

# Zanvyl Krieger School of Arts and Sciences Advanced Academic Programs

410.734.81 Practical Introduction to Metagenomics

Topic: Introduction to Unix/Linux

Instructor: Joshua Orvis

Some of you are taking this course because you want to be on the computational side of bioinformatics and, if so, Unix/Linux is the *de facto* primary operating system for the field.

If you see your career following more the role of the biological analyst and not a programmer, you'll most likely still need to learn this. Just as you expect to work with programmers who have some idea about the biology of a project, they'll expect you to have some clue about their side of the work.

Throughout my career it has been my experience that the best analysts are those who can do at least rudimentary shell/Perl/Python coding and it's becoming more common that analysts positions require it.

For this specific class, all work will be performed on the DIAG compute cluster and to keep up you'll need to know these basics.

If you're attending this course it is presumed you have already taken 410.634 or have equivalent experience.

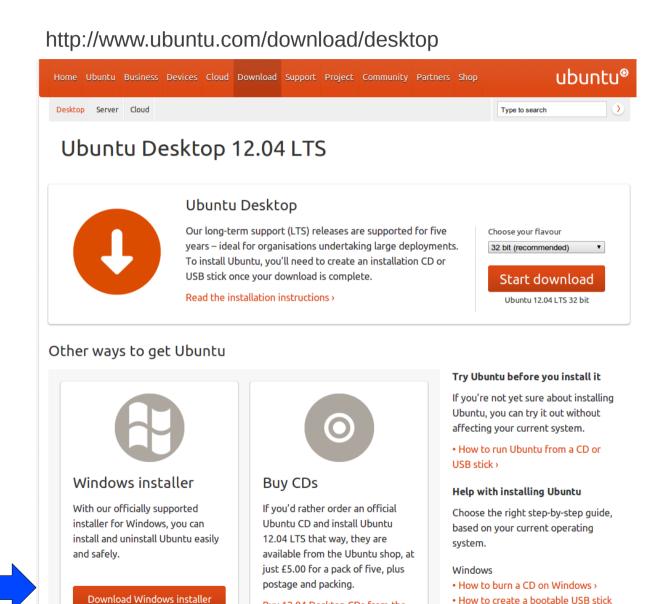
While there are many tools available on all platforms, bioinformatics is primarily a UNIX-based field, so it is important for your careers that you become an expert in using at least one UNIX-based operating system.

Aside from code editing, nearly all of this class will consist of performing operations using the Unix/Linux command line, so you **must** get comfortable using this.

If you are not already using Linux, Mac OS X or another UNIX-based operating system it is assumed that you will follow the "Running Ubuntu" guide on the next slide to either install Linux on your computer or boot from a LiveCD for a temporary working environment.

If you're using Windows and want to try Linux you can do so and keep your Windows installation at the same time.

If you use the "Download Windows Installer" option near the bottom-left, it will install Windows and Linux apart from each other, and you'll have a menu to choose which one you want to boot into each time you turn your computer on.



Buy 12.04 Desktop CDs from the

shop >

Read the installation instructions >

on Windows >

installer >

• Installing Ubuntu with the Windows

"Unix is simple. It just takes a genius to understand its simplicity." – Dennis Ritchie

Historically, the term "UNIX" evokes images of green/black monochrome screens with a blinking cursor, ready to accept commands from a bearded fat man on a clicky tan keyboard.



Modern UNIX-based operating systems, such as Ubuntu Linux, have very appealing, highly-customizable interfaces that can be used in a familiar point-and-click way but still provide terminal/shell access that is far more powerful for some applications.





#### What is UNIX?



"UNIX was not designed to stop its users from doing stupid things, as that would also stop them from doing clever things." – Dennis Ritchie

In modern usage, when we say 'UNIX' we are referring to a wide family of operating systems that include BSD, OS X, and numerous distributions Linux.

Specifically, they are mostly all portable, multi-user operating systems with hierarchical directory structures, text-based configuration files, access to command-line terminals and descendent from "Unics", developed by AT&T Bell Labs in the 1970s.

