SSC Intro to NGS Bioinformatics Course
May 2013

We will meet in Room 101B of the Flawn Academic Center (FAC) building. We STRONGLY encourage you to use the computers provided in the classroom, but you may also bring your personal laptops.

- May 2013
  - Your Instructors
    - Day 1: Linux/TACC Introduction and Read Mapping
      - Part 1: Linux/TACC Introduction
      - Part 2: Read Mapping
        - Enrichment modules (4:30-5:30)
        - Extras
    - Day 2: Handling Raw and Aligned sequences, and Calling Genome Variants
      - Part 1. Handling Raw and Aligned sequences
      - Part 2. Calling Genome Variants
        - Enrichment module (12:30-1:30)
        - Enrichment modules (4:30-5:30)
        - Extras
    - Day 3: RNA-seq
      - Part 1. Introduction to RNA-seq Counting
      - Part 2. The Tuxedo RNA-seq Pipeline (Tophat & Cufflinks)
        - Enrichment module (12:30-1:30)
        - Enrichment modules (4:30-5:30)
        - Extras
    - Day 4: Assembly and Annotation
      - Part 1. Genome Assembly
      - Part 2. Assembly Annotation
        - Enrichment module (12:30-1:30)
        - Enrichment module (4:30-5:30)
  - Resources

Resources tool list, file formats & more

Link to Etherpad: https://etherpad.mozilla.org/g2NxlEAFWL

Use this to post any questions you have about the lessons and tutorials.

Your Instructors

<table>
<thead>
<tr>
<th>Name</th>
<th>Initials</th>
<th>Affiliation</th>
<th>Expertise</th>
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</thead>
<tbody>
<tr>
<td>Scott Hunicke-Smith</td>
<td>SPHS</td>
<td>Director GSAF</td>
<td>Everything, if loosely defined (but especially awk)</td>
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<tr>
<td>Jeff Barrick</td>
<td>JB</td>
<td>Asst. Prof. Biochemistry</td>
<td>Microbes, Perl, C++, Mac, miscellanea</td>
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<tr>
<td>Dhivya Arasappan</td>
<td>DA</td>
<td>GSAF</td>
<td>RNA-seq, transcriptome assembly</td>
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<tr>
<td>Anna Battenhouse</td>
<td>AB</td>
<td>Iyer Lab</td>
<td>Eukaryotes, Bash scripting, UCSC Genome Browser</td>
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<tr>
<td>Daechan Park</td>
<td>DP</td>
<td>Iyer Lab</td>
<td>Eukaryotes, ChiP-seq, Python, Samtools</td>
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<tr>
<td>Nichole Bennett</td>
<td>NB</td>
<td>Parmesan/Singer Labs</td>
<td>Python, R, Unix</td>
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<tr>
<td>Dan Deatherage</td>
<td>DD</td>
<td>Barrick Lab</td>
<td>Unix, Python, NGS Library Prep</td>
</tr>
<tr>
<td>Nathan Abell</td>
<td>NA</td>
<td>Iyer Lab</td>
<td>Eukaryotes, RNA-Seq</td>
</tr>
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instructor action item list

- Info for the instructors
  - Day 1a: Scott 1b: Jeff
  - Day 2a: Jeff, Daechan, Anna, 2b: Scott
  - Day 3a: Jeff 3b: Iyer lab
  - Day 4a: Jeff, 4b: Scott

Instructors: meet 9am Monday for final check

Each Part 1/Part 2 section needs to be standardized with:
- Learning Objectives
Day 1: Linux/TACC Introduction and Read Mapping

Part 1: Linux/TACC Introduction

- General introduction (SPHS)
  - Introduction to Bioinformatics Prezi
- Linux refresher (SPHS)
  - Step 1: Start somewhere
  - Step 2: Establishing a profile on Lonestar
  - Step 3: Editing files
  - Step 4: Final explanations
- Using TACC's Lonestar Cluster (SPHS)
  - Diagram of Lonestar's directories
  - Diagram of running a job on Lonestar
  - Using SFTP for file browsing on Linux.
- Recap and "for further study"

Part 2: Read Mapping

- Introduction to next-gen sequencing technologies (JB)
  - Paper comparing NGS technologies (Liu et al., 2012)
  - Official Illumina video | Another Illumina video
  - Broad Center GA Boot Camp
- Variant calling workflow diagram:

Enrichment modules (4:30-5:30)

- Sharing Linux tricks - linux one-liners (SPHS)
- Working on TACC from your Mac or PC (AB)
  - Editing files, more detail

Extras

- Tutorial - Start diploid mapping for Day 2
- Running Unix & Perl for Biologists tutorial at TACC
- Installing Virtual machine & Linux on Windows (DP)

Day 2: Handling Raw and Aligned sequences, and Calling Genome Variants

Part 1. Handling Raw and Aligned sequences

- Overview and Resources
- Evaluating your raw sequencing data (AB)
  - GSAF adaptor and barcode sequence resource
Part 2. Calling Genome Variants

- Variant calling workflow diagram:
  - Variant calling tutorial (SAMtools)
  - Integrative Genomics Viewer (IGV) tutorial

Enrichment module (12:30-1:30)

- Shell Scripting (SPHS/AB)

Enrichment modules (4:30-5:30)

- Installing Linux tools (JB)

Extras

- Calling variants in diploid genomes (SPHS)
- Introduction to genome variation
- Variant calling with GATK (SPHS)
- Annotating variants (SPHS)
- Visualize mapped data at UCSC genome browser (AB)
- Genome variation in mixed samples (FreeBayes, deepSNV) (JB)
- Identifying structural variants (SVDetect) (JB)
- SRA toolkit (AB)

Day 3: RNA-seq

Part 1. Introduction to RNA-seq Counting

- Differential gene expression analysis (JB)

Part 2. The Tuxedo RNA-seq Pipeline (Tophat & Cufflinks)

- Differential expression with splice variant analysis

Enrichment module (12:30-1:30)

- Identifying mutations in microbial genomes (breseq) (JB)

Enrichment modules (4:30-5:30)

- Start tophat by submitting to lonestar (DA)
Extras

- Visualize mapped data at UCSC genome browser (AB)
- non-coding RNA analysis

Day 4: Assembly and Annotation

Part 1. Genome Assembly

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- Genome Assembly Examples (SPHS)
- Tutorial: Genome Assembly (velvet) (SPHS)

Part 2. Assembly Annotation

- Genome Annotation (Glimmer3) (SPHS)
- Evaluating & Visualizing assemblies (bacterial, SPHS)

Enrichment module (12:30-1:30)

- Office hours: “I want to learn how to install and use this tool called ______ that we didn’t talk about in class.” (JB).

Enrichment module (4:30-5:30)

- Transcriptome assembly & annotation
- Protein functional classification...
- Custom Genome Databases

Resources

- Scott’s list of linux one-liners
- Example BWA alignment script
- Exercises
- Key take home points
- Resources tool list, file formats & more

As you’re getting settled