Outline for today - 23 May 2012

1. Some technical details of amplicon sequencing

2. Overview of analysis process

3. Overview of `phyloseq`

4. Live demo "hackalong" of phyloseq in R

Stats 366
Joey McMurdie guest lecture

Bioconductor workshop in July:

https://secure.bioconductor.org/BioC2012/

Amplicon Sequencing
Repeat many times with different samples/replicates
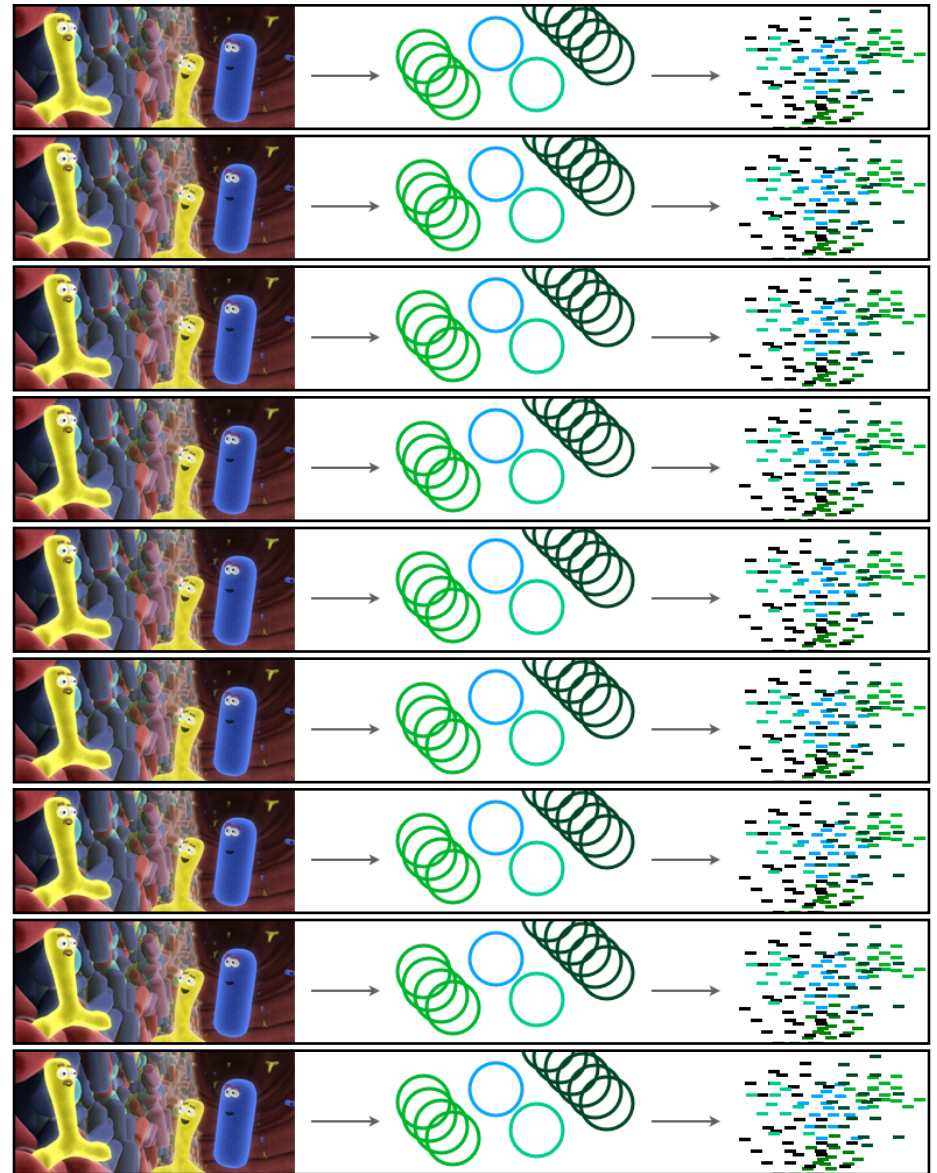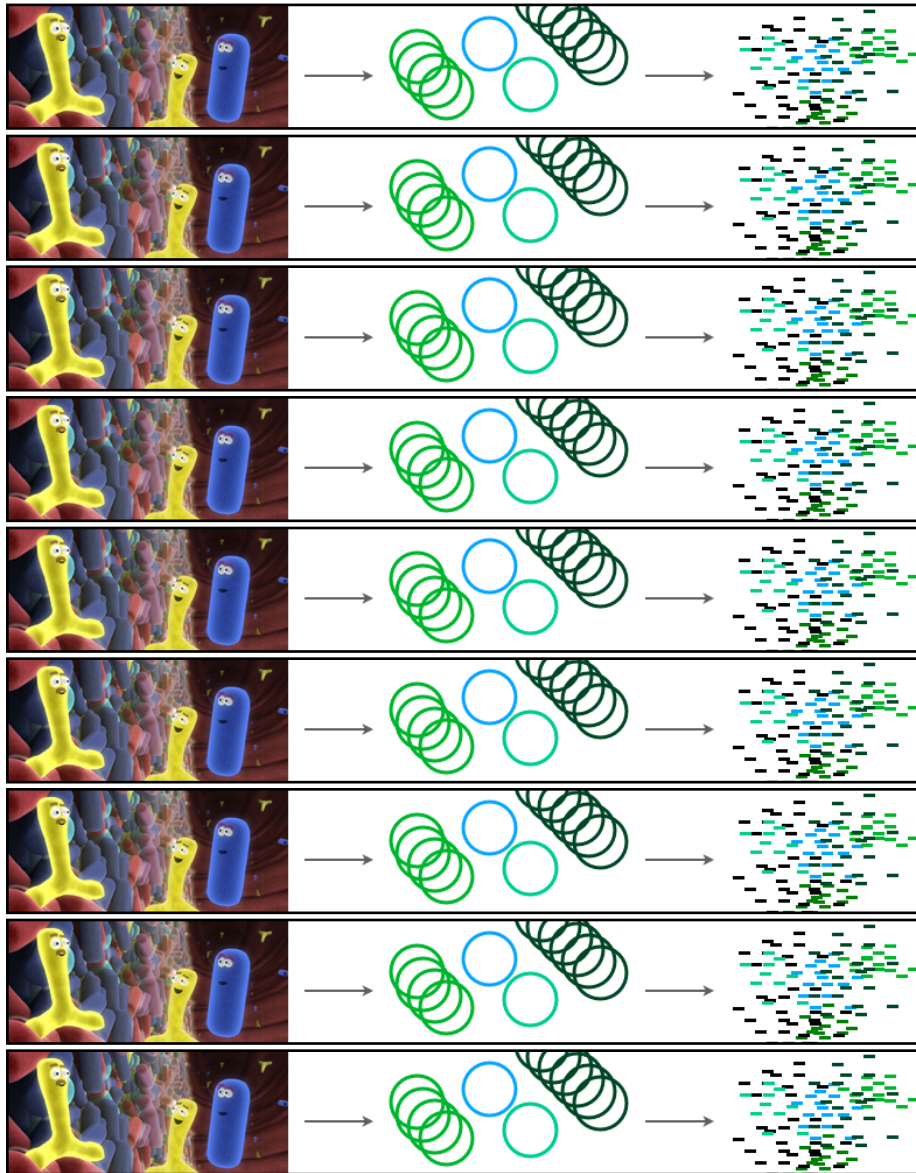
Amplicon Sequencing

