Algorithms for multiple sequence alignment

Lecture 13

https://phylo.cs.mcgill.ca/game.html

The basis of modern biology

- Cell theory
- Mechanism
- [✓ Evolution]

Observation I

- Species have great fertility, but not all their offspring survive
- Populations (groups of species) remain approximately the same size
- The resources (food, space, mates) are limited

Inference: there should be a struggle for survival

Observation II

- No 2 individuals are completely identical
- Much of this variation is inheritable

Inference: Those who survive pass their traits to the next generation

Mechanism of evolution: Darwin's Theory of Evolution

- Variation: There is variation in every population
- Competition: Organisms compete for limited resources
- Offspring: Organisms produce more offspring than can survive
- Genetics: Organisms pass genetic traits on to their offspring
- Natural Selection: Those organisms with the most beneficial traits are more likely to survive and reproduce.

All life: descent with modification

"*Probably all organic beings which have ever lived on this earth have descended from some one primordial life form. There is grandeur in this view of life that, whilst this planet has gone cycling on according to the fixed law of gravity, from so simple a beginning - endless forms most beautiful and most wonderful have been, and are being evolved*."

(**Charles Darwin**, The Origin of Species)

The fact of evolution

- It is a ***fact*** that:
  - The earth with liquid water, is more than 3.6 billion years old
  - Cellular life has been around for at least half of that period
  - Organized multicellular life is at least 800 million years old
  - Major life forms now on earth were not at all represented in the past. There were no birds or mammals 250 million years ago
  - Major life forms of the past are no longer living. There used to be dinosaurs and Pithecanthropus, and there are none now
  - All living forms come from previous living forms. Therefore, all present forms of life arose from ancestral forms that were different. Birds arose from nonbirds and humans from nonhumans
- No person who pretends to any understanding of the natural world can deny these facts any more than they can deny that the earth is round, rotates on its axis, and revolves around the sun

Molecular evolution - variation

- On the molecular level: the variation is achieved by random changes in the DNA:
  - Sequence mutations
  - Genome rearrangements
  - Combinatorics of sexual reproduction
  - Horizontal transfer of transposons
  - Gene duplications

Molecular evolution - selection

- The selection is applied only to the molecules with observable function: phenotype – proteins
- Evolutionary molecular “inventions” proven to be useful are preserved:
  - 40% of Human proteins are in Yeast: two species evolved independently, but this successful set of proteins changed minimally
  - Insulin of human and cow is so similar that the cow insulin was used for diabetic patients

Proteins seem to be a collection of distinct “approved” *domains* (amino acid chains which form a particular shape), which are preserved by selection

Comparing multiple strings. Motivation

- More than technical exercise - critical cutting-edge tool for extracting important faint commonalities from a set of strings

Arthur Lesk:“*One or two homologous sequences whisper. A full multiple alignment shouts out loud*.”

- We can reveal critical conserved motifs, common 2-3 dimensional structures, the clue to a common biological functions (HIV drug)

Multiple string comparison solves a different biological problem

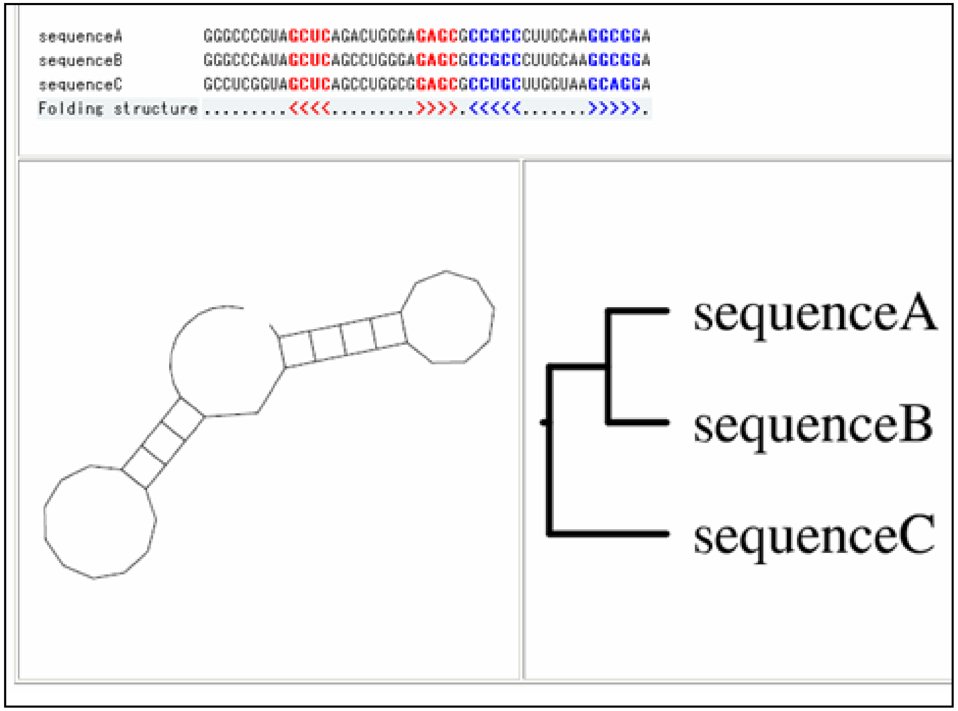
- When we are looking for sequence similar to a given sequence, performing the **pairwise** alignment, we try to find a **new biological relationship** based on the fact that the **two sequences are similar**
- When we are doing **multiple** alignment, the input **sequences may not be similar**, but they are known to have a **similar biological function** or shape, so we are looking for the similar regions to deduce what is responsible for their common biological function

