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The bioBakery microbial community analysis environment

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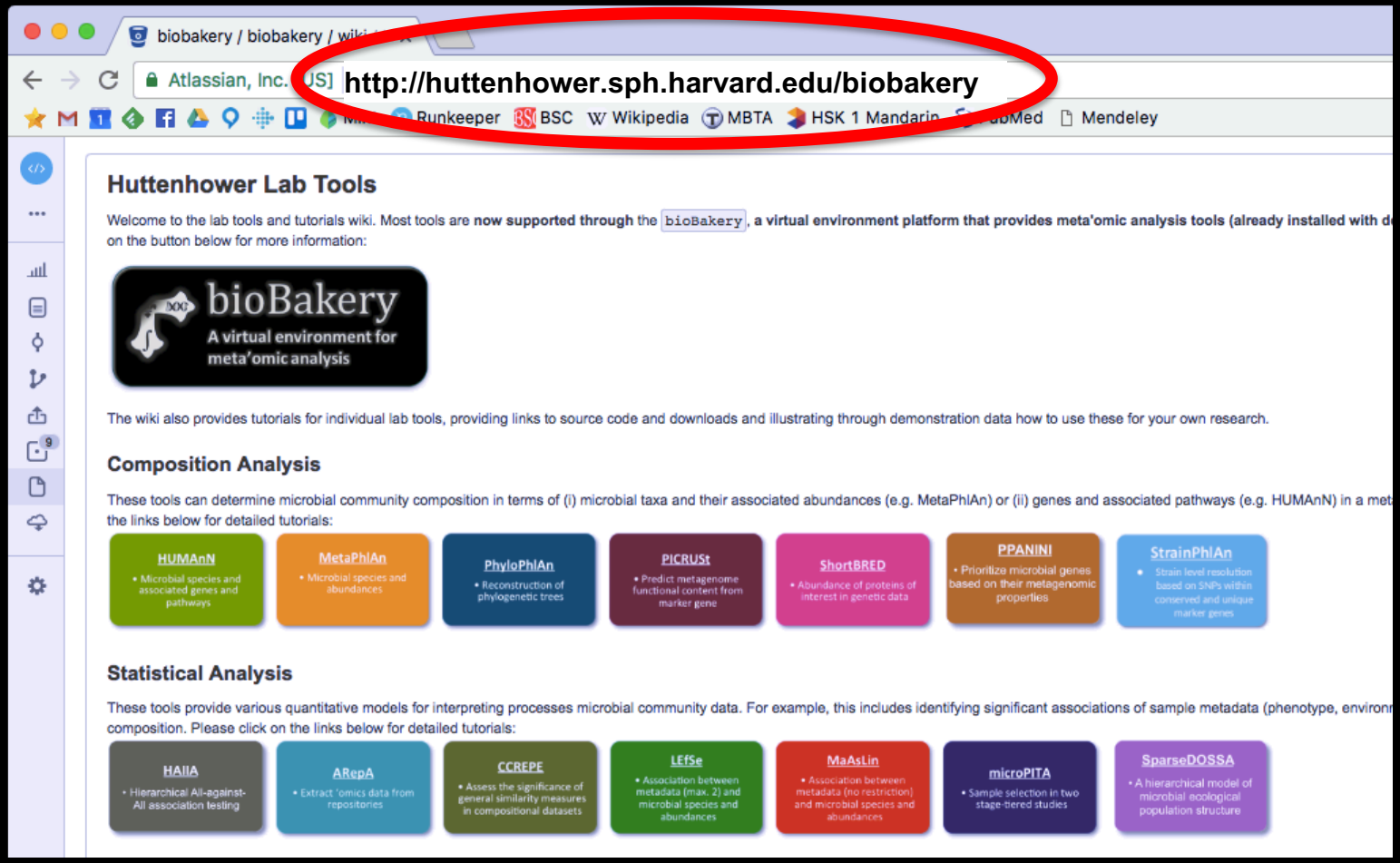
The bioBakery: a software environment for integrative microbiome analyses

- Environment for meta'ome analysis
  - Shotgun metagenomes/transcriptomes
  - Taxonomic and functional profiling
  - Experimental design, statistical analysis

- Pre-built one-click environment to run:
  - On your laptop graphically
  - On a server remotely
  - On the cloud (Amazon / Google)

Launching the bioBakery on Google Cloud

http://huttenhower.sph.harvard.edu/biobakery



Launching the bioBakery on Google Cloud

http://huttenhower.sph.harvard.edu/biobakery

Huttenhower Lab Tools

Welcome to the official Huttenhower Tutorials wiki.

We now support `bioBakery`, a virtual environment platform that provides Huttenhower tools (already installed!). Please click on the button below for more information:

bioBakery
A virtual environment for meta'omic analysis

...es tutorials for Huttenhower tools, illustrating through... ...nder three main categories as shown below. Click on the to...

bioBakery

Quick Start

1. Install Virtualbox
2. Install Vagrant
3. Download and unpack bioBakery
4. Run bioBakery by double clicking (or executing from the command line) the file for your operating system

- Linux and MacOS: `start_biobakery.command`
- Windows: `start_biobakery.bat`

Please note the first time you run bioBakery it will download and extract the box (~6 GB in size). This can take a bit of time depending on your internet connection. When you run bioBakery in the future, it will start up much faster (~30 seconds).

You can also run bioBakery in Google Cloud or you can install individual bioBakery tools.

Please see the sections and links that follow for detailed information about bioBakery.

Workshop Usage

Overview

bioBakery is an easy to use, virtual environment that provides a platform fo... ...unity to use the Huttenhower tools without having to install the tool suite directly on their personal machines. This documentation provides in depth instructions on all topics relate... ...basic setup and advanced configuration).

We provide support for bioBakery users. Please join the bioBa... ...and feel free to post questions directly to the group or by emailing biobakery-users@googlegroups.com.

Basic Usage	Workshop Usage	Advanced configuration	Internal documentation
• Launch bioBakery on your personal computer • Use bioBakery online with Google Cloud	• Connect to Google Cloud instances • Run tutorials	• Interacting with the VM from the command line • Sharing data with the VM • Building a new Google	• Adding new tools • Packaging new image • Releasing new image

Welcome to the bioBakery Workshop!

Web Browser

Your instructor will provide you with the information...

1. The connection method (Web Browser, V... ...or SSH)
2. The name or IP address of your instance
3. The password (and user name if required)

Please follow the instructions under the connection method above for your workshop to get started in bioBakery Google Cloud.

Launching the bioBakery on Google Cloud

http://huttenhower.sph.harvard.edu/biobakery

Option 1: Web Browser Connection

The web browser based connection will provide you with desktop access to the bioBakery Google Cloud instance. It will also provide you
method to download and upload files to your bioBakery instance.

