Install:

Analyzing microbial communities using high-throughput 16S rRNA sequencing data.

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1. Installing QIIME

To use this tutorial the user should install the following programs and set the appropriate environment variables, before downloading and installing QIIME.

1.1 Tutorial Software Dependencies

The following programs and datasets were used to generate this tutorial and it is recommended to use the same versions as shown below.

- Python 2.6
- PyCogent (from svn) with ReportLab
- PyNAST
- Numpy 1.3
- MatPlotLib 0.98.5.2
- jre1.6.0_16
- rdp_classifier-2.0
- Green_genes_core_set
- blast-2.2.21
- fasttree 2.0
- cd-hit 3.1

1.2 Installing QIIME

To install QIIME, the user must download the source code from sourceforge:

svn co https://qiime.svn.sourceforge.net/svnroot/qiime /path/to/download/qiime

1.3 Environment Variables

Make sure the following environment variable are set:

1.3.1 PATH Environment Variable

Your PATH variable should contain the following:

- /path/to/python/bin/
- /path/to/cd-hit/
- /path/to/FastTree/executable
- /path/to/blast-2.2.21/bin/
- /path/to/PyNAST/scripts/
- /path/to/jre1.6_0_16/

For all path description throughout this tutorial, the "/path/to/" refers to the physical location of each program on your local computer. For instance, the "/path/to/python/bin/" refers to "/Library/Frameworks/Python.framework/Versions/2.5/bin" on Mac OS X version 10.5.

In the bash shell, you can use the following command (example only shows the path to python, so other softwares can be added to the PATH, using a similar approach):

- export PATH=/path/to/python/bin/:$PATH

1.3.2 PYTHONPATH Environment Variable

Your PYTHONPATH should contain the following:

- /path/to/PyCogent
- /path/to/QIIME
- /path/to/PyNAST

In the bash shell, you can use the following command (example only shows the path to QIIME, so other softwares can be added to the PYTHONPATH, using a similar approach):

- export PYTHONPATH=/path/to/QIIME/:$PYTHONPATH

1.3.3 RDP_JAR_PATH Environment Variable

The user should also define an RDP_JAR_PATH variable, since this tutorial uses the RDP Classifier:

- /path/to/rdp_classifier-2.0.jar

In the bash shell, you can use the following command:

- export RDP_JAR_PATH=/path/to/rdp_classifier-2.0.jar

1.4 Testing QIIME Install

Once the source code is downloaded, the user should test QIIME to be sure all essential software is properly installed and the correct environment variables are set.

In a terminal window the user, should cd to their qiime/test directory using the following command:

```
cd /path/to/QIIME/tests/
```

Then run the following test command:

```
python all_tests.py -v
```

If all tests run properly, then QIIME was properly installed. Three of the test scripts may fail, but for this tutorial we are not using all the supported 3rd party applications which are supported, so we can disregard these errors. The following scripts may generate errors:

1. **test_align_seqs.py** - Make sure that the cd-hit test ran properly by parsing the traceback generated in . For this tutorial we are only using PyNAST to align sequences, so we are not worrying about installing MOTHER, DOTUR or MUSCLE at this time.
2. **test_pick_otus.py** - Make sure the cd-hit tests ran properly by parsing the traceback generated in your terminal window. For this tutorial we are only using cd-hit to pick OTUs, so we are not worrying about installing BLAST at this time.
3. **test_pyronoise.py** - For this tutorial, we are not denoising the sequences, so disregard the errors generated for this script.