parallel tagged sequencing "bar-coded" sequences

Binladen, *et al* (2007). The use of coded PCR primers enables. *PLoS ONE, 2*(2), e197. doi:10.1371/journal.pone.0000197

Amplicon Sequencing

Overview of amplicon sequencing and analysis
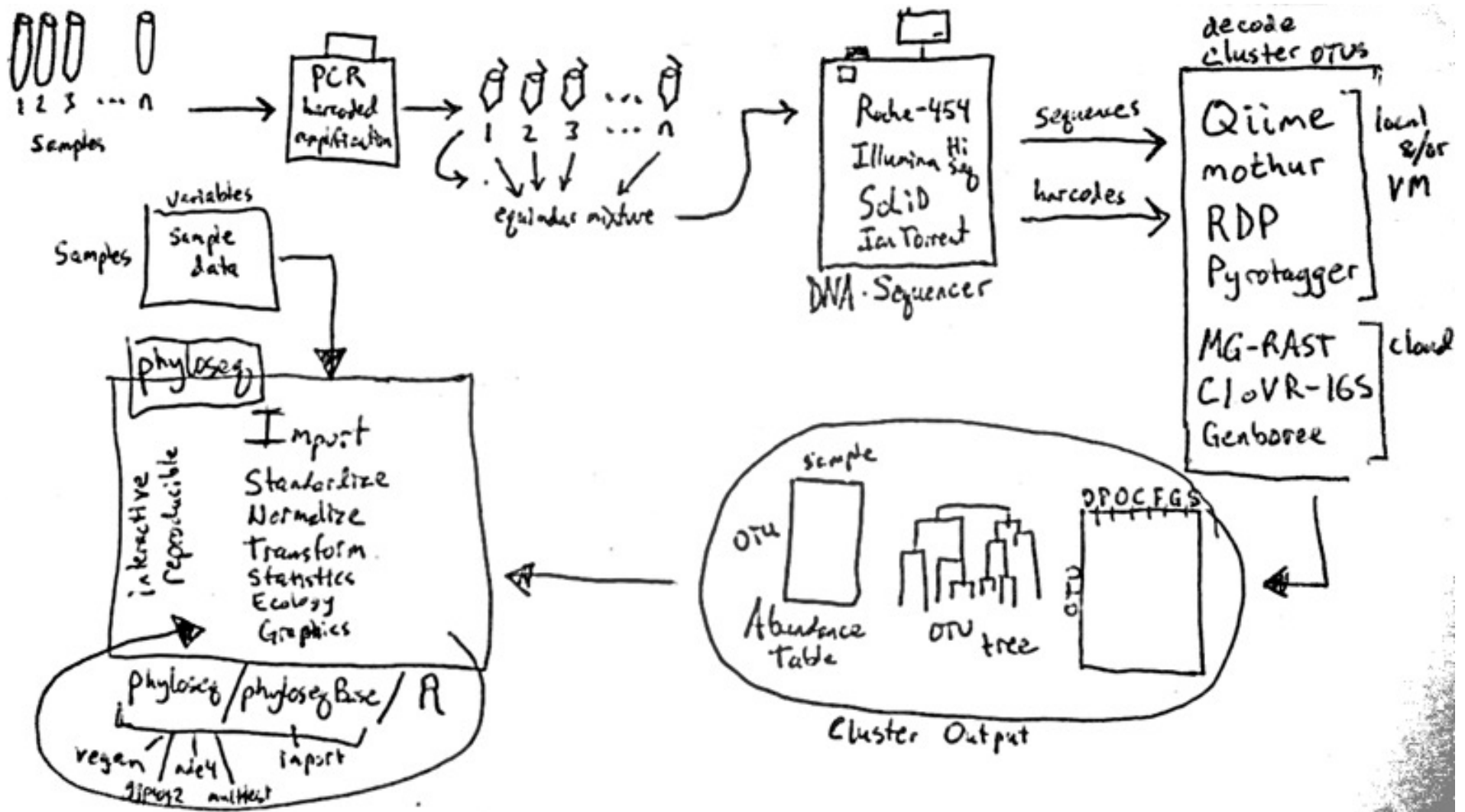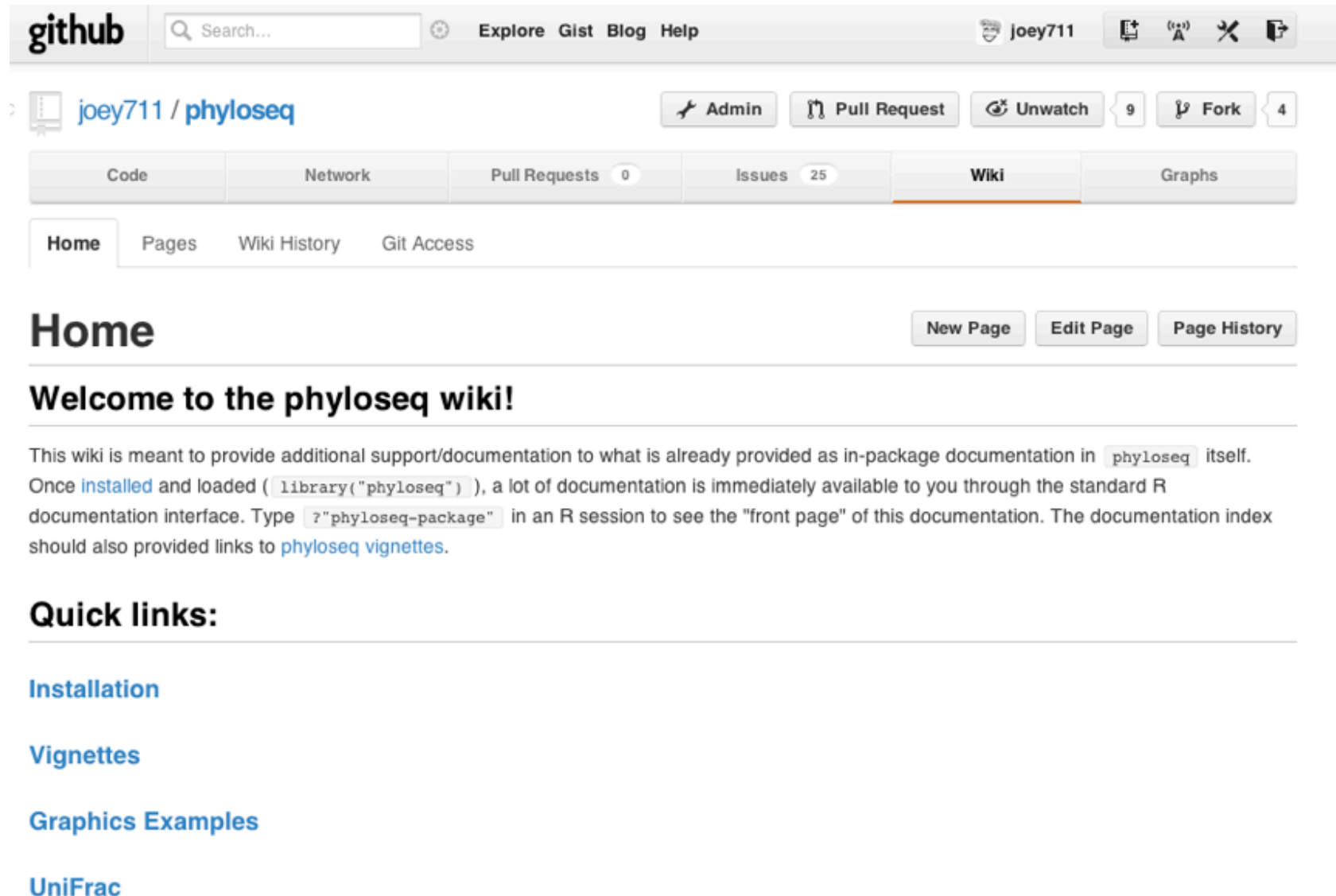
Overview of phyloseq