How to Connect

Follow the instructions below to connect to your instance through your web browser.

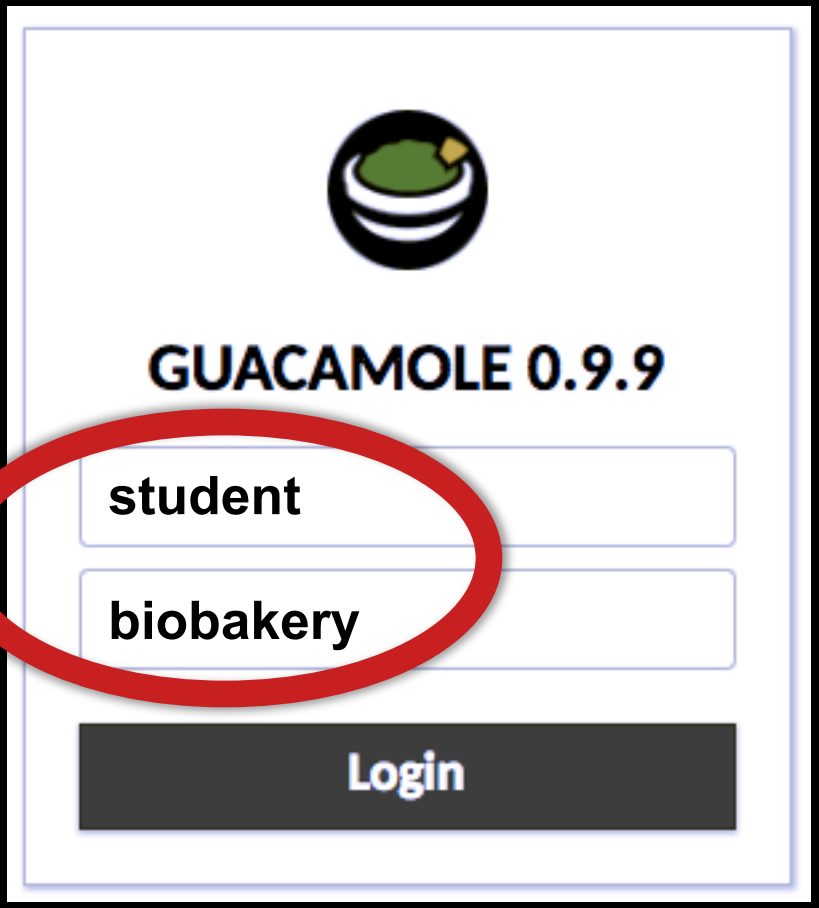
Step 1: Log in to the bioBakery Guacamole Server

1. Go to the http://huttenhower.sph.harvard.edu/guacamole
2. Login with the Guacamole username and password `provided by your instructor`

The Guacamole Server log in screen is shown below.

