

Comparison of Microbial DNA Sequences

Bacteria Genus name	NCBI Accession Number
<i>Chlorobium</i>	AJ299414
<i>Phaeospirillum</i>	D14433
<i>Chloroflexus</i>	AJ309653
<i>Halobacterium</i>	L37421
<i>Methanococcus</i>	AF051404
<i>Nitrosomonas</i>	AJ298739
<i>Beggiatoa</i>	L40997
<i>Delsulfovibrio</i>	U32578
<i>Bacillus</i>	AB050630

Directions

1. Go to the NCBI database at www.ncbi.nlm.nih.gov
2. For the input box that says “All Databases” click the dropdown arrow and select “nucleotide”
3. Enter the accession number for the first bacterium in the search box and click “search”
4. Your search will return only one result since accession numbers are unique identifiers. Under the “send” drop down menu on the right side on the screen, select “clipboard” and then “add to clipboard”. This will store the sequence on the clipboard temporarily.
5. Repeat steps 3 and 4 for the remaining bacteria ascension numbers.
6. When you have added all 9 sequences, click on the “9 items” beside the clipboard icon on the right side of the screen.
7. Under the “Display Settings” dropdown menu on the left side of the screen select “FASTA (text)”. Then, click “Apply”.
8. This will open a new browser window. This window will display all the sequences in the FASTA format. You’ll use these sequences in the copy and paste step below (10).
9. Go to the CLUSTALW website www.ebi.ac.uk/Tools/msa/clustalo/
- 10 The screen will read ClustalW2 Multiple Sequence Alignment. Copy and paste all the sequences into the input area in Step 1 box.
11. Click the red run “Submit” button at the bottom of the screen to go to the results page. The results are shown as all pairwise alignments and multiple alignments.
12. Click on “Guide Tree” tab to see the cladogram/phylogenetic tree at the bottom.

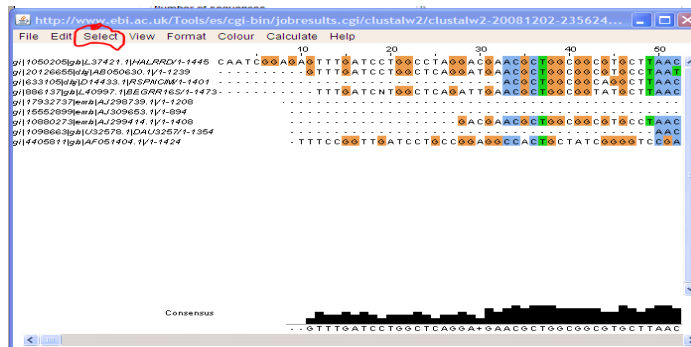
Analysis questions

1. Draw out the resulting **PHYLOGENETIC TREE** for all the bacteria including relative lengths of each line. (18pts)
2. All of the sequences above are taken from the 16s rRNA gene of each bacteria. Suppose you worked with a pharmaceutical company to design an antibiotic that disables 16s rRNA. Costly clinical trials have shown your antibiotic is very successfully in *Beggiatoa* but not in *Methanococcus*. **Explain why this is the case.** (10pts)
3. You are given just enough money to extend the clinical trials and test the effectiveness of your antibiotic on two more bacteria. **Which two would you choose for the trials and why?** (10pts)
4. Further research has found that your antibiotic specifically binds and interrupts the following sequence

ACAGGTGCTGCATGGCTGTCGTCAGCTCGTGTGAGATGTTGGGTTAAGTCCC GCAACGAGCGCAACCC

From the screen with your phylogenetic tree, click on the “Result Summary” tab. Click on “Start Jalview” to open an editable alignment view.

From here, click the “Select” menu and then click “Find”. In the find box paste the sequence from above to see where it appears in the bacteria. Matching sequences will be highlighted BLACK.



Based on this new information, what other bacteria are likely to be affected by your antibiotic? What evidence did you find to support your response? (4pts)

5. **Research the following about each genus of bacteria:** nutrition, habitat, ecological importance/impact, and/or other interesting facts. **Add a few sentences about each genus in the order that it occurs in the phylogenetic tree.** This can be done in popplet, other concept map builders on the ipad or on the internet, or hand written on your paper. (27pts)