Using BioGrids for RNA-Seq on AWS and Your Laptop

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Today we will

Install software with BioGrids

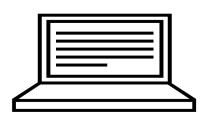
Run an RNA-Seq workflow

Replicate above on AWS and laptop



BioGrids on AWS

faithful laptop



AWS EC2 Instance



RNA-Seq Workflow

open a terminal

open a browser: biogrids.org/wiki/workshops



Subtle Things

- Capsule Environment
- .bashrc / .profile not changed
- binary installs



Avoid Time Sinks

https://www.biostars.org/p/189261/:

This seems to be a bug when installing fastqc using apt-get install fastqc

STARmanual.pdf

....which creates problems for STAR compilation. One option to avoid this problem is to install gcc

http://github.gersteinlab.org/exceRpt/

Manual Installation:

....generally not recommended ... <snip> ... instructions on how to install exceRpt and its various dependencies will [one day] be listed toward the bottom of this page.



Reproducible Research

Self Documenting

\$ STAR --sbapp:d

```
Capsule:STAR using star version 2.5.3a
Version information for: /programs/i386-mac/star
```

```
Default version: 2.5.3a In-use version: 2.5.3a Other available versions: none Overrides use this shell variable: STAR M
```

Include in workflow:

```
STAR --sbapp:d samtools --sbapp:d
```



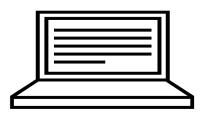
Config File

```
[installer]
site = biogrid-production
key = 70rYFBTDnmCr93VUklfbf1s3M4jdyC9bFVYHew==
user = jvincent1
[packages]
star@2.5.3a = i386-mac
samtools@1.5 = i386-mac
iqv@2.4.10 = i386-mac
```



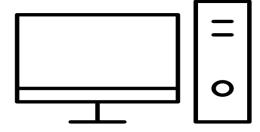
This Is Handy

faithful laptop



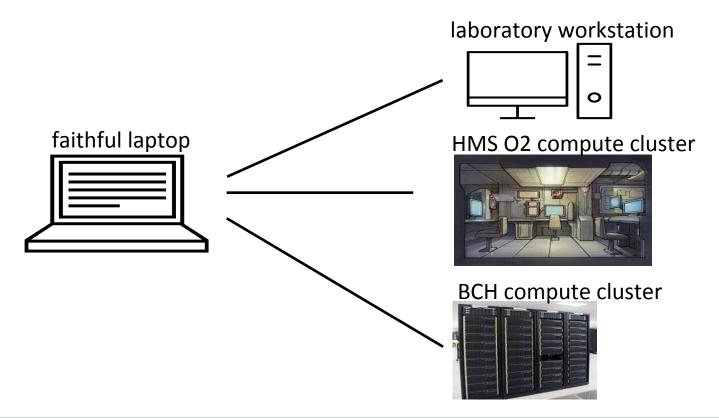
biogrids save mysetup.txt

new workstation



biogrids reactivate mysetup.txt

BioGrids is Portable



BioGrids Benefits

save time - reduce headaches scale and share workflows part of reproducible research

BioGrids Consortium















Personnel













Compute Infrastructure



Funding HMS Tools and Technologies Committee

Why BioGrids?

You — Cover of Nature

compile software compile libraries manage dependencies manage versions manage paths change versions

learn to use software optimize workflow get science done



RNA-Seq Overview

Harvard Chan Bioinformatics Core (HBC)

http://bioinformatics.sph.harvard.edu/training



RNA-Seq Overview

Biological samples / Library prep

sequence reads

quality check

adapter/quality trimming

splice aware mapping to genome

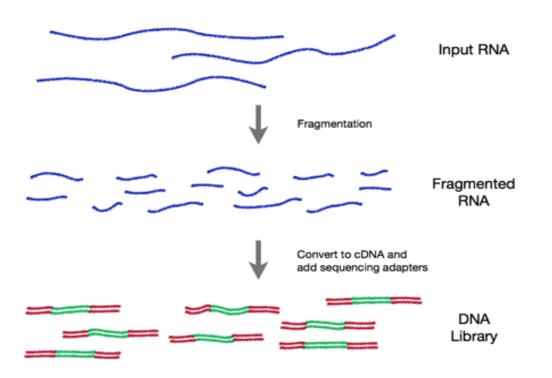
count reads associated with genes

statistical analysis identify differentially expressed genes

hbctraining.github.io/Intro-to-rnaseq-hpc-O2

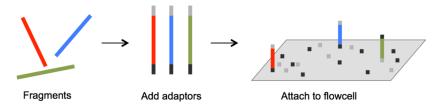


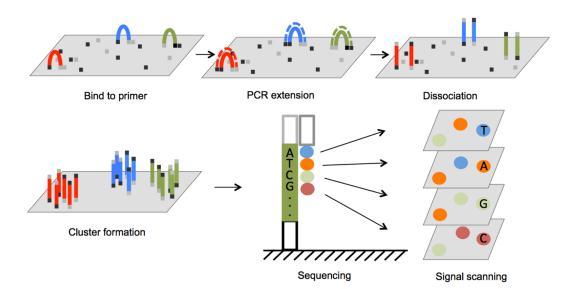
RNA Prep





Sequencing





RNA-Seq Overview

Biological samples / Library prep



adapter/quality trimming (trimmomatic)