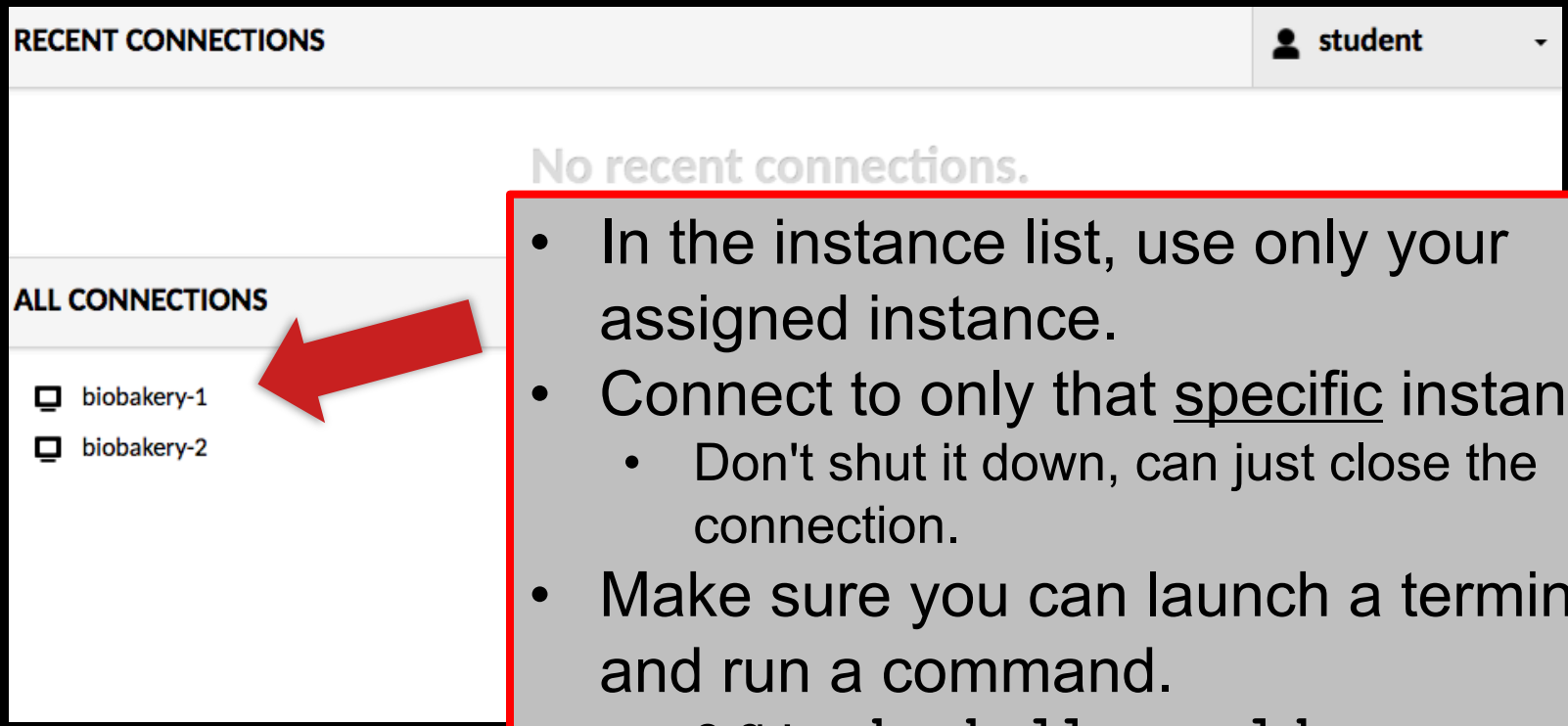
Launching the bioBakery on Google Cloud

http://huttenhower.sph.harvard.edu/guacamole



Launching the bioBakery on Google Cloud

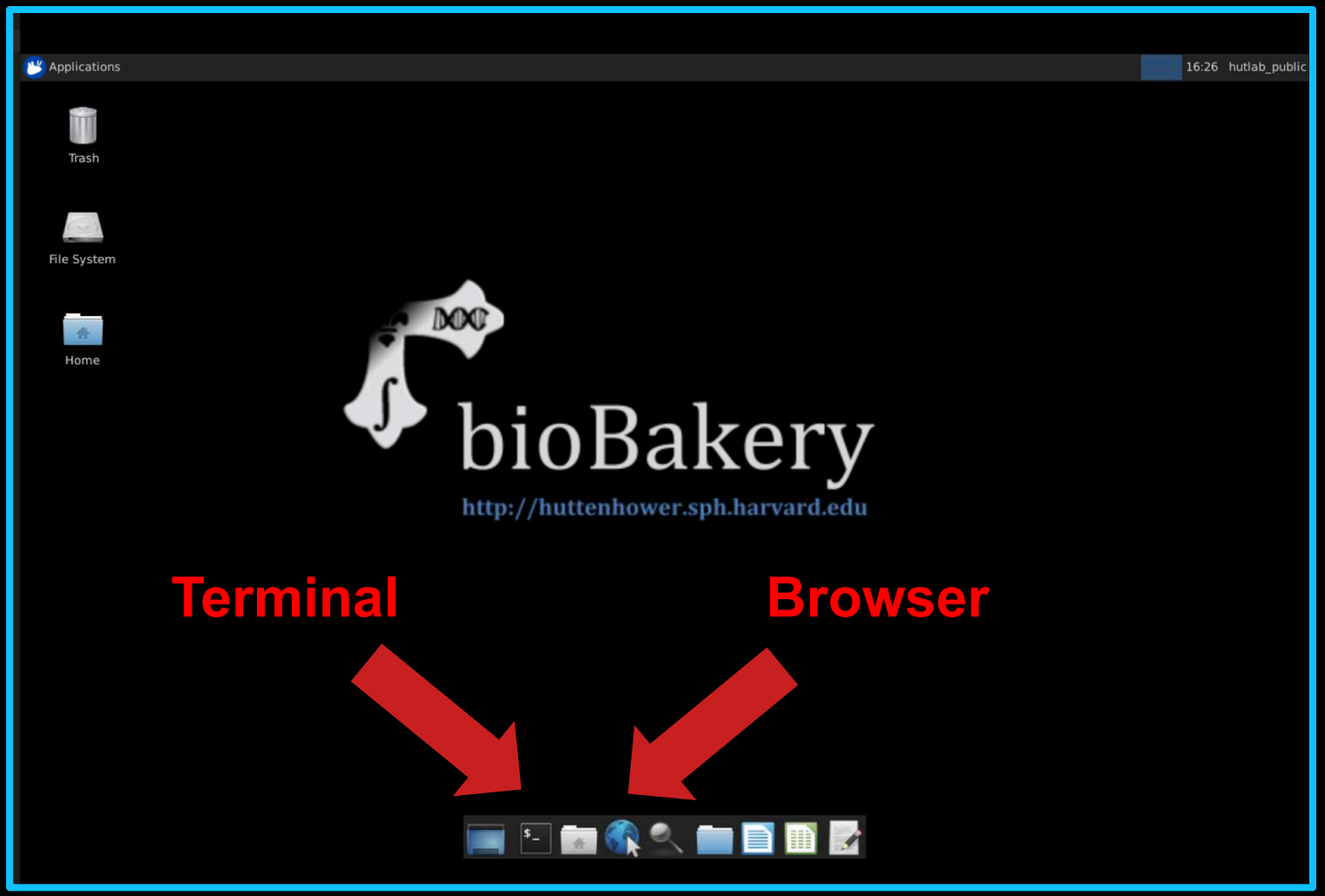
http://huttenhower.sph.harvard.edu/guacamole



- In the instance list, use only your assigned instance.
- Connect to only that specific instance!
  - Don't shut it down, can just close the connection.
- Make sure you can launch a terminal and run a command.
  - e.g.: `echo hello world`
- Try to upload/download a file.

