Installation

MSG currently runs on any linux platform.

Place msg-version.tar.gz in any directory: \$ tar zxf msg-version.tar.gz

- \$ cd msq_version/
- \$ make

Please ensure that the following dependencies are installed before running MSG (working versions are indicated in parentheses):

- Python (2.6.4)
- bwa (0.5.7)
- samtools (0.1.9-3)
- biopython-1.53
- Pyrex-0.9.9
- pysam-0.1.2 (apply fix*)
- R packages (HiddenMarkov 1.3-1, zoo 1.6-2, R.methodsS3 1.2.0 and R.oo 1.7.3)

Setting up the MSG analysis directory

Note that all data files must be located within your MSG analysis directory (links to files are acceptable). Also note that quality values in the sequence fastq files must be in Sanger format.

- 1. Create a text file called "msg.cfg". This file will specify the location of your data files, and a few other details. You can find an example of an msg.cfg file here:
 - http://genomics.princeton.edu/AndolfattoLab/MSG_files/msg.cfg

or

https://github.com/tinathu/msg/blob/master/example/msg.cfg.

2. Create a barcode file. You can find an example of a barcode file here: http://genomics.princeton.edu/AndolfattoLab/MSG_files/barcodes_file.txt

or

https://github.com/tinathu/msg/blob/master/example/barcodes_file.txt

- 3. Create (or download) two parental reference genomes in fasta format (links to examples are given at the end of this document).
- 4. Download read data from an MSG library for a backcross experiment and/or parental genomes (links to examples are given at the end of this document).
- 5. Create a link to the msg software within your MSG analysis directory:

```
$ In -s <path_to_msq> msq
```

6. To run MSG, simply type the following from within your MSG analysis directory:

```
$ perl msq/msqCluster.pl
```

^{*}http://code.google.com/p/pysam/issues/detail?id=22&can=1&q=dandavison0

Sample data

Short-read Illumina data set from manuscript

F1-parental backcross data ftp://ftp-trace.ncbi.nlm.nih.gov/sra/sra- instant/reads/ByExp/sra/SRX/SRX029/SRX029935/SRR071201/

Parental data for Dsim_w501 ftp://ftp-trace.ncbi.nlm.nih.gov/sra/sra- instant/reads/ByExp/sra/SRX/SRX032/SRX032362/SRR074287/

Parental data for Dsec_w1 ftp://ftp-trace.ncbi.nlm.nih.gov/sra/sra-instant/reads/ByExp/sra/SRX/SRX032/SRX032363/SRR074288/

To convert these to fastq format, download the SRA Toolkit (http://trace.ncbi.nlm.nih.gov/Traces/sra/sra.cgi?cmd=show&f=software&m=software&s=software) and type something like:

./<path to fastq-dump> -A <sra accession number> -D <path to sra file> -O <output directory> &

e.g.

./sratoolkit.2.0rc4-mac64/fastq-dump -A SRR071201 -D sra/SRR071201.sra -O fastq &

Reference genomes

D. simulans reference genome tp://ftp.flybase.net/genomes/Drosophila_simulans/current/fasta/dsim-all-chromosome-r1.3.fasta.gz

D. sechellia reference genome tp://ftp.flybase.net/genomes/Drosophila_sechellia/current/fasta/dsec-all-chromosome-r1.3.fasta.gz

Example barcode file

http://genomics.princeton.edu/AndolfattoLab/MSG_files/barcodes_file.txt or https://github.com/tinathu/msg/blob/master/example/barcodes_file.txt