phyloseq wiki

https://github.com/joey711/phyloseq/wiki

phyloseq wiki

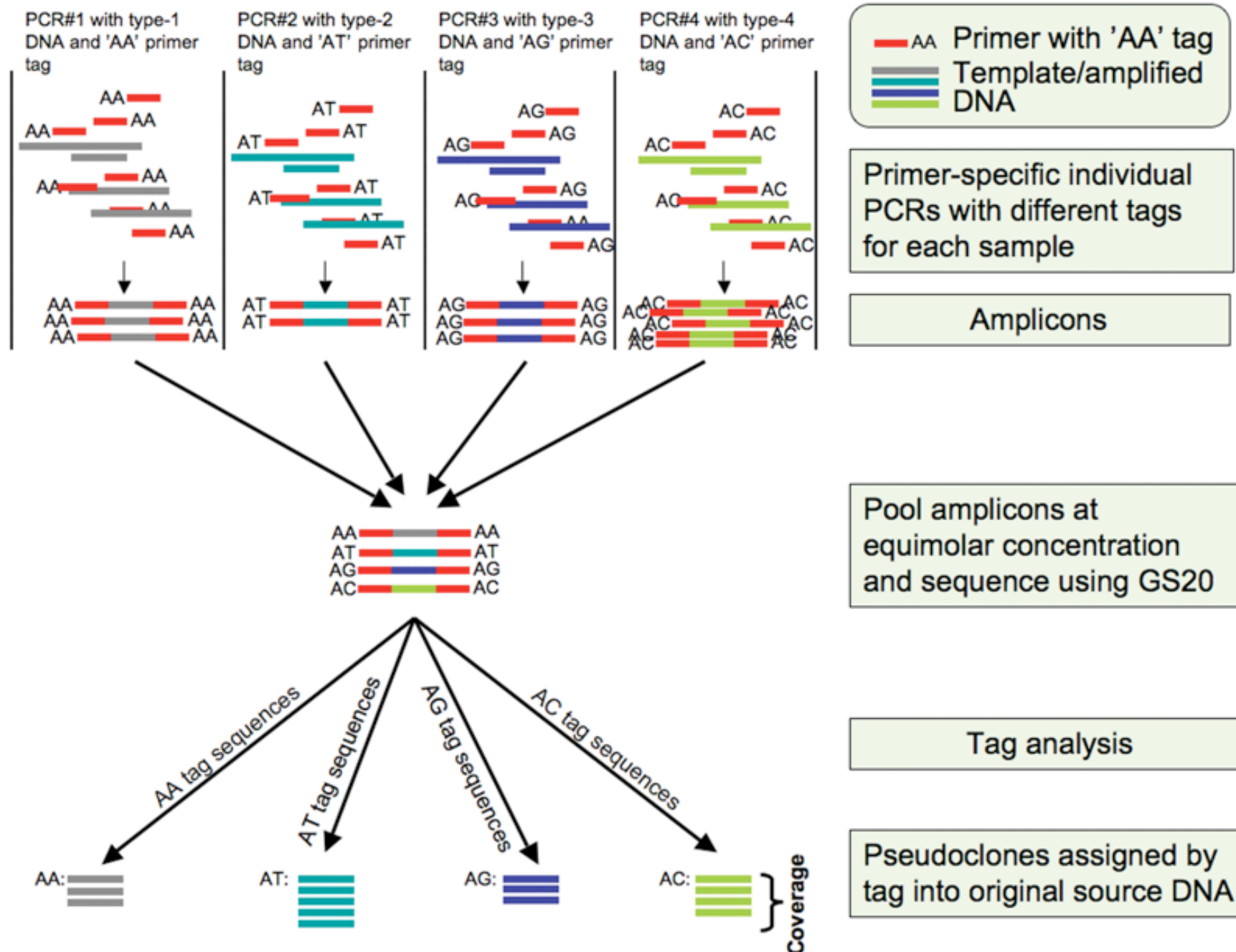https://github.com/joey711/phyloseq/wiki/Graphics-Examples