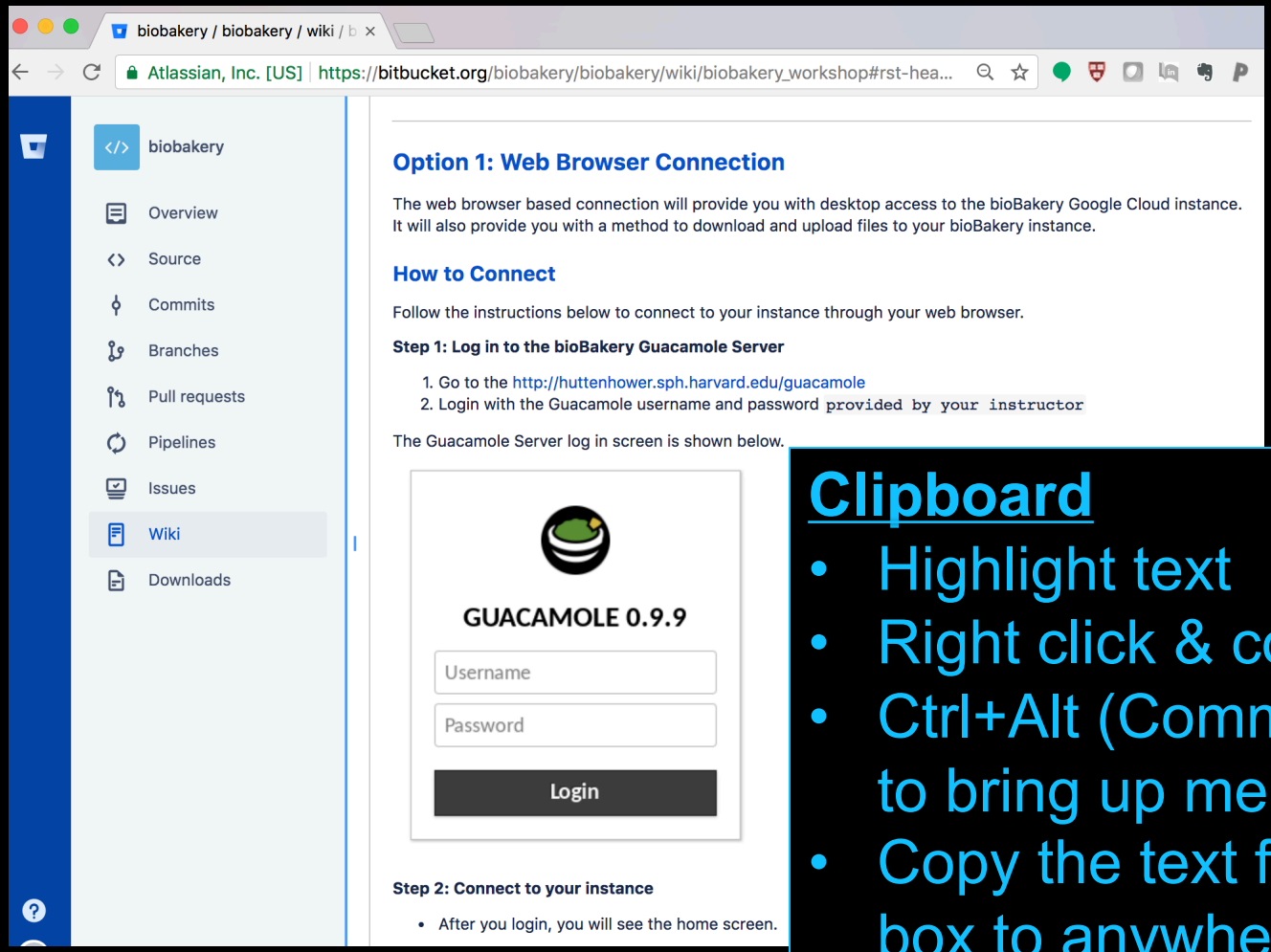
Google Cloud and Guacamole

http://huttenhower.sph.harvard.edu/guacamole



Guacamole: copy and paste

http://huttenhower.sph.harvard.edu/guacamole

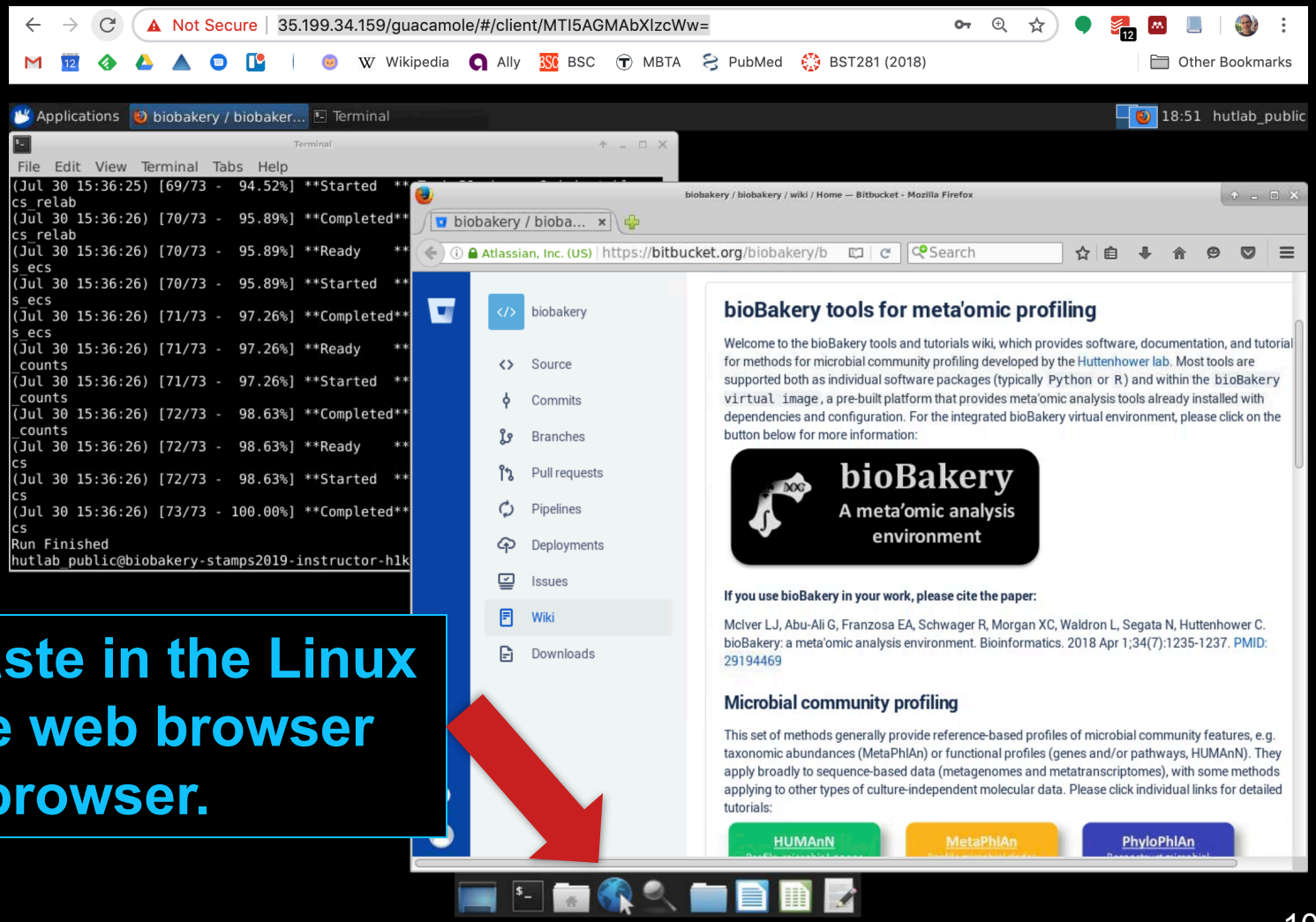


Clipboard

- Highlight text
- Right click & copy
- Ctrl+Alt (Command)+Shift to bring up menu
- Copy the text from the box to anywhere else

Guacamole: copy and paste

http://huttenhower.sph.harvard.edu/guacamole

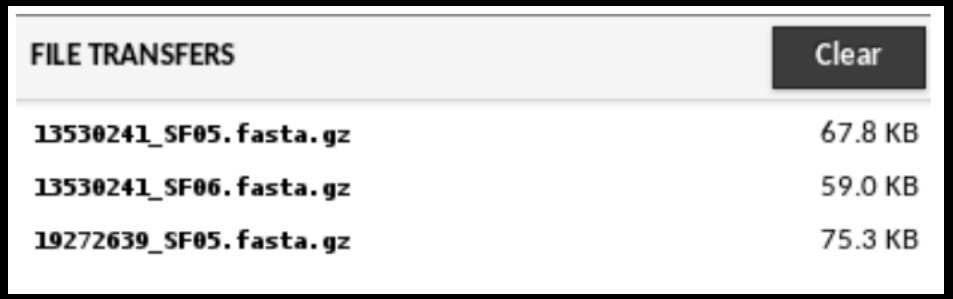


OR copy-paste in the Linux VM from the web browser *in* the web browser.

