

A Guide to the MLTreeMap imagemaker:

MLTreeMap is a software framework, designed for phylogenetic and functional analysis of metagenomics data. It searches for instances of marker genes on nucleotide sequences and deduces their most likely origin in a set of reference phylogenies. The MLTreeMap imagemaker has been designed to visualize your placements within the reference phylogenies. It is not part of the MLTreeMap stand-alone package and has to be installed individually.

A) Installation of the MLTreeMap imagemaker

The MLTreeMap imagemaker is based on Perl scripts and has the following dependencies:

```
GD
Math::Trig
```

If these dependencies are satisfied, the MLTreeMap imagemaker is ready to use.

B) Usage of the MLTreeMap imagemaker

The MLTreeMap imagemaker has to be accessed on the command line and needs MLTreeMap output files as input. An example for valid input files is given in the directory "example_input".

An example for a valid input command is: `./mltreemap_imagemaker.pl -i example_input`

This will concatenate all files of the same analysis type into one file and then generate the pictures (they will be written to the standard output directory `MLTreeMap_imagemaker_2_03/output/`).

If you want to enter a single file as input, this is also possible:

```
./mltreemap_imagemaker.pl -i
example_input/p_Aeropyrum_ernix_b.txt_contig158_RAxML_parsed.txt
```

Optional input option:

- o use this option if you want to write the output to another directory than the default.
- b parameter for the size of the placement bubbles (default: 0.9).
- d use different colors for different datasets (0: don't use this mode (default), 1: use this mode). See below for a more detailed description.

C) Detailed information to the -d input option.

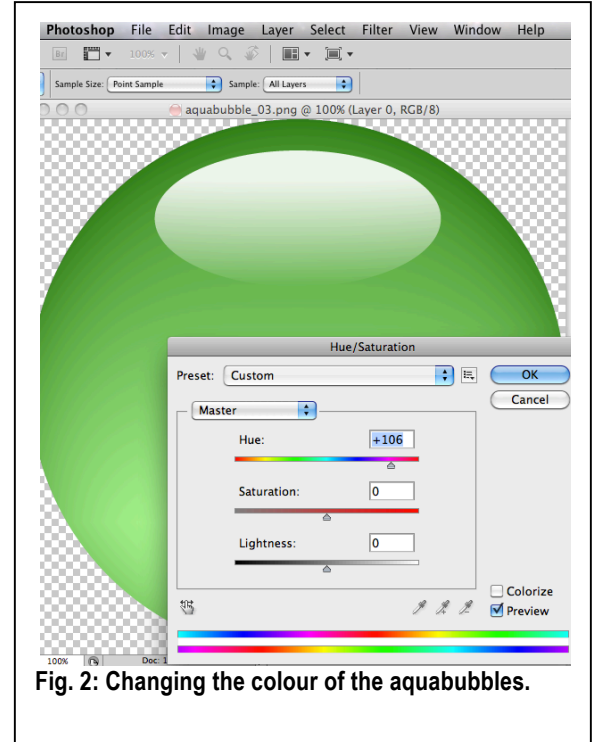
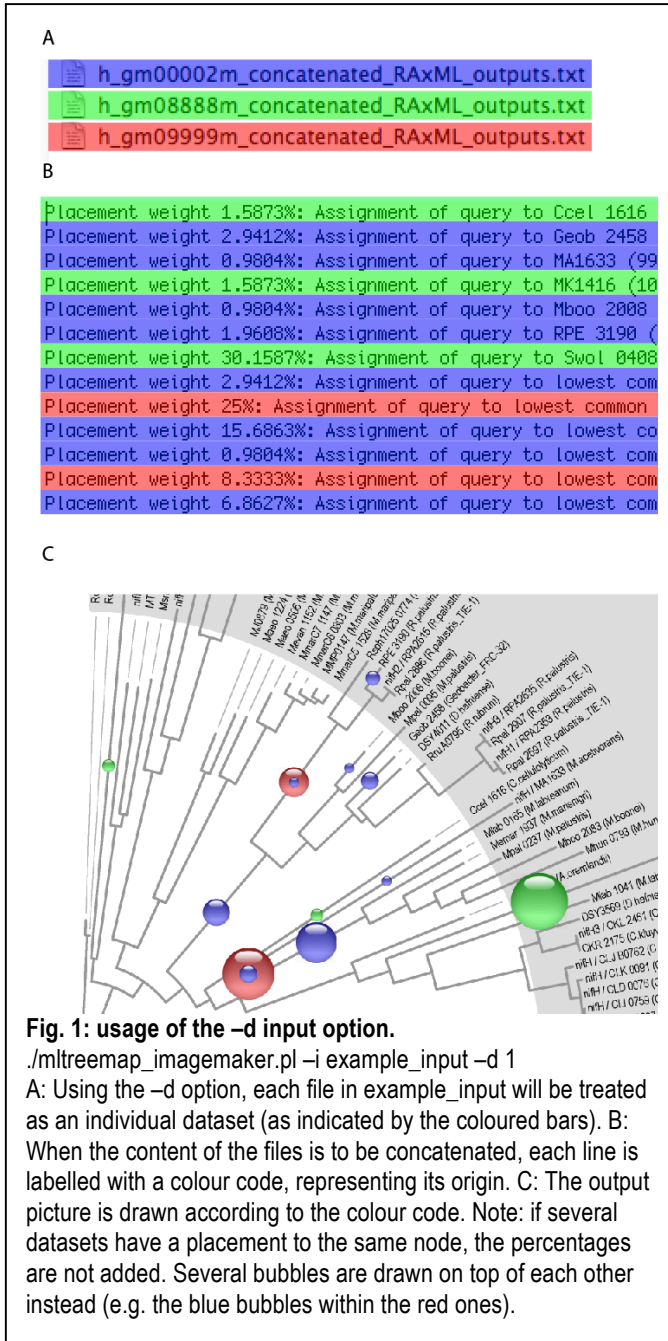
The -d option is designed to print the concatenated results from different datasets into one picture, while each dataset is represented by an own colour. For the principle see Fig. 1.

This mode works only if you enter a directory as input e.g. `-i example_input` (if you enter only one file as input, the -d option is irrelevant).

Each file in the input directory will be regarded as an independent dataset and assigned a different colour in the final picture.

Currently the imagemaker supports up to 4 datasets. Thus if your input directory contains more than 4 files of the same kind of analysis (p: phylogenetic, g: phylogenetic with the GEBA tree, h: functional nifH, n: functional nifD, r: functional RuBisCo, m: functional MMO) the program will die with an error message.

It is however very easy to increase this number:



The limit of supported datasets is set by the number of available “aquabubbles”. You will find them in the directory tree_data. They are called aquabubble_xx.png (xx representing the numbers 01-04). To increase the number of supported datasets just copy one of the bubbles as many times as you need and rename them with aquabubble_yy.png (while yy being a number > 04).

After having created the new bubbles, you have to change their colour. For this I suggest a graphic program such as Photoshop, on which the description of the last step is based:

In the menu bar you will find the entry “Image”. There chose “Adjustments” and then “Hue/Saturation”. Now play around with the “Hue” roll bar and you will be able to colour the bubble as you want (Fig. 2).