# Taxonomic classification using **pplacer**, *clst*, and *clstutils*

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### 1 Introduction

This vignette assumes that you have already created a reference package, and have used it to run pplacer against an alignment of reference sequences. For instruction on performing the above operations, see the documentation for pplacer here: http://matsen.fhcrc.org/pplacer/

## 2 Input files

The following input is required (file names defined as variables in parentheses):

```
1. a reference package (refpkg)
```

- 2. a pplacer file created using the same reference package (placefile)
- 3. a file providing distances between nodes in the reference tree (distfile)

Note that distfile is generated from placefile using placeutil (distributed with pplacer).

```
> library(clstutils)
> expand <- function(fname){</pre>
    orig.dir <- getwd()</pre>
+
   destdir <- tempdir()
+
+
    setwd(destdir)
    archive <- system.file('extdata', 'vaginal_16s.refpkg.tar.gz', package='clstutils')</pre>
+
   system(sprintf('tar --no-same-owner -xzf "%s"', archive))
+
+
    setwd(orig.dir)
+
    file.path(destdir, fname)
+ }
> refpkg <- expand('vaginal_16s.refpkg')</pre>
> placefile <- system.file('extdata','merged.json', package='clstutils')</pre>
> distfile <- system.file('extdata','merged.distmat.bz2', package='clstutils')</pre>
```

### 3 Reading the input

Classification requires a matrix representation of distances between "objects" being classified, in this case sequences in a phylogenetic tree. treeDists returns a list containing matrix representations of distances between internal and terminal edges (\$dists and \$paths), and \$dmat, a square matrix of distances between terminal edges.

```
> treedists <- treeDists(distfile=distfile, placefile=placefile)</pre>
```

We also need a description of the taxonomy of the reference sequences. This is read from the reference package using taxonomyFromRefpkg. The seqnames argument ensures that the output is arranged in an order compatible with treedists. We indicate that the most specific rank that we want to consider is "species" using lowest\_rank.

> taxdata <- taxonomyFromRefpkg(refpkg, seqnames=rownames(treedists\$dmat), lowest\_rank='species')

#### 4 Classification of a single sequence

Given the distances and taxonomic information describing the reference tree, the only additional data required to perform classification is the position of a sequence placed onto a tree. At a minimum, this consists of a data.frame with columns **at**, **edge**, and **branch**. This data will be used to generate a vector of branch lengths between the query sequence and each of the reference sequences on the tree.

> placetab <- data.frame(at=49, edge=5.14909e-07, branch=5.14909e-07)

The function classifyPlacements is a wrapper around *clst*::classifyIter. The output is a data.frame describing the taxonomic assignment, along with a description of the confidence of the classification. See the man page for *clst*::classify for details on the output.

```
> cdata <- classifyPlacements(taxdata, treedists, placetab)
> cdata
tax_id tax_name rank below above score match min median max
1 2702 Gardnerella vaginalis species 10 0 0.95 1 0 0.06 0.1
    d at
1 0.16 49
```