Guacamole: uploading files

http://huttenhower.sph.harvard.edu/guacamole

- **Drag & drop** the file into the **desktop of your instance**
- The uploaded file will be located in your $HOME directory, which is `/home/hutlab_public/`
- A file transfer status box will be displayed in the lower right-hand corner of the screen



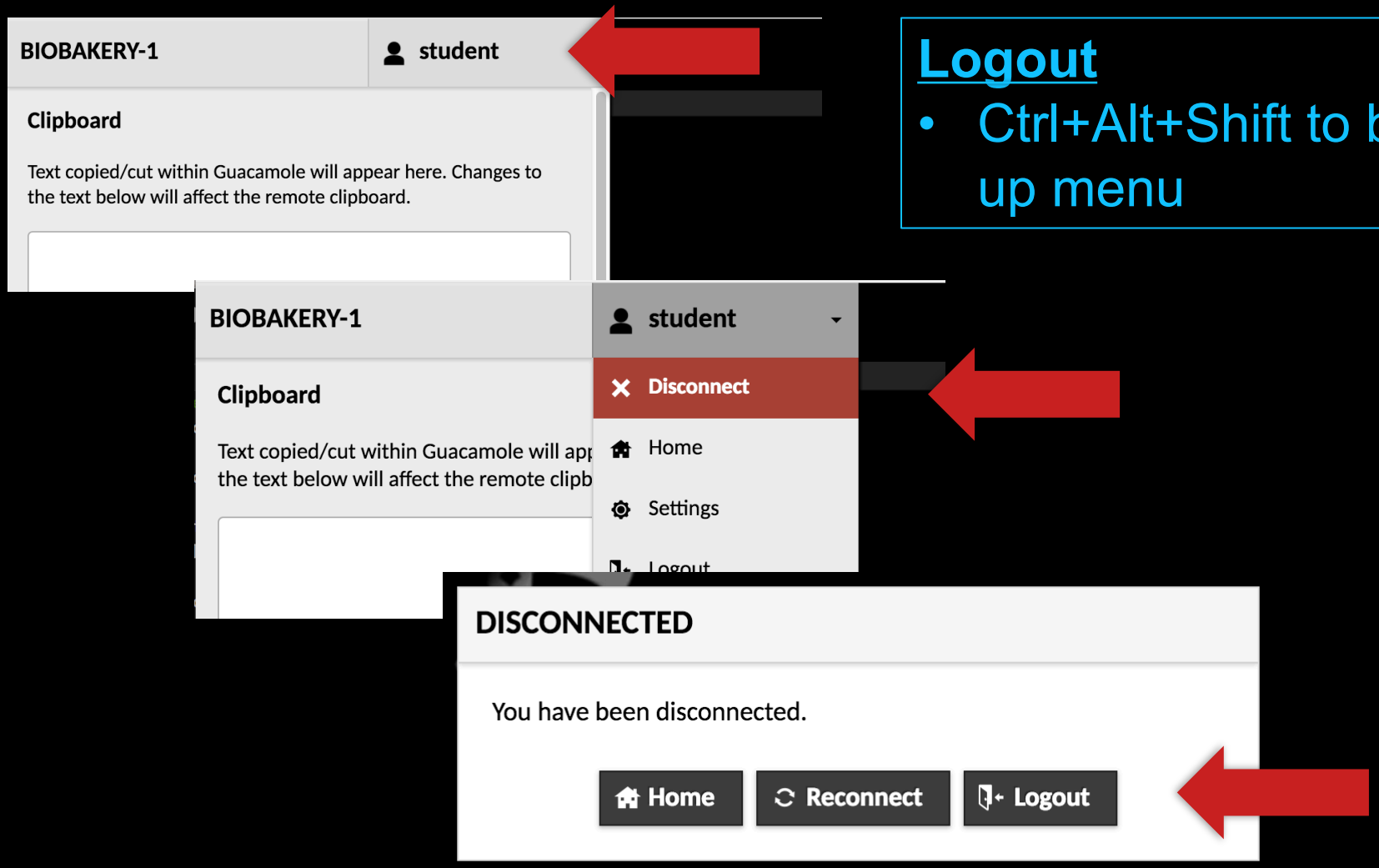
Guacamole: downloading files

http://huttenhower.sph.harvard.edu/guacamole

- Type **Ctrl+Alt+Shift** to bring up the menu
- Click on the **box under Devices**
- Double click on the folders to **get to the folder containing the file(s)** on your instance you would like to download
  - E.g. **`/home/hutlab_public/`**
- **Double click on the file(s)** you would like to download
- A file transfer status box will appear
- Click on the **links in the file transfer status box** to download the files to the Downloads folder on your computer

Guacamole: logging off

http://huttenhower.sph.harvard.edu/guacamole



Logout

- Ctrl+Alt+Shift to bring up menu

The bioBakery:
Methods for integrative meta'omic analysis

1. Upstream microbial gene and genome curation.
2. Microbial community bioinformatics (e.g. taxonomic and functional profiling of meta'omes).
3. Microbial community statistics and association discovery.
4. Reproducible workflows & infrastructure.

Reproducible Workflows & Infrastructure

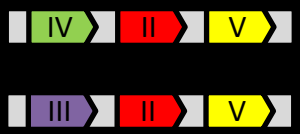
Microbial Genomics | Meta'omic Profiling | Association Discovery

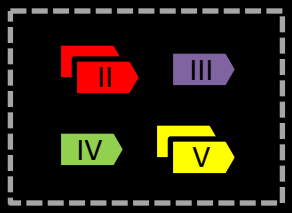
The bioBakery:
1. Microbial ‘omics and reference sequences

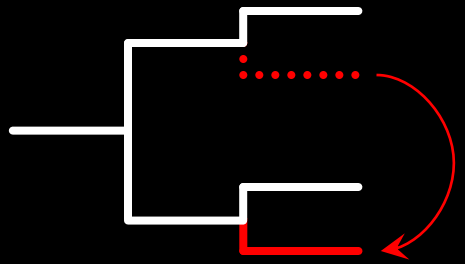
RepoPhlAn
collects & organizes microbial genomes

ChocoPhlAn
constructs & curates microbial pangenomes

PhyloPhlAn
assigns or corrects microbial taxonomy





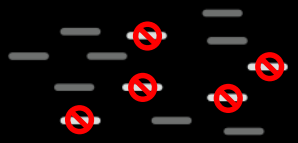


Reproducible Workflows & Infrastructure

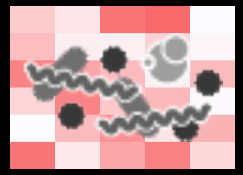
Microbial Genomics | Meta’omic Profiling | Association Discovery

The bioBakery:
2. Microbial community bioinformatics

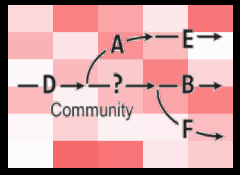
KneadData
quality controls meta'omic seq.