Multiple Strings Comparison: Computational problems

- The mutation rate between organisms is high.
- Up to some extent the changes in DNA do not impact the functionality of the molecule, so all these similar regions we want to find are *inexact* matches

Sample application: Structure prediction

- For proteins with the similar shape or function, compute a multiple alignment and find what regions are conserved between all of them.
- These regions must play important role in defining their common 3D structure (function)



```
sequenceA         GGGCCCGUAGCUCAGACUGGGAGAGCGCCGCCCUUGCAAGGCGGA
sequenceB         GGGCCCAUAGCUCAGCCUGGGAGAGCGCCGCCCUUGCAAGGCGGA
sequenceC         GCCUCGGUAGCUCAGCCUGGCGGAGCGCCUGCUUGGUAAGCAGGA
Folding structure .........<<<<.........>>>>.<<<<<.......>>>>>.
```

Sample application: inferring evolutionary relationships

- Inferring evolutionary relations between species

S1	A	-	X	-	Z
S2	A	-	X	-	Z
S3	A	-	X	X	Z
S4	A	-	Y	-	Z
S5	A	Y	X	X	Z

Global Multiple Sequence Alignment (MSA)

- A global multiple alignment for k>2 strings is a table with k rows
- The spaces are inserted in chosen positions of any of the aligned strings, then each string is arrayed in a separate row such that each character and space is in a unique column

S1	A	-	X	-	Z
S2	A	-	X	-	Z
S3	A	-	X	X	Z
S4	A	-	Y	-	Z
S5	A	Y	X	X	Z

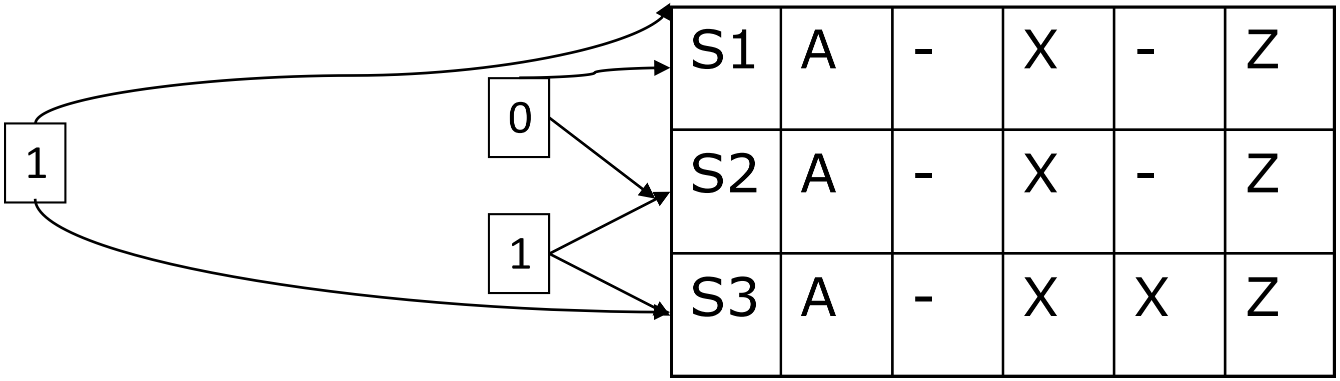
How to score MSA with an objective score function

- Sum of pairs
- Consensus
- Tree

- But better: to have an expert to look at the alignment (subjective score function)

The sum-of-pairs (SP) score

- The SP score is the sum of scores of pairwise global alignments for each pair of strings in the MSA
- Example: suppose the pairwise alignment scores are edit distances



S1	A	-	X	-	Z
S2	A	-	X	-	Z
S3	A	-	X	X	Z

Total SP-score (edit distance) is 2

The consensus score

S1	A	-	X	-	Z
S2	A	-	X	-	Z
S3	A	-	X	X	Z
S4	A	-	Y	-	Z
S5	A	Y	X	X	Z
S*	A	-	Y	-	Z
	0	1	4	2	0

Consensus string

Consensus score: 7

The consensus score of MSA is

$\mathrm{score}(MSA, S^*)=\Sigma_{\text{all columns } j}\ \Sigma_{1\leq i\leq k}\ \mathrm{score}(Si[j],S^*[j])$

