**General syntax notes**

* **~ (the tilde)** is a shortcut for your home directory. This is generally something like /Users/NAME/ (Mac) or C:/Users/NAME/ (Windows). We will use the tilde throughout the code here but if you are having issues with it, replace the tilde with your full home directory path name.
  + Note that throughout these instructions, we give example file paths including **NAME**. You cannot use these directly in your terminal. You need to change NAME to your username instead.
* Your command line terminal opens in this **home directory**. You can see the full home path by running pwd (print working directory) in a new terminal window
* The terminal does not like **special characters** in directory and file names. This includes spaces. If at all possible, install and save everything to a path without any special characters in it. If this is unavoidable (like your username has a space), you can use the backslash \ before the special character. For example, my home directory might be C:/Users/first\ lastname/
* In general, a **WARNING** is okay, but an **ERROR** or **FAILURE** is not. ERROR means the command did not run. When you get an error, stop and determine if you can fix the issue (like a typo) and if not, contact Kim for help.

**Install git and SEAsnake**

1. Install git
   1. Windows: <https://gitforwindows.org/>
   2. Mac: <https://git-scm.com/book/en/v2/Getting-Started-Installing-Git>
2. Use git to download SEAsnake (our RNAseq pipeline)
   1. Open a terminal window
      1. Windows: You will use the terminal that came with git, not the one that came with Windows. This terminal is called “Git Bash”. Search for this in your finder.
      2. Mac: Go to Applications/Utilities/Terminal
   2. Navigate to where you want to download the files
      1. You move around your files in the terminal with cd (change directory)
      2. For example, you can move to the Desktop with cd ~/Desktop
   3. Make a directory for SEAsnake and navigate into it

mkdir SEAsnake

cd SEAsnake

* 1. Download SEAsnake by running the following command in the terminal
     1. git clone https://github.com/BIGslu/SEAsnake .
     2. If this for some reason fails, you can download with the big green code button at <https://github.com/BIGslu/SEAsnake>

**Install conda and mamba**

1. Install anaconda for your operating system <https://www.anaconda.com/docs/getting-started/anaconda/install>
2. WINDOWS ONLY. Setup the path to conda in the terminal. Skip to #5 if using a Mac.
   1. Open a Git Bash terminal window
   2. Navigate to where you installed anaconda. For example, something like

cd C:/Users/NAME/anaconda3

* 1. Navigate to the profile.d directory

cd etc/profile.d

* 1. Add conda to your path by running the following in the terminal

echo ". '${PWD}'/conda.sh" >> ~/.bashrc

1. Activate the path changes by running the following in the terminal

source ~/.bashrc

1. Check that conda works by running conda in the terminal. You should see the conda help page. If you instead see something like “command not found”, contact Kim for assistance before moving forward.
   1. If this does not work and you must move on now without fixing it, you can give the full file path *every time* you run conda like

Windows: ~/anaconda3/condabin/conda.bat

Mac: ~/anaconda3/bin/conda

1. Install mamba with the following command. This can take several minutes.

conda install -n base -c conda-forge mamba -y --repodata-fn repodata.json

1. WINDOWS ONLY. Setup path to mamba. Skip to #8 if using a Mac
   1. Navigate to where you installed anaconda. For example, something like

cd C:/Users/NAME/anaconda3

* 1. Navigate to the condabin directory

cd condabin

* 1. Add mamba to your path by running the following in the terminal

echo ". '${PWD}'/mamba.bat" >> ~/.bashrc

1. Activate the path changes by running the following in the terminal

source ~/.bashrc

1. Check that mamba works by running mamba in the terminal. You should see the mamba help page. If you instead see something like “command not found”, contact Kim for assistance before moving forward.
   1. If this does not work and you must move on now without fixing it, you can give the full file path *every time* you run mamba like

Windows: ~/anaconda3/condabin/mamba.bat

Mac: ~/anaconda3/bin/mamba

**Install bioinformatics software**

We will use conda and mamba to more easily install all the software for SEAsnake.

Option 1: Install with mamba

1. Open the file in SEAsnake/environment/Hissss\_env.yaml in any text editor
2. Replace - adapterremoval >=2.3.2 with - cutadapt >=5.0
3. Save and close the file
4. Open a terminal window (Terminal for Mac, Git bash for Windows)
5. Navigate to within the SEAsnake directory you downloaded early. For example

cd ~/Desktop/SEAsnake

1. Run mamba installation of all the SEAsnake softare with the following

mamba env create --name SEAsnake2 --file environment/Hissss\_env.yaml

Option 2: Install with conda (only use if Option 1 fails)

1. Open a terminal window (Terminal for Mac, Git bash for Windows)
2. Navigate to within the SEAsnake directory you downloaded early. For example

cd ~/Desktop/SEAsnake

1. Create a conda environment

conda create --name SEAsnake

1. Activate that environment

conda activate SEAsnake

1. Make sure you are in the environment. You should see a \* next to SEAsnake

conda env list

1. Setup channels for software install

conda config --add channels defaults

conda config --add channels bioconda

conda config --add channels conda-forge

~~conda config --set channel\_priority strict~~

conda config --set channel\_priority flexible

1. Install each software individually. If one fails, save the error to a text file to share with Kim and move on to the next program

conda install -y bioconda::fastqc

conda install -y bioconda::bcftools

conda install -y bioconda::samtools

conda install -y bioconda::bwa

conda install -y bioconda::bedtools

conda install -y bioconda::subread

conda install -y bioconda::star

conda install -y bioconda::picard

conda install -y bioconda::snakemake-minimal

conda install -y bioconda::pysam

conda install -y bioconda::adapterremoval

conda install -y bioconda::cutadapt

conda install -y anaconda::jinja2

conda install -y anaconda::networkx

conda install -y anaconda::graphviz

conda install -y anaconda::matplotlib

1. Exit the conda environment before closing your terminal

conda deactivate

**Install Amazon command line interface (CLI)**

1. Install the CLI for your operating system following instructions. <https://docs.aws.amazon.com/cli/latest/userguide/getting-started-install.html#getting-started-install-instructions>

Install R and RStudio

Packages

For Kim only

conda env remove –-name SEAsnake

Git Bash

C:/Users/NAME

/c/Users/NAME

# >>> conda initialize >>>

# !! Contents within this block are managed by 'conda init' !!

\_\_conda\_setup="$('/Users/kadm/anaconda3/bin/conda' 'shell.bash' 'hook' 2> /dev/$

if [ $? -eq 0 ]; then

eval "$\_\_conda\_setup"

else

if [ -f "/Users/kadm/anaconda3/etc/profile.d/conda.sh" ]; then

. "/Users/kadm/anaconda3/etc/profile.d/conda.sh"

else

export PATH="/Users/kadm/anaconda3/bin:$PATH"

fi

fi

unset \_\_conda\_setup

# <<< conda initialize <<<

Running SEAsnake notes

* May need to do sudo
* Don’t use nohup so can see process