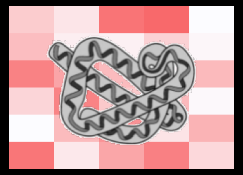
MetaPhlAn2
profiles community membership



HUMAnN2
profiles species-specific functions



ShortBRED
detects & quantifies proteins of interest

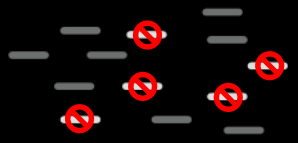


Reproducible Workflows & Infrastructure

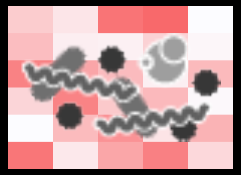
Microbial Genomics | **Meta'omic Profiling** | Association Discovery

The bioBakery:
2. Microbial community bioinformatics

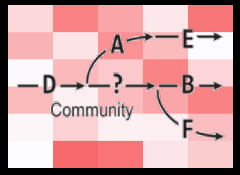
KneadData
quality controls meta'omic seq.



MetaPhlAn2
profiles community membership



HUMAnN2
profiles species-specific functions



ShortBRED
detects & quantifies proteins of interest

QIIME
integration
amplicon-based taxonomic profiling

PICRUSt
predictive functional profiling

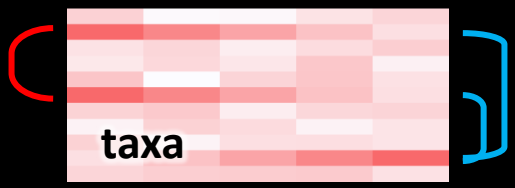
Knight Lab

Reproducible Workflows & Infrastructure

Microbial Genomics | **Meta'omic Profiling** | Association Discovery

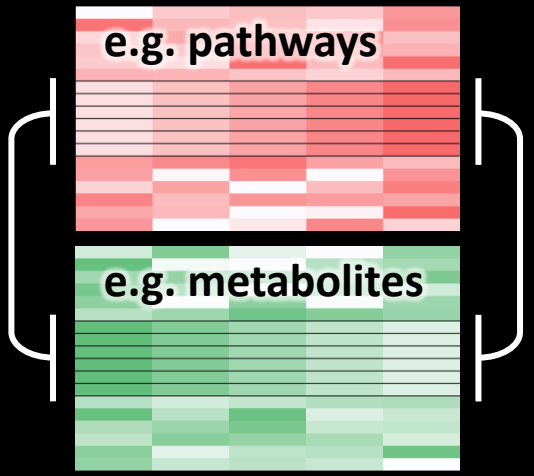
The bioBakery:
3. Microbial community biostatistics

BANoCC
identifies ecological associations among taxa



MaAsLin2
links microbial features with sample metadata

HAllA
identifies associated "blocks" in paired multi'omic datasets



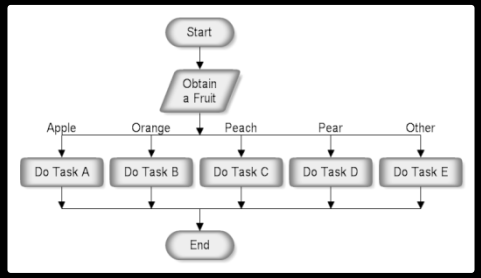
Reproducible Workflows & Infrastructure

Microbial Genomics | Meta'omic Profiling | Association Discovery

The bioBakery:
4. Infrastructure and reproducible science

BreadCrumbs
data visualization & utility library

AnADAMA
scientific workflow management



bioBakery VM & workflows
deployment system

Reproducible Workflows & Infrastructure

Microbial Genomics | Meta'omic Profiling | Association Discovery

AnADAMA2: Another Automated Data Analysis Management Application

http://huttenhower.sph.harvard.edu/anadama2

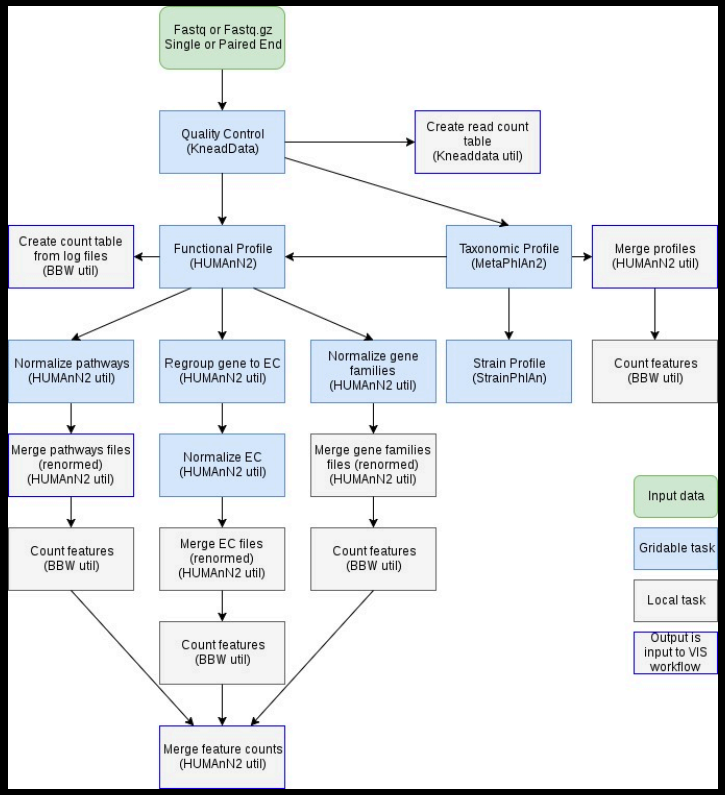
- **Creating efficient, reproducible workflows**
  - A set of modular tasks to transform inputs into outputs
  - Data, tables, visualizations, statistics…
- Reproducible
  - All tasks are logged
    - Includes commands and software versions
  - Automated documentation generation
- Efficient
  - Only those tasks that need to be rerun will run
  - Make-like operation using targets and dependencies
  - Local and/or grid parallelization
  - Jobs are dispatched/monitored/logged
  - Resubmit if job exceeds time/memory

