20.0M	99.0%	98.0%	150.0 bp	0.01	0.6%	0.0%	0.0%	1.0

Alignment Scores



Salmon

Quantify expression of transcripts using RNA-seq data. https://github.com/COMBINE-lab/salmon 10.5281/zenodo.5127



Sample relationships

Plots that display pairwise sample relationships, based on their count matrices.

SALMON DESeq2 sample similarity

A pairwise heatmap created by the function plot() from the matrix for each sample in the flash deseq2_pca module.



SALMON DESeq2 PCA plot

PCA plot between samples in the experiment. These scores are calculated using DESeq2's in the flash deseq2_pca module.



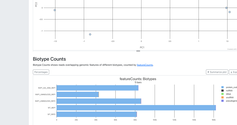
STAR_SALMON DESeq2 sample similarity

A pairwise heatmap created by the function plot() from the matrix for each sample in the flash deseq2_pca module.



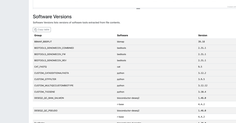
STAR_SALMON DESeq2 PCA plot

PCA plot between samples in the experiment. These scores are calculated using DESeq2's in the flash deseq2_pca module.



Biotype Counts

Biotype counts stats were calculated using genomic features of different biotypes, counted by featureCounts.



Software Versions

Software versions that versions of software tools extracted from the systems.

Group	Software	Version
ANACONDA	Anaconda	20.09
BIOCONDA_BRANDON-CHUANG	bedtools	2.29.2
BIOCONDA_BRANDON-CHUANG	bedtools	2.29.2
BIOCONDA_BRANDON-CHUANG	bedtools	2.29.2
FASTQC	fastqc	0.11.9
FASTQ_SCREEN_STEPHEN-KRUEGER	fastq_screen	0.14.0
CUTTSLAB_CUTTLETOOLS	cuttletools	0.9.3
CUTTSLAB_FASTQ-DUPLICATE-REMOVER	fastq-duplicate-remover	0.10.1.post0
CUTTSLAB_FASTQ2BAM	cuttletools	0.9.3
PEDRO_DE-LIMA_SALMON	bioconductor-deseq2	1.30.0
	r-base	4.0.3
PEDRO_DE-PRADO	bioconductor-deseq2	1.30.0
	r-base	4.0.3
TOPHAT	bioconductor-biostrings	2.58.0
CACTUS_CUTTSEQ	bcl	1.2.0
FASTQC	fastqc	0.11.9
TRIMGALORE	fastqc	0.11.2
FQ_HAT	fq	0.12.0
FQ_LINT_MITCH_MARUF	fq	0.12.0
FQ_LINT_MITCH_TRIMMING	fq	0.12.0
FQ_SUBSAMPLE	fq	0.12.0
GUNZIP_AGRITHINA_HAZRA	gunzip	1.10
GUNZIP_JAP	gunzip	1.10
PICARD_MARKDUPLICATES	picard	2.2.4
PIGZ_UNPIGZ	pigz	2.3.4
PIGZ_UNPIGZ	pigz	2.3.4
PIGZ_WWEMEYERMANY	rseqc	3.0.1
PIGZ_WWEMEYERMANY	rseqc	3.0.1
PIGZ_JUSTINCHAPMAN	rseqc	3.0.1
PIGZ_JUSTINCHAPMAN	rseqc	3.0.1
PIGZ_MAHESHBINNANI	rseqc	3.0.1
PIGZ_MAHESHBINNANI	rseqc	3.0.1
SALMON_QUANT	salmon	1.3.0
SAMTOOLS_FLAGSTAT	samtools	1.10.2
SAMTOOLS_FLAGSTAT	samtools	1.10.2
SAMTOOLS_IDXSTATS	samtools	1.10.2
SAMTOOLS_IDXSTATS	samtools	1.10.2
SAMTOOLS_SORT	samtools	1.10.2
SAMTOOLS_SORT_QUALIMAP	samtools	1.10.2
SAMTOOLS_STATS	samtools	1.10.2
SE_GENE_SYMPY	bioconductor-summarizedexperiment	1.20.0
SE_TRANSCRIPT_SYMPY	bioconductor-summarizedexperiment	1.20.0
UNPIGZ_STAR	pigz	2.3.4
	star	2.7.7a
UTAH_GRAHAMHAWKS	gcc	7.5.0
	perl	5.2.0
	samtools	1.10
XPRESS	xpress	1.7.1.linux
STRINGTIE_STRINGTIE	stringtie	2.1.3
SUBREAD_FEATURECOUNTS	subread	2.0.1
TRIMGALORE	trimgalore	0.6.6
TRIMGALORE_TRIMMOMATIC	trimmomatic	0.39
UCSC_BEDGRAPHTOBIGWIG	ucsc	377
UCSC_BEDGRAPHTOBIGWIG	ucsc	377
UNTOOL_GENERAL_MAPS	untool	1.1.0
Workflow	Nextflow	20.09.1
	all used tools	v1.20.1b-47-g1e03

nf-core/rnaseq Methods Description

Automatically generated by nf-core/rnaseq v3.0dev of the nf-core collection of workflows. https://github.com/nf-core/rnaseq

Methods

Data was processed using nf-core/rnaseq v3.0dev ([10.5281/zenodo.1400710](https://doi.org/10.5281/zenodo.1400710)) of the nf-core collection of workflows [Ewels et al., 2020], utilising reproducible software environments from the Bioconda [Grüning et al., 2018] and Biocontainers [da Veiga Leprevost et al., 2017] projects.