In this class we'll focus on Linux, and specifically our examples will use a *distribution* called Ubuntu Linux. There are hundreds of competing distributions, but this is one of the most popular and is easy to get started with. While I might often say just "Linux" during these lecture slides, you should consider most of this to be true with any UNIX variant.

## **Open-source software**



The Linux *kernel* (the core part of the operating system) is open-source which, at its purest, just means that you have the ability to download and view the source code for it. In practice this is also true for nearly all of the other programs within a given Linux distribution, but not always. Often this is also referred to as free software, but "free" is more difficult to define than you might suspect:





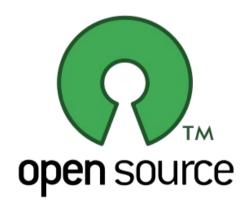


Free as in Beer – First coined by software activist Richard Stallman, meaning that something doesn't cost any money. (I don't know what bars he goes to.)

Free as in Speech – This sort of software is free in the sense of having no restrictions. You can download it, change it, redistribute it, etc.

Free as in Pretzels – Free to acquire, and it may whet the appetite, but you have to pay to get additional, often ultimately necessary, functionality. iPhone users should be familiar with this.

Corporate lawyers for companies who use (and write) open-source software do not run around courtrooms talking about pretzels and beer. There are a host of specific open-source licenses to choose from when you want to make your project open-source, saving you the hassle of legal document preparation. The Open-Source Initiative (OSI) provides these along with guides for applying them to your project.



www.opensource.org

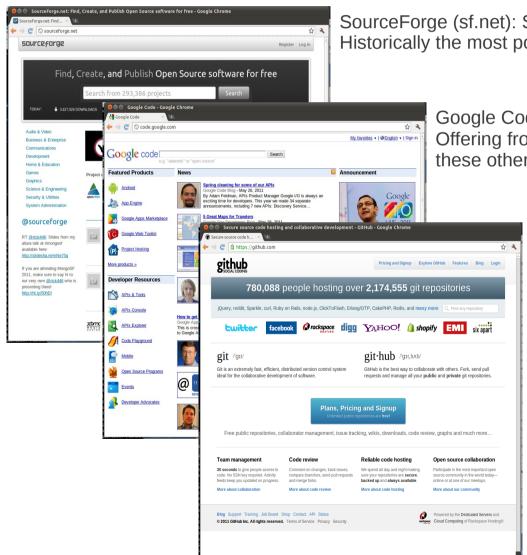
Choose your license carefully, as they all specify different combinations of rules governing whether users can do things like redistribute, modify or sell your code. Popular examples include:

GNU General Public License – popular but "viral" license that requires any programs that link or use GPL code to also be released under the same license.

Artistic License – Written by Larry Wall, the author of Perl, this license has been successfully tested in court.

BSD License – Relatively open and permissive set of licenses that are approved by the Free Software Foundation.

There are several websites dedicated to hosting your code and providing it to others. Each of these examples provides free source code management access as well as project organization tools, bug trackers, mailing lists, etc. for your user community.



SourceForge (sf.net): SVN and CVS code management. Historically the most popular, though losing ground to newer resources.

Google Code (code.google.com): SVN or Mercurial repositories. Offering from Google – A distant 3<sup>rd</sup> place when compared with these other two, but is well integrated into Google's other services.

GitHub (www.github.com): Git repositories only. The fastest rising and currently most popular of these, GitHub uses a distributed code management system called 'Git' that encourages software forks and merges. This is the home of projects like Rails and JQuery.

The terminal is your primary method for accessing your operating system and telling it what to do. Embrace it. Love it. Learn it. As bioinformatics developers you will learn that it is an indispensable tool for getting things done quickly.

To open a terminal in Ubuntu hit "CTRL + Alt + t" or click Applications → Accessories → Terminal

On a Mac open the Finder and search for Terminal.

