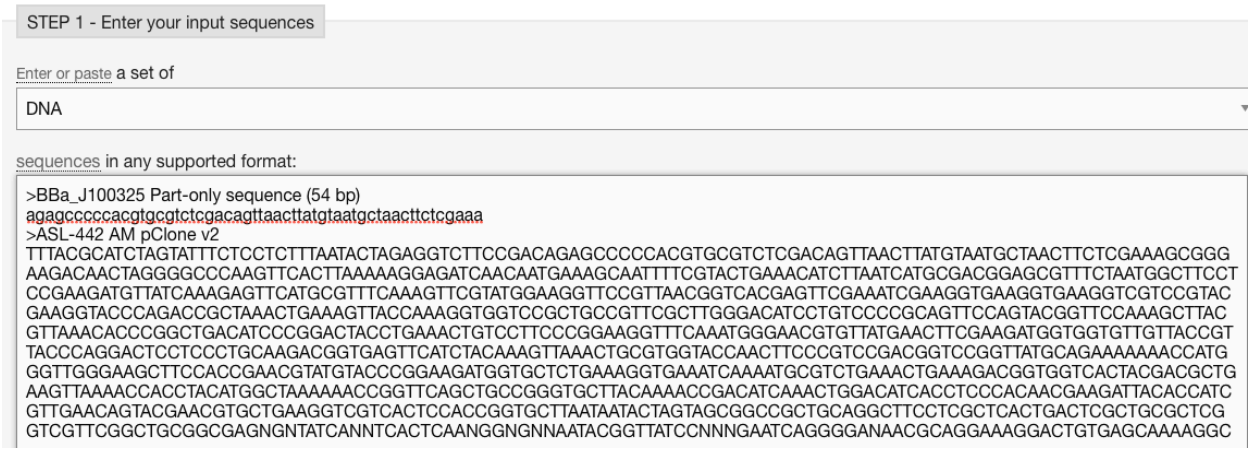
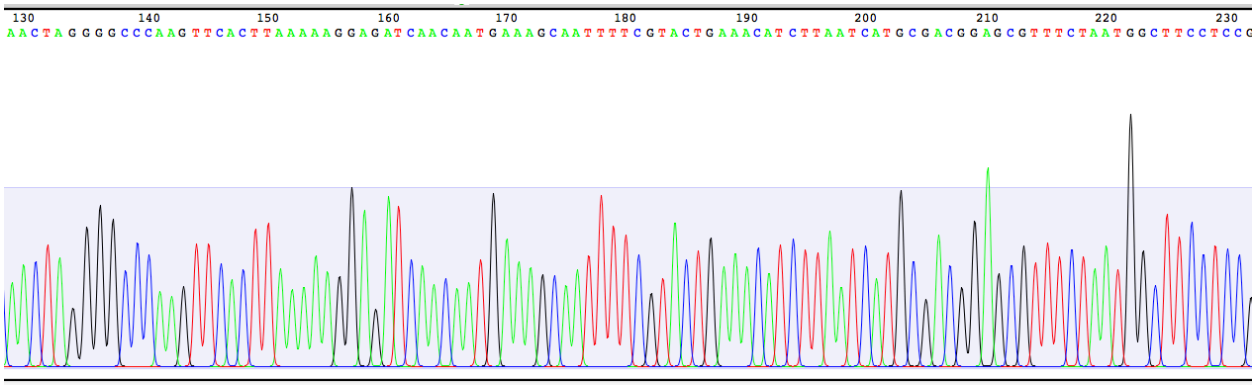
Verifying Bases of Interest: xClone Red

1. Right click on one of your file (.seq and .ab1). On a Mac, you can Get Info and have it do this every time as demonstrated in lab.
2. Go to CLUSTAL Omega (https://www.ebi.ac.uk/Tools/msa/clustalo/) web tool. Change the data type to DNA.
3. Copy and paste your designed sequence using FASTA format (see below). You will need to remember the part numbers for v1 and v2 designs. Do the same with your sequencing results. Open a .seq file to get the sequence. Omit the initial Ns and terminal Ns. Use FASTA format to create a name as shown below. Submit using "CLUSTAL w/ numbers".



4. If there are bases in your region of interest, then you can open the chromat (.ab1) using ApE. You can verify any bases about which the sequencing software was uncertain (see below).



5. Record the results for each of your 4 sequencing reactions. If your sequencing reaction did not produce usable data, look at the chromat and confirm that the sequence cannot be read.