RUM 2

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The RUM Pipeline

Enhancements in RUM 2

Installation Command-line interface Job status

Demo

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Issues



About me

- Software engineering background
- Working at Penn since January
- Experience in a variety of languages (Perl, Java, Clojure (lisp), Python, Ruby)

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About RUM

- "RUM is an alignment, junction calling, and feature quantification pipeline specifically designed for Illumina RNA-Seq data"
- Written by Gregory Grant (ggrant@grant.org)
- Runs on Linux / UNIX / Mac
- Can distribute work across multiple machines
- Written in Perl 5

The RUM Pipeline

- Align all reads against genome using Bowtie
- Align all reads against transcriptome using Bowtie
- Merge genome and transcriptome alignments and identify unmapped reads
- Align unmapped reads against genome using BLAT
- Merge Bowtie and Blat alignments
- Produce some output files based on merged results

RUM output files

- Files of unique and non-unique alignments
- All alignments in SAM format
- Coverage plots
- Feature quantifications
- Junction calls
- List of novel inferred internal exons



Work distribution



- RUM can run alignment phase in parallel
- Splits input reads into N chunks
- Alignment for each chunk is independent of other chunks
- Results are merged together for coverage, quantification, junction calling

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RUM 2 Features

- Standard installation process
- New command-line interface
- Get status of running job
- Restart a job where it left off
- More reliable kill command
- Run a chunk or postprocessing by itself
- Relocatable indexes
- SAM file is closer to conforming to standard

Installation

- Uses standard Perl Makefile.PL
- Should be familiar to system administrators
- Download tarball from https://github.com/PGFI/rum/downloads
- Run perl Makefile.PL
- Install indexes using bin/rum_indexes
- That's it!



Command-line interface

Usage is rum_runner ACTION [OPTIONS] where action is one of:

- align Run an alignment
- status Check the status of a job
- stop Stop a job (can be restarted later)
- kill Stop a job and clean it up (to restart from scratch)
- clean Remove output files for a job
- help Get help
- version Show version number

rum_runner align

Use rum_runner align to run an alignment:

```
rum_runner align \
  --output dir \
  --index ~/rum_indexes/hg19 \
  --name TestJob \
  --chunks 25 \
  ~/samples/forward.fq ~/samples/reverse.fq
```

Job status

 Use rum_runner status to check on the status of a running job.

Processing in 25 chunks

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```
X XXX XX XXXX XXXXX Run bowtie on transcriptome
X XXX XX XXXX XXXXX Parse transcriptome Bowtie output
     XX XX XXXX
               XXXXX Merge unique mappers together
X XXX
X XXX XX XXXX XXXXX Merge non-unique mappers together
X XXX XX XXXX XXXXX Make unmapped reads file for blat
X XXX
    XX XX XXXX XXXXX Run blat on unmapped reads
X XXX
    XX XX XXXX XXXXX Run mdust on unmapped reads
     XX XX XXXX XXXXX Parse blat output
X XXX
X XXX
     XX XX XXXX
                XXXXX Merge bowtie and blat results
X XX
      XX XX
           XXX
                 XXXX
                      Clean up RUM files
X XX
      XX XX
           XXX
                 XXXX Produce RUM Unique
X XX
     XX XX
           XXX
                 XXXX Sort RUM_Unique by location
X X
      XX XX
           XXX
                 XXXX
                      Sort cleaned non-unique mappers by ID
X X
           XXX
     XX XX
                 XXXX
                      Remove duplicates from NU
X X
     XX XX
           XXX
                 XXXX Create SAM file
X X
     XX XX
           XXX
                 XXXX Create non-unique stats
X X
           XXX
      XX XX
                 XXXX Sort RUM_NU
ΧХ
      XX XX
           XXX
                 XXXX
                      Generate quants
```

Job status

Postprocessing

Merge RUM_NU files
Make non-unique coverage
Merge RUM_Unique files
Compute mapping statistics
Make unique coverage
Finish mapping stats
Merge SAM headers
Concatenate SAM files
Merge quants
make_junctions
Sort junctions (all, bed) by location
Sort junctions (all, rum) by location
Sort junctions (high-quality, bed) by location
Get inferred internal exons
Quantify novel exons

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All the chunk error log files are empty. That's good. Main error log file is empty. That's good.

RUM is running (job ids 815718, 815720).

Recovering from errors



- Sometimes bad things happen
- RUM 2 allows easier recovery from infrastructure failures
- Running "rum_runner align" again will restart a job from where it left off
- Can save a lot of time when recovering from infrastructure failure
- Determines state of job by looking at which output files exist

Killing a job

- To stop a job and remove all of its output:
 - rum_runner kill -o dir
- Useful if you've run a job with incorrect parameters and need to start over

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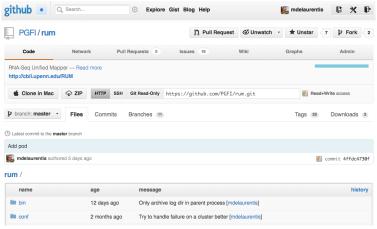
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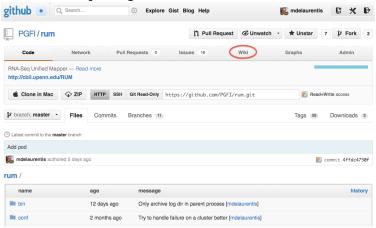
Main Github page

https://github.com/PGFI/rum



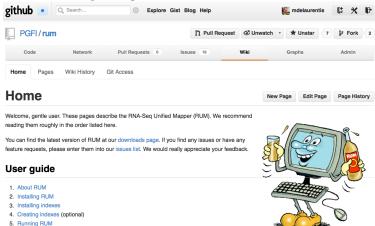
User guide

https://github.com/PGFI/rum/wiki



User guide

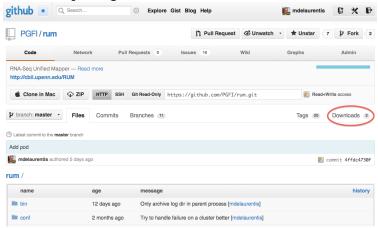
https://github.com/PGFI/rum/wiki



RUM output files
 Running RI IM on a cluster (optional)

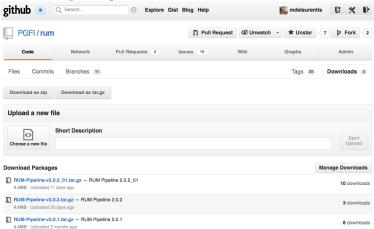
Downloads

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Downloads

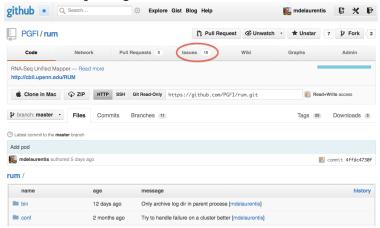
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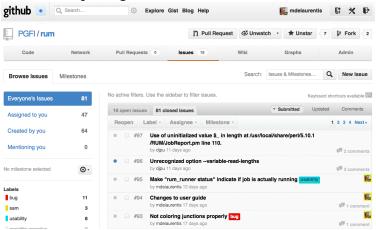
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Issues

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Possible future enhancements

- Use original read names throughout the pipeline
- Performance improvements