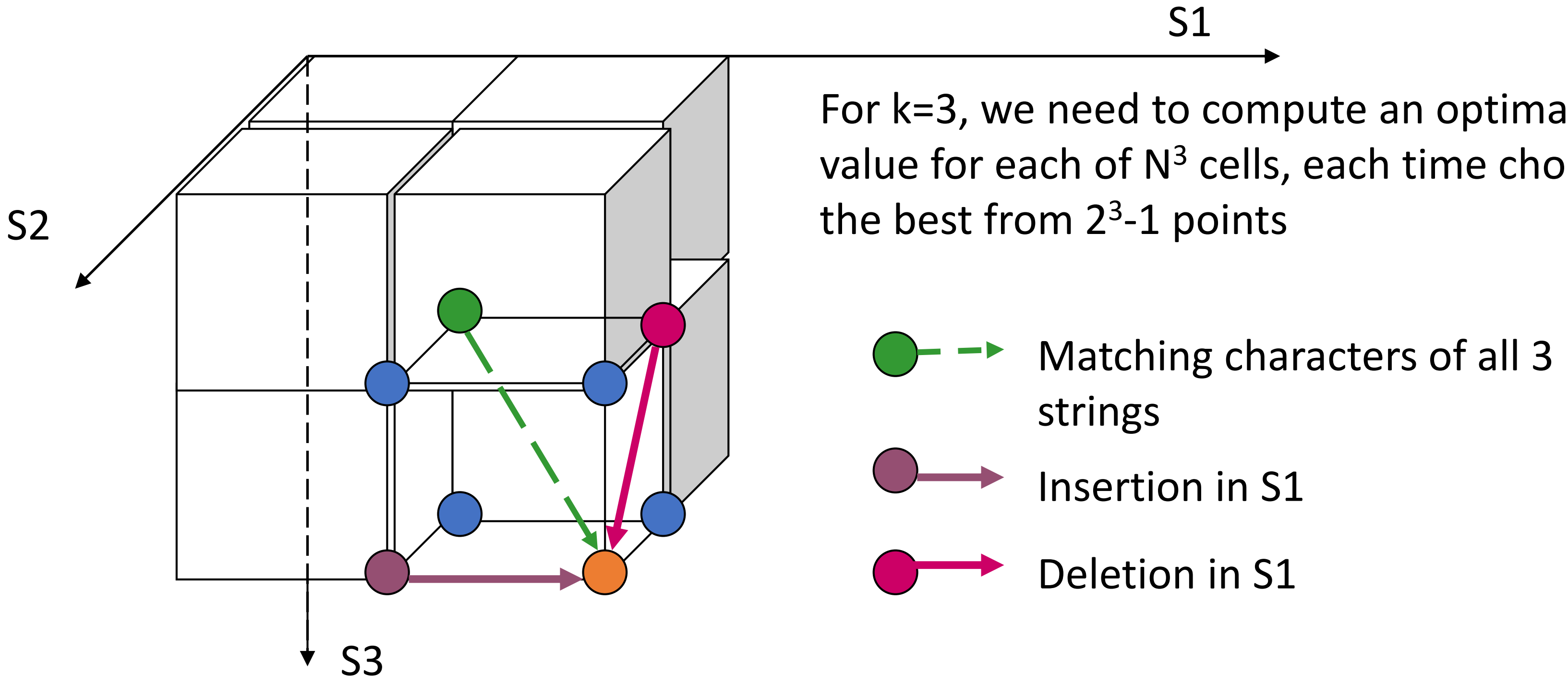
Multiple alignment problem

- Given a set S of k strings and an objective scoring function, compute multiple alignment with an optimal score
- There is no known efficient method for solving this problem for a *consensus score*, so we try to solve it for an *SP-score*

Dynamic programming solution for MSA with an SP-score objective function

- The solution is analogous to computing an optimal path in a multi-dimensional grid, exactly as for a pairwise alignment in a 2-dimensional grid.