Graphics Examples

New Page | Edit Page | Page History

This page links you to a "show and tell" of wiki-pages describing different graphics options supported in the phyloseq-package, documented with exam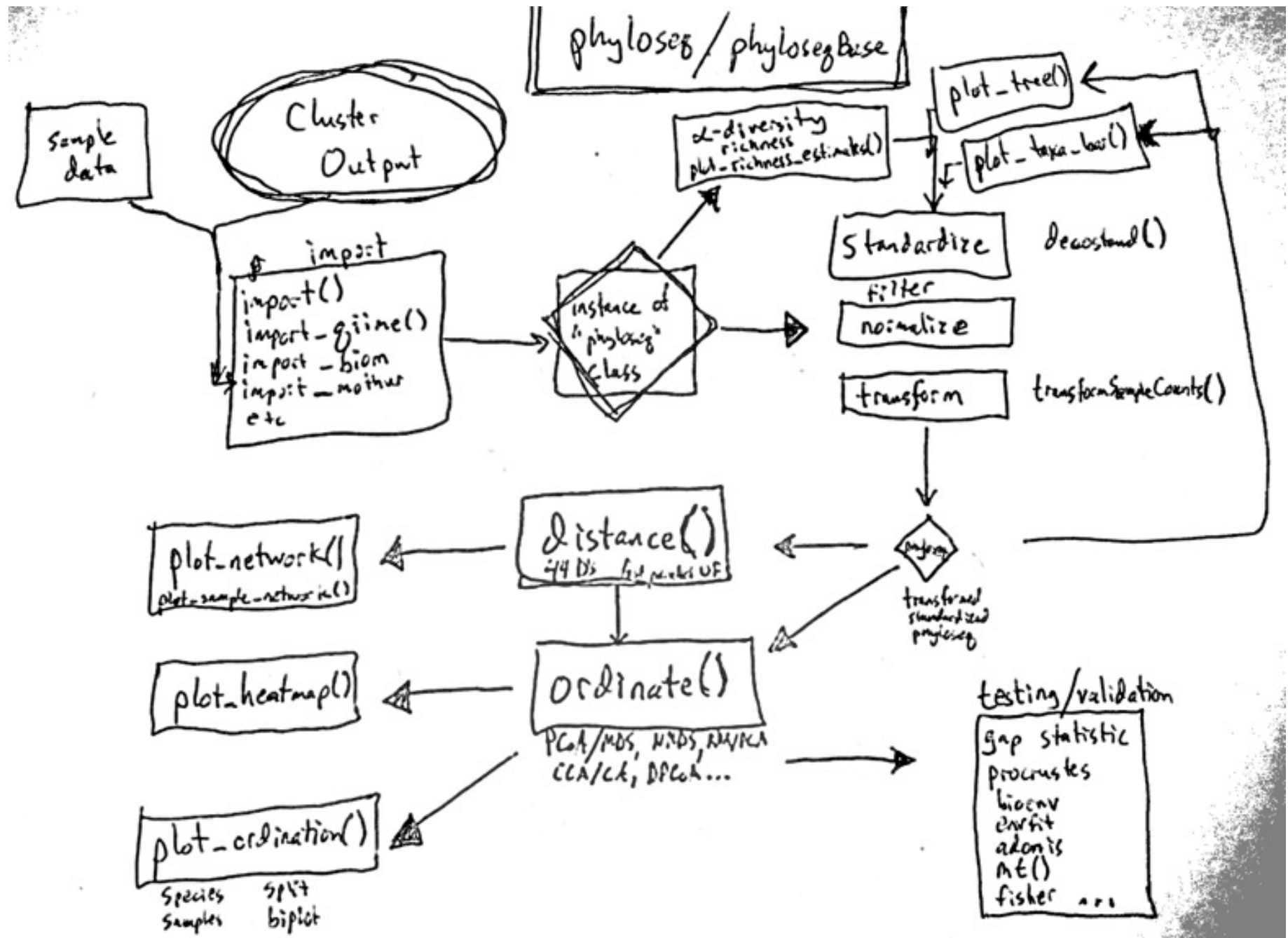ple code that for making the plot. If you have data of your own to try, it is strongly recommended that you