splice aware mapping to genome STAR

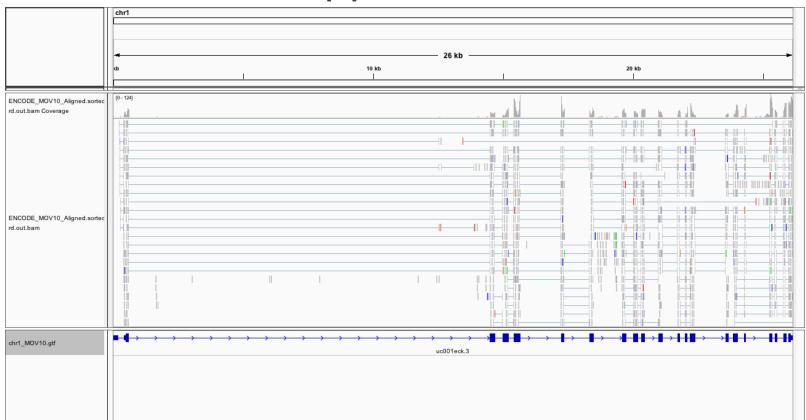
count reads associated with genes ——— subRead

statistical analysis identify differentially expressed genes

hbctraining.github.io/Intro-to-rnaseq-hpc-O2



Mapped Reads



Check Results

IGV

- 1: Genomes / Load Genome from File... (chr1_MOV10.fa)
- 2: File / Load from file... (.gtf file)
- 3: File / Load from file... (.bam file)



DevOps with BioGrids

workflow

software stack

compute resources

bioinformatics

BioGrids

laptop HMS O2 AWS



AWS Hands On

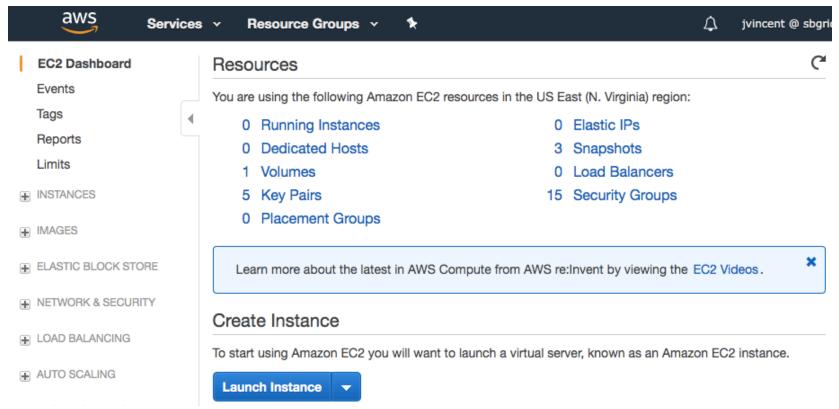
https://sbgrid.signin.aws.amazon.com/console

username: workshop21

password: Biogrids_Workshop1

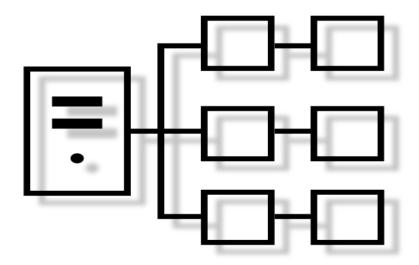


AWS - Amazon Web Services



AWS Parallel Cluster

aws-parallelcluster.readthedocs.io



scalable HPC cluster



help@biogrids.org

BioGrids is funded by the Harvard Medical School Tools and Technologies Committee



Additional Resources

ENCODE data files can be found here for CalTech RNA-Seq: http://hgdownload.cse.ucsc.edu/goldenPath/hg19/encodeDCC/wgEncodeCaltechRnaSeq/

Use this bam file: wgEncodeCaltechRnaSeqK562R1x75dAlignsRep1V2

Region of MOV10 gene: chr1:113,214,934-113,243,900

How to download whole genome:

- UCSC ftp site: hgdownload.cse.ucsc.edu
- UCSC web site: http://hgdownload.cse.ucsc.edu/goldenPath/hg19/chromosomes/
- UCSC recommends using an ftp client for large file downloads
- chr1 is only 70M



References

TRAINING

hbctraining.github.io/Intro-to-rnaseq-hpc-O2

<u>AWS</u>

https://aws.amazon.com/ec2/getting-started

ENCODE

https://www.encodeproject.org

IMAGES

https://www.diagenode.com/en/categories/Library-preparation-for-RNA-seq https://rnaseq.uoregon.edu

