

10.3 UNDERSTANDING OUTPUT

The ClustalW output will give you two main forms results – the **multiple sequence alignment** and a **phylogram/cladogram**.

The score table is the first section of the page below, the results summary box. The score table shows the scoring of the pairwise alignment of all sequences (Fig. 10.5).

SeqA Name	Len (aa)	SeqB Name	Len (aa)	Score
1 1-Halomicrobium	243	2 2-Halomicrobium	231	31
1 1-Halomicrobium	243	3 3-Halomicrobium	237	16
1 1-Halomicrobium	243	4 4-Halomicrobium	273	18
1 1-Halomicrobium	243	5 1-Halobacterium	239	40
1 1-Halomicrobium	243	6 2-Halobacterium	236	56
1 1-Halomicrobium	243	7 3-Halobacterium	237	35

Fig. 10.5: Showing scores table.

Take a screen shot of this table, or download by right-clicking the Output File (.output) found in the result summary box at the top of the page (Fig. 10.6).

Results of search	
Number of sequences	8
Alignment score	6793
Sequence format	Pearson
Sequence type	aa
JalView	Start Jalview
Output file	clustalw2-20091008-0844088435.output
Alignment file	clustalw2-20091008-0844088435.ali
Guide tree file	clustalw2-20091008-0844088435.dnd
Your input file	clustalw2-20091008-0844088435.input
SUBMIT ANOTHER JOB	

Fig. 10.6: Showing how to save the output file.

CLUSTAL omega aligns all of the input sequences, an HTML text version is listed just below the Scores Table. A more extensive view of the alignment can be seen using JalView. Under alignment, you can click “Show Colors” to view a coloured version of an amino acid alignment (Fig. 10.7).



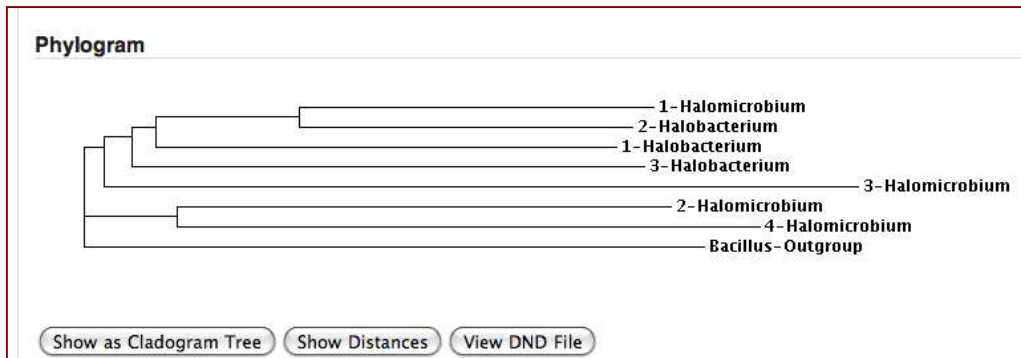
Fig. 10.7: Showing “normal” and “coloured” alignment results.

In the row below the last sequence of the alignment, there may be symbols like:

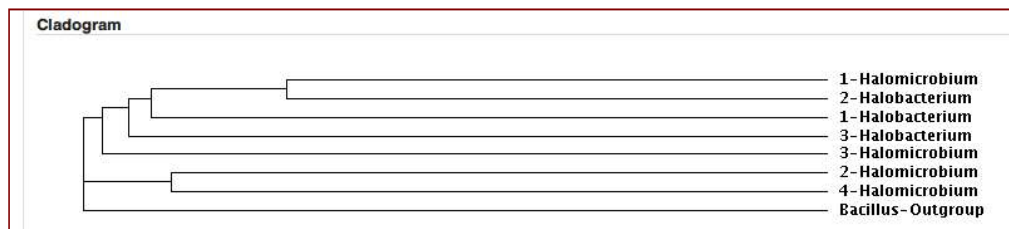
- " * " – the residues or nucleotides in that column are identical in all sequences
- " : " – conserved substitutions have been observed, according to the colour data
- " . " – semi-conserved substitutions are observed

The generated phylogenetic tree is at the very bottom of the results page. You'll notice above this there is a “Guide Tree” section. You can save the **Guide Tree**. The tree can be viewed as a **phylogram** or a **cladogram**.

A **phylogram** explicitly represents the number of sequence character changes through the horizontal branch length. The sum of the horizontal distances between two leaves is the predicted evolutionary difference in sequences. A **cladogram** only depicts branching patterns, not evolutionary time by branch length (Fig. 10.8 A and B).



A)



B)

Fig. 10.8: A) Phylogram B) Cladogram.

10.4 SUMMARY

- Multiple Sequence alignment is aligning of three or more biological sequences of similar length.
- From the output of MSA applications, homology can be inferred and the evolutionary relationship between the sequences studied.
- In the current exercise you have learnt to use the multiple sequence alignment tool Clustal Omega for analysing evolutionary relationships among sequences and interpret relationships among the sequences or organisms through a phylogenetic tree.

10.5 LAB EXERCISES

1. Retrieve any three or more protein sequences from protein database, copy the sequences in FASTA file format, align the sequences each other and report the pair wise score using Clustal Omega.