attempt some of the examples on your own dataset as well, and see if you like the results. Many of the default settings are modifiable within the function arguments directly, and virtally everything about these plots can be further modified via the layers interface of ggplot2.

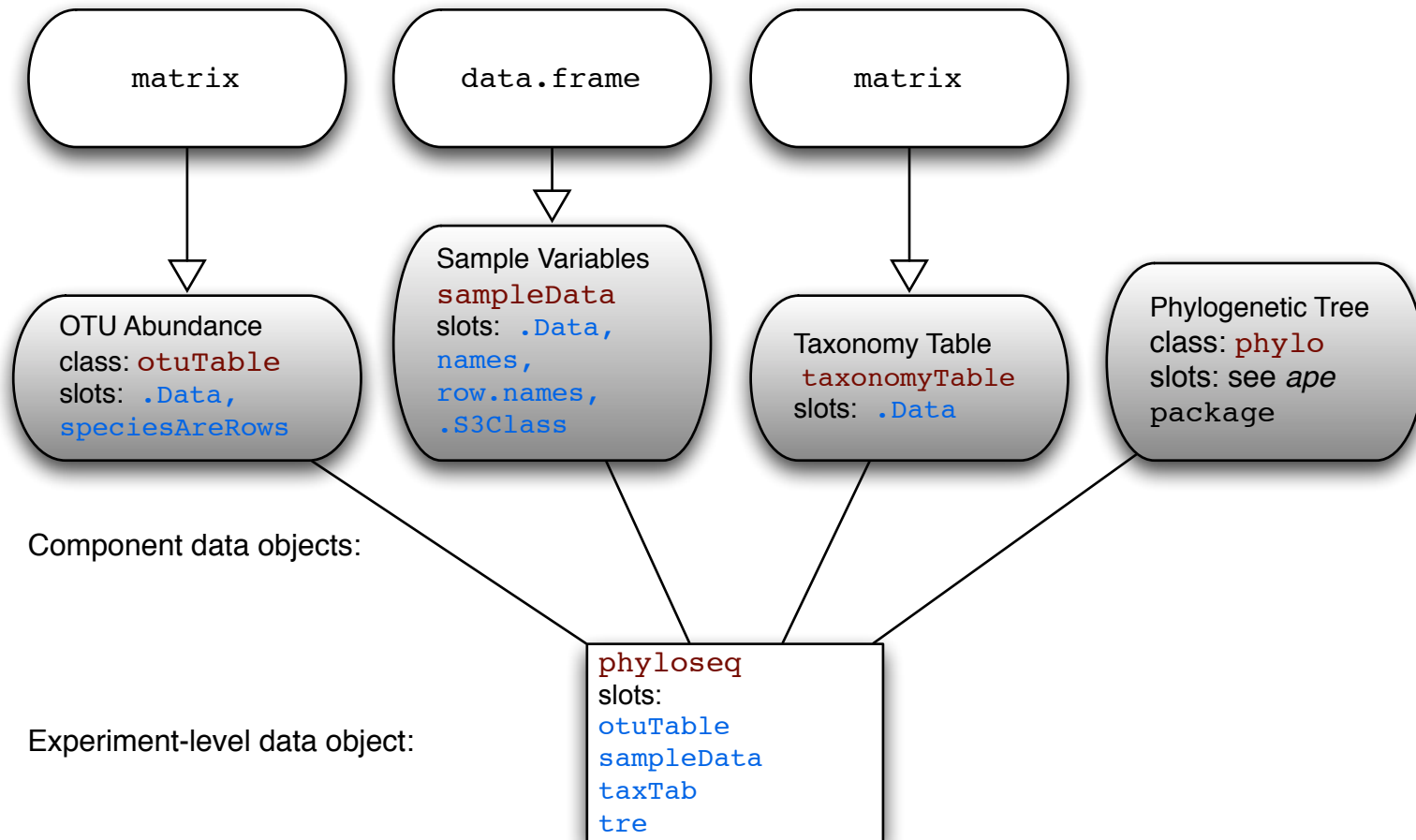Some key graphics-producing functions:

plot_heatmap

plot_tree

plot_ordination

plot_sample_network

plot_richness_estimates

plot_taxa_bar

phyloseq classes

phyloseq accessors

Function	Description
[	Standard extraction operator. works on otuTable, sampleData, and taxonomyTable
access	General slot accessor function for phyloseq-package
getslots.phyloseq	Return the slot names of phyloseq objects
getSpecies	Returns the abundance values of sample 'i' for all species in 'x'
getSamples	Returns the abundance values of species 'i' for all samples in 'x'
getTaxa	Get a unique vector of the observed taxa at a particular taxonomic rank
getVariable	Returns an individual sample variable vector/factor
nsamples	Get the number of samples described by an object
nspecies	Get the number of species (taxa) described by an object
otuTable	Build or access otuTable objects
rank.names	Get the names of the available taxonomic ranks
sampleData	Build or access sampleData objects
sample.names	Return the names of the samples described by an object
species.names	Return the names of the species described by an object
sampleSums	Returns the total number of individuals observed from each sample
sample.variables	Returns the names of sample variables in an object
speciesSums	Returns the total number of individuals observed from each species
speciesAreRows	Returns the orientation of the abundance table
taxTab	Build or access taxTab objects
tre	Access the tree contained in a phyloseq object

phyloseq constructors

Functions for building component data objects

Function	Input Class	Output Description
`otuTable`	numeric matrix	`otuTable` object storing taxa abundance
`otuTable`	data.frame	`otuTable` object storing taxa abundance
`sampleData`	data.frame	`sampleData` object storing sample variables
`taxTab`	character string	`taxonomyTable` object storing taxonomic identities
`tre`	file path char	phylo4-class tree, read from file
`tre`	phylo-class tree	phylo4-class tree, converted from argument
`read.table`	table file path	A matrix or data.frame (Std Rcore function)
`read.tree`	Newick file path	phylo-class tree object (ape)
`read.nexus`	Nexus file path	phylo-class tree object (ape)
`readNexus`	Nexus file path	phylo4-class tree object (phylobase)

Functions for building complex data objects

Function	Input Class	Output Description
`phyloseq`	2 or more component objects	phyloseq-class, "experiment-level" object
`merge_phyloseq`	2 or more component or phyloseq-class objects	Combined instance of phyloseq-class

Importing Data

```
import()
import_biom()
import_qiime()
import_mothur()
import_pyrotagger_tab()
import_RDP_cluster()
import_RDP_otu()

Soon:
import_mgrast()
import_clovr16s()
import_genboree()
```

Example Data

(Begin live demo)