This is a called a terminal *prompt*. It usually has this format:

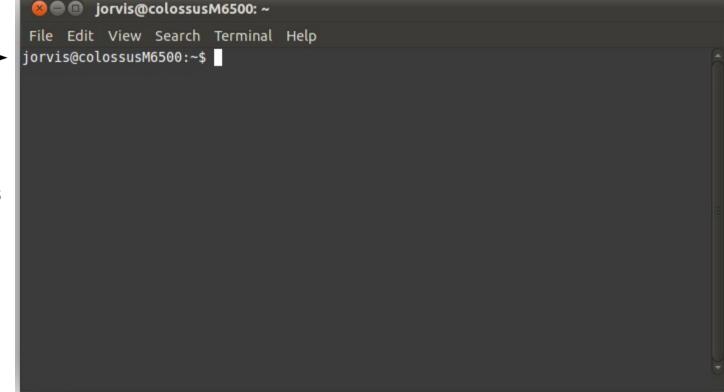
user@host:path\$

user: who you are logged in as

host: computer's name

path: position in the filesystem\$: symbol of the shell in use

(Why is my host called colossusM6500? Because it's a 17" behemoth of a laptop and is nearly unusable on planes. The M6500 is just the model #)



When using a terminal you are issuing commands within a *shell* and, naturally, there are a variety of different shells to choose from. Each has slightly different syntax when you really get into the details of them, but the general commands we'll use in class will be the same.

The default shell on Ubuntu and recent versions of OS X is *bash*, which stands for <u>Bourneagain shell</u>. Here are some example commands so you can get a feel for the syntax:

\$ pwd (command only)

\$ Is -I (command with an option)

\$ head -n 5 class.notes (command with an option with an argument (5) and file)

\$ ssh -l jsmith -XY someserver.com (command with multiple options)

We'll cover these commands in detail on upcoming slides, so just focus on the elements of each command here. (Also remember that the '\$' symbol just shows the start of a command for our bash shell, and you don't type it.)

In each of these examples the *command* is first. The first ('pwd') doesn't have any *arguments* or *options* passed to it. The second has a '-l' option alone. The next has an option '-n' that requires an argument (5) and a file to operate on. The ssh command has multiple options and arguments.

Let's learn how to log in to a server next and then see what these and other commands actually do.

The **ssh** command allows you to remotely log into other servers, such as the one set up for this class. You must pass it a server name or IP address. (You'll need to replace the IP address here with the one you were sent for this class.)

\$ ssh -l jsmith 192.168.1.200

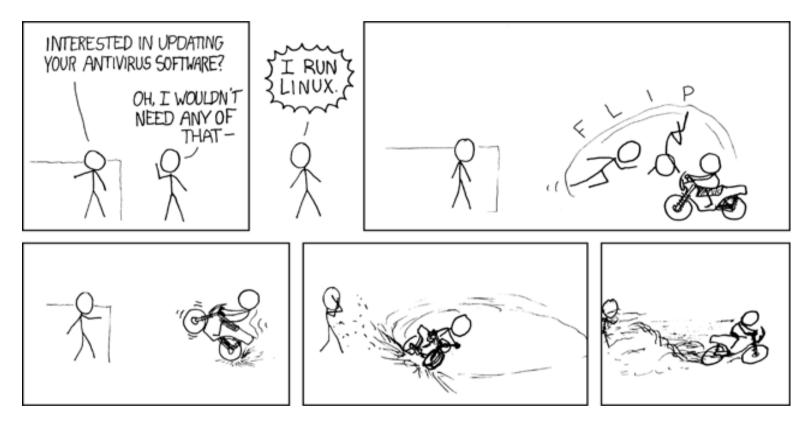
You will be prompted for a password (again, use the one you were given) and, once successful, will find a shell waiting to accept your commands. Some initial commands to try from here are:

\$ whoami (returns the name of your logged-in user)
\$ hostname (gives the name of your server)
\$ pwd (prints your working directory)
\$ ls -l (lists the files in your working directory, with details)

This set of simple commands can help orient you when you're on a new server. The next thing you should do if this is your first time is change your password with *passwd*:

\$ passwd

You will be prompted to enter your current password before entering a new one. You can change this as often as you like. When you are finished with a terminal type 'exit'.



xkcd.com/272/

(There is no point to this at all except to break up slides with lots of text. If you are anti-humor, carry on to the next slide.)