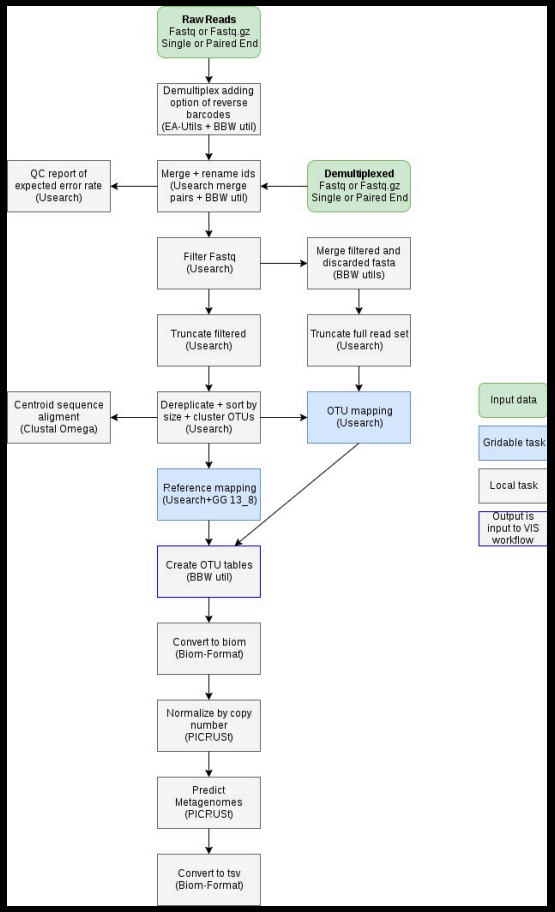

Randall Schwager


Lauren McIver

AnADAMA2: Workflows

- Workflows are AnADAMA2's container for a series of steps that consume a series of inputs and produce outputs.





An example workflow

```
from anadama2 import Workflow

workflow = Workflow(remove_options=["input","output"])
workflow.do("ls /usr/bin/ | sort > [t:global_exe.txt]")
workflow.do("ls $HOME/.local/bin/ | sort > [t:local_exe.txt]")
workflow.do("join [d:global_exe.txt] [d:local_exe.txt] > [t:match_exe.txt]")
workflow.go()
```

A fancier example workflow

```
1 ### Section #1: Import anadama2 and create a workflow instance (Required)
2 from anadama2 import Workflow
3 workflow = Workflow(version="0.0.1", description="A workflow to run MetaPhlAn2" )
4
5 ### Section #2: Add custom arguments and parse arguments (Optional)
6 workflow.add_argument("input-extension", desc="the extensions of the input files", default="fastq")
7 args = workflow.parse_args()
8
9 ### Section #3: Get input/output file names (Optional)
10 in_files = workflow.get_input_files(extension=args.input_extension)
11 out_files = workflow.name_output_files(name=in_files, tag="metaphlan2_taxonomy")
12
13 ### Section #4: Add tasks (Required)
14 workflow.add_task_group("metaphlan2.py [depends[0]] --input_type [extension] > [targets[0]]", depends=in_files,
15
16 ### Section #5: Run tasks (Required)
17 workflow.go()
```

But all of that runs with a single command

```
# all tasks are run
$ python myworkflow.py
```

```
# all tasks are skipped (because they were just run and everything is up-to-date)
$ python myworkflow.py
```

```
# all tasks are run (because of the flag applied)
$ python myworkflow.py --skip-nothing
```

```
(Dec 08 11:50:43) [0/4 -   0.00%] **Ready    ** Task 2: kneaddata
(Dec 08 11:50:43) [0/4 -   0.00%] **Started  ** Task 2: kneaddata
(Dec 08 11:50:43) [0/4 -   0.00%] **Ready    ** Task 0: kneaddata
(Dec 08 11:50:43) [0/4 -   0.00%] **Started  ** Task 0: kneaddata
(Dec 08 11:50:44) [1/4 -  25.00%] **Completed** Task 2: kneaddata
(Dec 08 11:50:44) [1/4 -  25.00%] **Ready    ** Task 5: humann2
(Dec 08 11:50:44) [1/4 -  25.00%] **Started  ** Task 5: humann2
(Dec 08 11:50:44) [2/4 -  50.00%] **Completed** Task 0: kneaddata
(Dec 08 11:50:44) [2/4 -  50.00%] **Ready    ** Task 4: humann2
(Dec 08 11:50:44) [2/4 -  50.00%] **Started  ** Task 4: humann2
(Dec 08 11:52:48) [3/4 -  75.00%] **Completed** Task 5: humann2
(Dec 08 11:52:49) [4/4 - 100.00%] **Completed** Task 4: humann2
Run Finished
```

bioBakery workflows

http://bitbucket.org/biobakery/biobakery_workflows

- A collection of “baked” workflows that execute common microbial community analysis.
- Currently supports 16S, metagenomic, and metatranscriptomic data.
  - Plus downstream visualization and statistics.

```
$ biobakery_workflows wmgx --input examples/wmgx/paired/ --output workflow_output
```

```
(Dec 08 14:33:07) [0/3 -   0.00%] **Ready    ** Task 4: kneaddata
(Dec 08 14:33:07) [0/3 -   0.00%] **Started  ** Task 4: kneaddata
(Dec 08 14:33:07) [0/3 -   0.00%] **Ready    ** Task 2: kneaddata
(Dec 08 14:33:07) [0/3 -   0.00%] **Started  ** Task 2: kneaddata
(Dec 08 14:33:07) [0/3 -   0.00%] **Ready    ** Task 0: kneaddata
(Dec 08 14:33:25) [0/3 -   0.00%] **GridJob  ** Task 4: kneaddata <Grid JobId 76918649 : Submitted>
(Dec 08 14:33:34) [0/3 -   0.00%] **GridJob  ** Task 2: kneaddata <Grid JobId 76918676 : Submitted>
(Dec 08 14:34:25) [0/3 -   0.00%] **GridJob  ** Task 4: kneaddata <Grid JobId 76918649 : PENDING>
(Dec 08 14:47:26) [0/3 -   0.00%] **GridJob  ** Task 4: kneaddata <Grid JobId 76918649 : PENDING>
(Dec 08 14:47:26) [0/3 -   0.00%] **GridJob  ** Task 4: kneaddata <Grid JobId 76918649 : Getting benchmarking data>
(Dec 08 14:47:35) [0/3 -   0.00%] **GridJob  ** Task 2: kneaddata <Grid JobId 76918676 : PENDING>
(Dec 08 14:47:35) [0/3 -   0.00%] **GridJob  ** Task 2: kneaddata <Grid JobId 76918676 : Getting benchmarking data>
(Dec 08 14:49:35) [0/3 -   0.00%] **GridJob  ** Task 4: kneaddata <Grid JobId 76918649 : Final status of COMPLETED>
(Dec 08 14:49:35) [0/3 -   0.00%] **GridJob  ** Task 2: kneaddata <Grid JobId 76918676 : Final status of COMPLETED>
(Dec 08 14:49:35) [0/3 -   0.00%] **Started  ** Task 0: kneaddata
(Dec 08 14:49:35) [1/3 -  33.33%] **Completed** Task 4: kneaddata
(Dec 08 14:49:35) [2/3 -  66.67%] **Completed** Task 2: kneaddata
(Dec 08 14:49:44) [2/3 -  66.67%] **GridJob  ** Task 0: kneaddata <Grid JobId 76922265 : Submitted>
(Dec 08 14:50:44) [2/3 -  66.67%] **GridJob  ** Task 0: kneaddata <Grid JobId 76922265 : Waiting>
(Dec 08 14:50:44) [2/3 -  66.67%] **GridJob  ** Task 0: kneaddata <Grid JobId 76922265 : Getting benchmarking data>
(Dec 08 14:54:46) [2/3 -  66.67%] **GridJob  ** Task 0: kneaddata <Grid JobId 76922265 : Final status of COMPLETED>
(Dec 08 14:54:46) [3/3 - 100.00%] **Completed** Task 0: kneaddata
Run Finished
```