The pipeline was executed with Nextflow v20.09.1 [Di Tommaso et al., 2017] with the following command:

```
nextflow run nf-core/rnaseq -r 3.0dev -profile docker --input samplesheet.csv --outdir <OUTDIR> --genome GRCh37
```

References

- Ewels, P., Peltzer, A., Fillinger, S., Patel, H., Alneberg, J., Wilm, A., Garcia, M. U., Di Tommaso, P., & Nahnsen, S. (2020). The nf-core framework for community-curated bioinformatics pipelines. Nature Biotechnology, 38(3), 276–278. [10.1038/s41587-020-0439-x](https://doi.org/10.1038/s41587-020-0439-x)
- Grüning, B., Dale, R., Sjödin, A., Chapman, B. A., Rowe, J., Tomkins-Tinch, C. H., Valieris, R., & Köster, J. & Bioconda Team. (2018). Bioconda: sustainable and comprehensive software distribution for the life sciences. Nature Methods, 15(7), 475–476. [10.1038/s41592-018-0046-7](https://doi.org/10.1038/s41592-018-0046-7)
- da Veiga Leprevost, F., Grüning, B. A., Alves Aflitos, S., Röst, H. L., Uszkoreit, J., Barsnes, H., Vaudel, M., Moreno, P., Gatto, L., Weber, J., Bai, M., Jimenez, R. C., Sachsenberg, T., Pfeuffer, J., Vera Alvarez, R., Griss, J., Nesvizhskii, A. I., & Perez-Riverol, Y. (2017). BioContainers: an open-source and community-driven framework for software standardization. Bioinformatics (Oxford, England), 33(16), 2580–2582. [10.1093/bioinformatics/btx192](https://doi.org/10.1093/bioinformatics/btx192)

Notes:

- The methods above does not include parameters or software versions that may have been used. Ensure this config file is also uploaded with your publication!
- You should also cite all software used within this run. Check the "Software Versions" of this report to get more information.

nf-core/rnaseq Workflow Summary

- this workflow was run with -profile docker and parameters set/used:

Input/output options

```
input                 samplesheet.csv
outdir                <OUTDIR>
```

Reference genome options

```
genome                GRCh37
```

Read trimming options

```
trim_fastq            false
```

Alignment options

```
aligner               star_salmon
```

Process skipping options

```
skip_bigwig           false
skip_stringtie        false
skip_fastqc           false
skip_preseq           false
skip_dupradar         false
skip_qualimap         false
skip_rseqc            false
skip_biotype_qc       false
skip_deseq2_qc        false
skip_multiqc          false
```

Custom options

```
custom_config_version 1.0
custom_config_base    https://raw.githubusercontent.com/nf-core/configs/master
```

Reference genome options

```
fasta                 https://raw.githubusercontent.com/nf-core/test-datasets/rnaseq/reference/genome.fa
gtf                   https://raw.githubusercontent.com/nf-core/test-datasets/rnaseq/reference/genes_with_empty_tid.gtf.gz
gff                   https://raw.githubusercontent.com/nf-core/test-datasets/rnaseq/reference/genes.gff.gz
transcript_fasta      https://raw.githubusercontent.com/nf-core/test-datasets/rnaseq/reference/transcriptome.fasta
additional_fasta      https://raw.githubusercontent.com/nf-core/test-datasets/rnaseq/reference/gfp.fa.gz
```

Read trimming options

```
trim_fastq            false
```

Alignment options

```
aligner               star_salmon
```

Process skipping options

```
skip_bigwig           false
skip_stringtie        false
skip_fastqc           false
skip_preseq           false
skip_dupradar         false
skip_qualimap         false
skip_rseqc            false
skip_biotype_qc       false
skip_deseq2_qc        false
skip_multiqc          false
```

Custom options

```
custom_config_version 1.0
custom_config_base    https://raw.githubusercontent.com/nf-core/configs/master
```

Reference genome options

```
genome                GRCh37
```

Quality Control

kraken_db
https://raw.githubusercontent.com/nf-core/test-datasets/eb0cbf73c3f103f8aeda9878ba200e92b4d045d8/data/genomics/sarscov2/genome/db/kraken2.tar.gz

Process skipping options

skip_bbsplit
false

Institutional config options

config_profile_contact
/Teresa Shippy (tshippy@ksu.edu)

config_profile_description
Beocat at Kansas State University cluster profile provided by nf-core/configs.

config_profile_name
Test profile

Generic options

trace_report_suffix
2026-08-25_20-40-32

Core Nextflow options

configFiles
/homes/tshippy/.nextflow/assets/.repos/nf-core/rnaseq/clones/e7ca46272c8f9d5ceee3f71759f4ba551d3217a4/nextflow.config,
/bulk/bioinfo/Nextflow_demos/beocat_rnaseq.config

container
[.*PARABRICKS_STARGENOMEGENERATE:community.wave.seqera.io/library/htslib_samtools_star_gawk:4de2f983041d42e6]

containerEngine
singularity

launchDir
/bulk/bioinfo/Nextflow_demos

profile
test,singularity

projectDir
/homes/tshippy/.nextflow/assets/.repos/nf-core/rnaseq/clones/e7ca46272c8f9d5ceee3f71759f4ba551d3217a4

revision
master

runName
big_maxwell

userName
tshippy

workDir
/bulk/bioinfo/Nextflow_demos/work