Most of you are already familiar with the hierarchical way in which files and directories are stored in most operating systems. (Note that 'directories' and 'folders' are generally interchangeable terms.) You can create files and folders, and place them all inside any other folders, creating a tree-like structure. In UNIX file systems the beginning of this tree is called the *root* and has a very simple path:

/

Most UNIX file systems have several directories here. I'll highlight some important Ubuntu ones for this class that will show the multi-level structure of the hierarchy:

/home/jsmith/
/opt/
/tmp/
/tmp/
/var/lib/mysql/
/var/log/apache2/
/var/www/

(each user has a home area)
(where custom-installed software will go)
(anyone can write temporary files here)
(MySQL data file locations)
(apache writes error and access logs here)
(web application files will go here)

Windows users will notice that there are no drive letters (C:, D:, etc.) In UNIX there is a single directory hierarchy and other drives can be *mounted* to show up as a directory in almost any location.

When moving around the file system (command: *cd*) you need to know where you are (*pwd*) and see what's there (*l*s). Here are some examples:

In this series of commands we check our location, see what's there, change directories into the 'docs' folder, show our path, cd back 'up' one directory (using '..'), then show our position again.

# STAND BACK



When you log in you'll be placed in your own area called your *home directory*.

\$ pwd
/home/jsmith

Your home area is your own personal work space to create, upload and organize files and directories in any way you like. The quickest way to create a file is with the *touch* command:

\$ touch test.file

This creates a file called 'test.file' that is initially empty. We'll learn how to populate it soon. You can create a directory like this:

\$ mkdir notes

This creates a directory called 'notes' that you could place other files in. The 'rm' command is used to remove files and directories, though it requires special options when removing a directory as slight protection from accidentally removing more than you intended to. You can even *pass* multiple arguments to rm if you want to delete multiple things:

\$ rm -rf test.file notes

The '-rf' options tell rm to remove the 'notes' directory recursively and forcefully, meaning that it will also remove the contents of 'notes' without validation prompts. Notice that there are no warnings when deleting files you own, so take care when using 'rm'.

Two simple commands are used to copy (cp) or move (mv) files on the UNIX file system. The mv command is also used to rename files. Both commands have the same basic syntax:

\$ command source destination

For example, to make a backup copy of a file you might do this:

\$ cp journal.doc journal\_backup.doc

If this journal file was instead no longer current and you wanted to rename it, you could do:

\$ mv journal.doc journal.doc.old

If you want to copy a directory instead of a single file you have to pass an option to the cp command to tell it to copy *recursively*. This would copy my docs directory into the temp area:

\$ cp -r /home/jorvis/docs/ /tmp/

Be warned that both commands will overwrite any existing files with the destination you specify without warning first. This makes it very easy to lose data if you aren't careful!

You can pass an option to the *Is* command to get a 'long listing' of files and directories that provides a lot more information, including the permissions allowed for each.

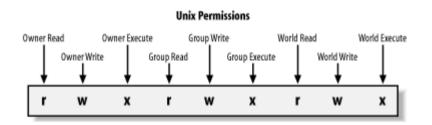
### \$ Is -I

```
drwxr-xr-x 2 jorvis adm 4096 2011-05-30 20:56 bin
drwxr-xr-- 2 jorvis adm 4096 2011-05-30 20:56 docs
drwxr-xr-x 2 jorvis adm 4096 2011-05-30 20:56 lib
drwxr-xr-x 2 jorvis adm 4096 2011-05-30 20:56 music
drwxr-xr-x 2 jorvis adm 4096 2011-05-30 20:56 notes
-rw-r--r-- 1 jorvis adm 62656 2011-05-30 21:45 resume.pdf
drwxrwx--- 2 jorvis adm 4096 2011-05-30 21:45 share
```

There is a lot of useful information here. The columns returned are:

- 1. Object permission block (see next slide for detailed info here)
- 2. Number of links to the object (not commonly used)
- 3. Object user owner
- 4. Object group owner
- 5. Object size
- 6. Last modified date
- 7. Last modified time
- 8. Object name

Let's return to the first column in the 'ls -l' output on the previous slide. It is a concise shorthand – the first character tells you what type of object it is (file, directory, symlink, etc.) After that are three triplet characters that describe who should be able to read, write and execute that object. For directories, 'execute' specifies whether the user can cd into it. See the graphic below:



Looking back on our previous slide, we see that our 'docs' directory has 'drwxr-x---'. This means the user can do anything to the file, group members can only read or execute, and the rest of the world can't do anything at all.