bioBakery automated reports

Metagenome and Metatranscriptome Report

Project: UChicago

Date: 06/01/2017

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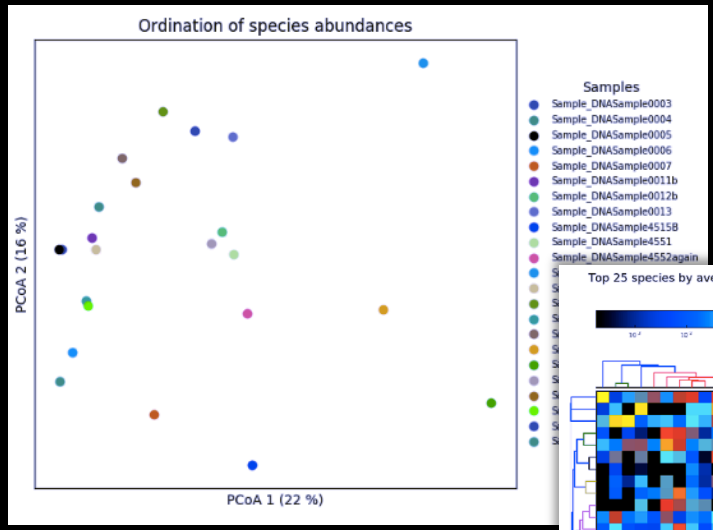

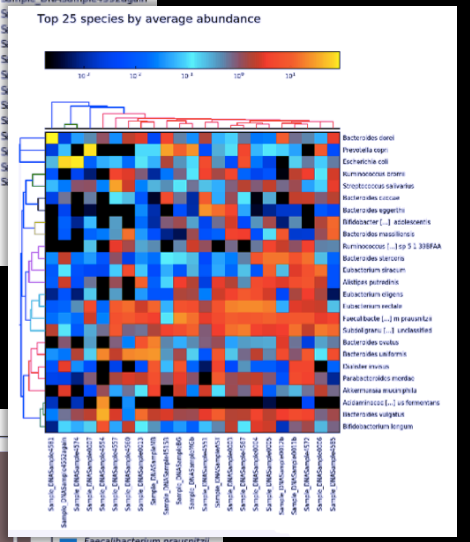

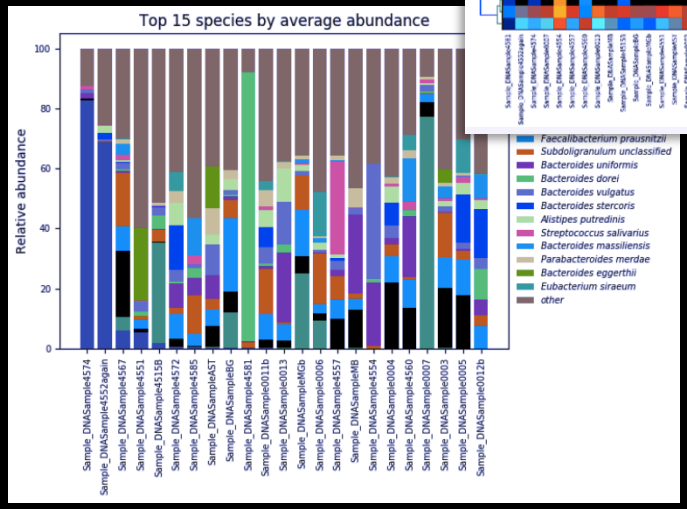

Packaging tools and building bioBakery

Individual tools packaged as Homebrew formulae

- **Simple to install**
- **Supports MacOS & Linux**
- **No root permissions required**
  - **Supports $HOME install**
- **Dependencies satisfied automatically**

Starting with base Ubuntu image, a master homebrew formula runs inside vagrant to install all tools…

…provisioning different "flavors" of virtual machine image

Packaging tools and building bioBakery

$ brew tap biobakery/biobakery

```
==> Tapping biobakery/biobakery
Cloning into '/home/user/.linuxbrew/Library/Taps/biobakery/homebrew-biobakery'...
remote: Counting objects: 23, done.
remote: Compressing objects: 100% (23/23), done.
remote: Total 23 (delta 11), reused 2 (delta 0), pack-reused 0
Unpacking objects: 100% (23/23), done.
Checking connectivity... done.
Tapped 19 formulae (65 files, 254.4K)
```

$ brew install maaslin

```
==> Installing maaslin from biobakery/biobakery
==> Downloading
https://bitbucket.org/biobakery/maaslin/get/ced3ca2a3b1d.tar.gz
==> R -q -e install.packages('agricolae',lib='/home/user/.linuxbrew/
==> R -q -e install.packages('gam', lib='/home/user/.linuxbrew/
==> R -q -e install.packages('gamlss', lib='/home/user/.linuxbrew/
==> R -q -e install.packages('gbm', lib='/home/user/.linuxbrew/
==> R -q -e install.packages('glmnet', lib='/home/user/.linuxbrew/
==> R -q -e install.packages('inlinedocs', lib='/home/user/.linuxbrew/
==> R -q -e install.packages('logging', lib='/home/user/.linuxbrew/
==> R -q -e install.packages('MASS', lib='/home/user/.linuxbrew/
==> R -q -e install.packages('nlme', lib='/home/user/.linuxbrew/
==> R -q -e install.packages('optparse', lib='/home/user/.linuxbrew/
==> R -q -e install.packages('outliers', lib='/home/user/.linuxbrew/
==> R -q -e install.packages('penalized', lib='/home/user/.linuxbrew/
==> R -q -e install.packages('pscl', lib='/home/user/.linuxbrew/
==> R -q -e install.packages('robustbase', lib='/home/user/.linuxbrew
==> R -q -e install.packages('tools', lib='/home/user/.linuxbrew/
/home/user/.linuxbrew/Cellar/maaslin/0.0.3-dev-ced3ca2a3b1d: 2,860 files,
44.6M, built in 2 minutes 31 seconds
```

http://huttenhower.sph.harvard.edu/biobakery

bioBakery workflows tutorial

https://bitbucket.org/biobakery/biobakery/wiki/biobakery_workflows