Phylogenomics

Work locally on your laptop

The following instructions have been tested on Ubuntu. Mac and Windows users can use Ubuntu in the Virtual Machine.

- 1. Download MEGA 7:
 - Go to the website http://www.megasoftware.net/ and download the appropriate MEGA7
 - To get help:
 - **GUI:** http://www.megasoftware.net/docs or menu Help → Contents
 - CC: http://www.megasoftware.net/pdfs/MEGA7-CC-Quick-Start-Tutorial.pdf or type man megacc
 - 1. use megaproto to create analysis options files
 - 2. run megacc -a megaOptionFile.mao -d dataFile.meg [options] to run MEGA analyses
- 2. Download RAxML:
 - Download RAxML under https://github.com/stamatak/standard-RAxML
 or type sudo apt-get install raxml

Exercise 1: MEGA

In this exercise you will work with an example file, which is included in the MEGA distribution: mtCDNA.meg (can be found in the Examples folder of MEGA, which is located in your home (Mac) or Documents (Windows) directory). It can also be downloaded from http://www.megasoftware.net/examples/mtCDNA zip). This data set contains mitochondrial cDNA sequences of several primate species.

- a) Open the mtCDNA.meg file with MEGA
- b) Find the appropriate substitution model for this data set with MEGA. Try to understand the result table.
- c) Create a NJ, maximum parsimony and maximum likelihood tree. Compare the different phylogenies

BIO634 – Next-Generation Sequencing 2

d) Try to time calibrate the maximum likelihood tree by clicking on "Compute" → "Timetree".
 You know that Gibbon is the outgroup and that chimpanzee and bonobo diverged between 1.2 –
 5.0 Ma. Additionally you know that orangutan and Sumatran diverged latest 13 Ma.

Exercise 2: RAxML

a) Convert the .meg file to a RAxML input file (relaxed PHYLIP format) using my converter program PGDSpider (http://www.cmpg.unibe.ch/software/PGDSpider/).

Hint: In "PHYLIP (RAxML) – Writer Questions" you have to specify that you want to save a relaxed PHYLIP file!

(if everything goes wrong:

```
wget https://www.dropbox.com/s/sk545mqslufjosi/mtCDNA.txt)
```

b) Estimate a maximum likelihood tree using RAxML by specifying the input file (-s), the substitution model (-m) and the output file name (-n) (for further options type raxmlHPC -h):

```
raxmlHPC -p 1 -T 2 -s mtCDNA.txt -n out.nwk -m GTRCATI
```

c) Run a bootstrap analysis in RAxML:

```
raxmlHPC -p 1 -m GTRCATI -b 1234 -N 100 -s mtCDNA.txt -n out2.nwk (the -b turns on bootstrapping with 1234 as seed and -N specifies the number of runs)
```

d) This can now be used to put bootstrap values on the maximum likelihood tree:

```
raxmlHPC -m GTRCATI -f b -t RAxML_bestTree.out.nwk -z
RAxML_bootstrap.out2.nwk -n out_bootstrap.nwk
```

e) Open the resulting file (e.g. with MEGA) and compare it with the maximum likelihood tree of MEGA