In today's lab we will use the program FastQC to perform quality control check of our data and Trimmomatic to remove bad data such as adapters.

Start by sshing into your scientific computing account :

ssh np17d@pamd.sc.fsu.edu

Type:

classroom

First copy the lab folder to your account. You must replace np17d with you account login:

cp -R /usr/common/Bioinformatics/Bioinformatics2020/Sep_11_2020/Lab4Student/ /home/np17d/Documents/

Navigate into the FastQC Folder :

cd /home/np17dDocuments/Lab4Student/FastQC

Give the executable fastqc permission to run :

chmod 755 fastqc

Run fastqc on both data files:
./fastqc ../Data/I31820_S129_L001_R1_001.fastq.gz

./fastqc ../Data/I31820_S129_L001_R2_001.fastq.gz

Navigate to the output files:
cd ../Data

See the files:
ls

Get the Directory:
pwd

Copy the html files to your local machine to view them. You must change the paths accordingly. The first path is the location on the pamd server, and the second path is where you want it on your local machine.
/home/newking9088/Downloads/ needs to be replaced by where you want the file on your local computer.
Open a new terminal
scp np17d@pamd.sc.fsu.edu:/home/np17d/Documents/Lab4Student/Data/I31820_S129_L001_R1_001_fastqc.html /home/newking9088/Downloads/

**np17d@pamd.sc.fsu.edu:/home/np17d/Documents/Lab4Student/Data/
I31820_S129_L001_R2_001_fastqc.html /home/newking9088/Downloads/**

Double click the files to view them :

What does the report warn about?

Lets us Trimmomatic to remove the adapters.

Return to the Terminal with the ssh connection:

Nagivate to the Trimmomatic Folder

cd ../Trimmomatic-0.39

Run the program
java -jar trimmomatic-0.39.jar PE ../Data/I31820_S129_L001_R1_001.fastq.gz

**../Data/I31820_S129_L001_R2_001.fastq.gz lane1_forward_paired.fq.gz
lane1_forward_unpaired.fq.gz lane1_reverse_paired.fq.gz lane1_reverse_unpaired.fq.gz
ILLUMINACLIP:adapters/TruSeq3-PE.fa:2:30:10 LEADING:3 TRAILING:3
SLIDINGWINDOW:4:15 MINLEN:36**

The commands we used did the following:
ILLUMINACLIP: Cut adapter and other illumina-specific sequences from the read.
LEADING: Cut bases off the start of a read, if below a threshold quality
TRAILING: Cut bases off the end of a read, if below a threshold quality
SLIDINGWINDOW: Performs a sliding window trimming approach. It starts scanning at the 5‟ end and clips the read once the average quality within the window falls below a threshold.
MINLEN: Drop the read if it is below a specified length
Run FastQC on one of the output files

Nagivate to the FastQC Folder
cd ../FastQC

Run
./fastqc ../Trimmomatic-0.39/lane1_forward_paired.fq.gz

Copy the file to your local machine to view it. Again you must replace the paths with what you are using.
Go to your other terminal:

**scp np17d@pamd.sc.fsu.edu:/home/np17d/Documents/Lab4Student/Trimmomatic-0.39/
lane1_forward_paired_fastqc.html /newking9088/kevin/Downloads/**

Submit all three FastQC html files!