This shorthand can (and most commonly is) shortened even more using octal notation. For each triplet r=4, w=2, and x=1. These values are summed within each triplet then concatenated with the other two. For our 'docs' directory the triplets are:

User 
$$rwx = 4+2+1 = 7$$
  
Group  $r-x = 4+0+1 = 5 = 750$   
World  $--- = 0+0+0 = 0$ 

Using this notation you can set all permissions on any object with a single chmod command:

\$ chmod 755 docs

(opens permissions to give world readability)

(AN UNMATCHED LEFT PARENTHESIS CREATES AN UNRESOLVED TENSION THAT WILL STAY WITH YOU ALL DAY. You will often need to transfer files between computers, either downloading data from a server or uploading code/data from your workstation to a server. UNIX provides many ways to do this, but two simple ones you can use throughout this class are:

wget – If you have the URL for a file you would like to download, such as a large DNA database, this is an appropriate command-line choice. It has options to do much more, such as full site mirroring, but here is an example of downloading a single file (please don't do this on the class server).

\$ wget ftp://ftp.ncbi.nih.gov/blast/db/FASTA/nr.gz (get the non-redundant protein db)

**scp** – The <u>secure copy</u> tool provides a way to transfer files between computers using syntax similar to native UNIX 'cp' commands. The main difference is that the source or destination can contain the user and host information of a remote server. For example, to transfer a file from your home directory locally to your home directory on a remote server you can do this:

\$ scp resume.pdf jsmith@192.168.1.200:/home/jsmith/docs/

This is the best way to transfer files you work on to the class server, so you should practice this.

There are several ways to read the contents of a file using the terminal and others that invoke a graphical code editor, which are discussed on the next slide. Using the terminal, the most common way to inspect the contents of a file is to use *less*:

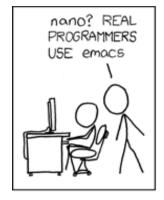
\$ less dna.fna

The less program reads the size of your terminal and displays one 'page' at a time. You can use the space bar to scroll down or your keyboard arrows to go either direction. If you want to search the file you can type '/', the string you want to find, then enter to search. Finally, to quit just press 'q'.

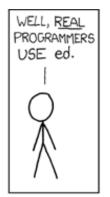
If you know you only want to see the first or last number of lines in a file, the *head* and *tail* commands are useful.

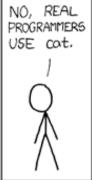
\$ head -n 10 dna.fna (shows the first 10 lines of the dna.fna file) \$ tail -n 20 dna.fna (shows the last 20 lines of the dna.fna file)

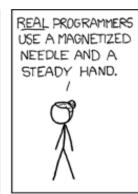
Next we'll cover more advanced editors to view and modify files.





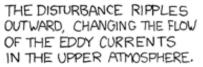








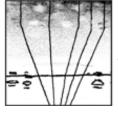


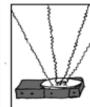


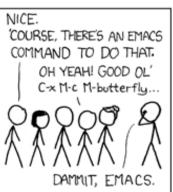


THESE CAUSE MOMENTARY POCKETS OF HIGHER-PRESSURE AIR TO FORM,

WHICH ACT AS LENSES THAT DEFLECT INCOMING COSMIC RAYS, FOCUSING THEM TO STRIKE THE DRIVE PLATTER AND FLIP THE DESIRED BIT.







I am not going to join the incessant, almost religious, debates about the superiority of one code editor over another. It is much more important that you choose one and spend a lot of time learning all of its features than choose the 'right' one. Some editors run within your terminal window (like vi, and older emacs) while others open a new window within your graphical environment. How do you know where to start? I'll give you my (mostly) unbiased short summaries of some of the most popular, but you should try them all out yourself.

<u>eclipse</u> – This is a full-fledged, professional-level graphical IDE. It is the *de facto* standard for Java developers and those in structured, team-driven development environments.

<u>emacs or xemacs</u> – This uses keystrokes for operations you might normally use a mouse for. It has a very steep learning curve but those developers I've seen master it seem almost godlike in their ability to fling code around.

<u>nedit</u> – A simple, graphical editor with a very low learning curve and surprising amount of features including languages modes, syntax highlighting and keyboard macros.

 $\underline{vi}$  – This is similar to emacs in that it uses keystrokes to perform most operations rather than a mouse, but works only within your terminal windows. This is the most common editor I use when I want to quickly modify a single part of the file, save, and close it.

This is in no way an exhaustive list but includes enough of the most popular alternatives to get started.

Many commands that you run generate output, and UNIX allows you to redirect that output to a file or other programs instead of just displaying it on the screen. There are a few basic tenets of core UNIX programs that make this very useful:

Programs should do one thing, well.

They should be written to connect to other programs via input/output streams.

This allows you to both redirect the output of these tools either to the screen, files on disk or directly as input to another tool. Powerful operations can be performed on the command-line by streaming these tools together. For example, the following series of commands would count the number of entries in a directory full of FASTA files:

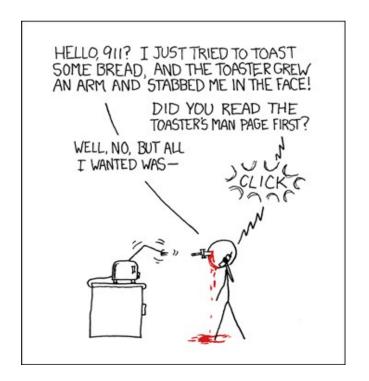
\$ cat \*.fna | grep ">" | wc -l

There is a lot of new material here. The *cat* command spews out the contents of the passed files, and in this case a wildcard '\*' character is used to say "anything followed by .fna". So this same command would work if there were 10 or 10,000 files in the directory. This output is then streamed to the *grep* command using the '|' (pipe) symbol. Grep is used to search for strings or patterns within an input file or stream, and will return only those lines of the input that match the search pattern. In this case, we use grep to filter the input to match the FASTA header lines. This output is then piped to *wc*, which is used to count words, lines or characters. The '-l' option to wc specifies that we want a count of the lines returned by grep.

We have only scratched the surface of the functionality of many of these commands. The best way to learn more, or get clarification, is to use some of the built-in resources and documentation. The most common way is to use the *man pages*. For any command, such as the scp command you just learned, you can pull up that command's <u>man</u>ual like this:

\$ man scp

This will provide often extensive documentation from the tool's author. This should always be your first resource when you need help with a command.



Other great resources (besides Google) when learning Linux and its tools include:

linuxquestions.org help.ubuntu.com/community/UsingTheTerminal

Also, IRC is very old but is still very active and one of the best ways to get immediate answers from dozens of other users. There are channels for thousands of topics using a live chat mechanism. Here's a useful getting started guide:

www.linux.com/archive/feed/61439

In this series of slides we have gone through many common UNIX commands and learned how to get information on new ones we encounter. You should use these resources to find information on these other commands we will use throughout the class.

```
sort
cut (filter sections of input)
find (searches for files/directories)
ps (view the processes on a computer)
kill (kill a running process)
tar (creates an archive of files/directories as a single file)
gzip (compresses a file)
```

If you are newer to the command line you might find it helpful to refer to a 'cheat sheet' that you keep printed nearby. There are many of these online, but here's an example:

http://fosswire.com/post/2007/08/unixlinux-command-cheat-sheet/

Unix commands tutorial and reference: www.unixtutorial.org/commands

Comparison of open-source licenses en.wikipedia.org/wiki/Comparison\_of\_free\_software\_licenses