For k=3, we need to compute an optimal value for each of N^3 cells, each time choosing the best from 2^3-1 points



● Matching characters of all 3 strings

● Insertion in S1

● Deletion in S1

The complexity of the DP solution

- $O(N^{k*}2^k)=O(N^k)$
- The problem is NP-complete

Heuristic: Iterative alignment

- We have 5 strings:

S1. AXZ

S2. AYZ

S3. AXXZ

S4. AYXXZ

S5. AXZ

- Let us try to add them to an alignment *iteratively*:

Iterative alignment – adding S2 to S1

S1. AXZ

S2. AYZ

S3. AXXZ

S4. AYXXZ

S5. AXZ

S1	A	X	Z
S2	A	Y	Z

Iterative alignment – adding S3 to M(S1,S2)

S1. AXZ

S2. AYZ

S3. AXXZ

S4. AYXXZ

S5. AXZ

S1	A	X	-	Z
S2	A	Y	-	Z
S3	A	X	X	Z

Iterative alignment – adding S4 to M(S1,S2,S3)

S1. AXZ

S2. AYZ

S3. AXXZ

S4. AYXXZ

S5. AXZ

S1	A	-	X	-	Z
S2	A	-	Y	-	Z
S3	A	-	X	X	Z
S4	A	Y	X	X	Z

or

S1	A	X	-	-	Z
S2	A	Y	-	-	Z
S3	A	X	-	X	Z
S4	A	Y	X	X	Z

Which is better? How many different possibilities are for longer strings?

Iterative alignment – result

S1. AXZ

S2. AYZ

S3. AXXZ

S4. AYXXZ

S5. AXZ

S1	A	X	-	-	Z
S2	A	Y	-	-	Z
S3	A	X	-	X	Z
S4	A	Y	X	X	Z
S5	A	-	X	-	Z

	S2	S3	S4	S5
S1	1	1	3	3
S2		2	2	3
S3			2	3
S4				2

SP score (M)=22

How good is it comparing to an optimal alignment?

How to choose the right order of sequences to insert?

An approximation algorithm for MSA with an SP-score objective function: SP-star

- Practical methods use *heuristics* to find sub-optimal SP alignment. Little is usually known about how much a produced alignment deviates from the optimal SP alignment.
- A bounded-error *approximation algorithm* is an algorithm which finds a sub-optimal solution, but which allows to exactly evaluate the difference between the computed solution and the optimal solution

SP-star algorithm for MSA

- For this algorithm, the scoring distance must have the following properties:

Property 1. D(S1, S1)=0 **identity**

Property 2. D(S1, S3) <= D(S1, S2) + D (S2, S3)

triangle inequality for strings

(the cost of transforming S1 into S3 is no more than transforming S1 into S2 and then transforming S2 into S3)

Property 3. D(S1, S2)= D(S2, S1) **symmetry**

Edit Distance has these properties

Edit Distance: formal definition

For each character x in S1 and y in S2:

d(x,z)= 0 if x=z
1 if x<>z

Definition 1. Distance $D(S1,S2)=\sum_{i=1}^{L}[d(S1_{[i]}, S2_{[j]})]$

Definition 2. Edit distance

ED(S1, S2)=min { D(S1, S2)}

A Center Star tree

- **Definitions**

Definition 1. Given a set S of k strings, define a center string Sc $\in$ S as a string that minimizes

$$\sum_{Sj \in S} \text{EDistance}(Sc, Sj)$$

$\forall i \quad \sum_{j=1}^{k}\text{EDistance}(Si, Sj) >= \sum_{j=1}^{k}\text{EDistance}(Sc, Sj)$

Definition 2. Center start tree - a tree of k nodes with Sc as a center and adjacent nodes – the remaining (k-1) strings of the set.

Produce an alignment Mstar by optimally aligning each string to a center string.

SP-tree algorithm – demo 1

S1. AXZ

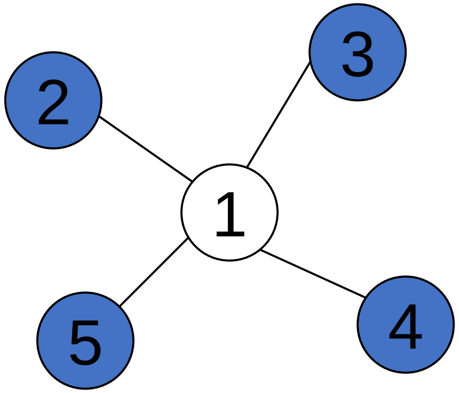
S2. AYZ

S3. AXXZ

S4. AYXXZ

S5. AXZ

	S1	S2	S3	S4	S5	
S1	0	1	1	2	0	4
S2	1	0	2	2	1	6
S3	1	2	0	1	1	5
S4	2	2	1	0	2	7
S5	0	1	1	2	0	4



We chose S1 to be a center string Sc

Performed in time $O(K^2N^2)$

SP-start algorithm - demo 2

S1. AXZ

S2. AYZ

S3. AXXZ

S4. AYXXZ

S5. AXZ

S1	A	-	X	-	Z
S2	A	-	Y	-	Z
S3	A	-	X	X	Z
S4	A	Y	X	X	Z
S5	A	-	X	-	Z

	S2	S3	S4	S5
S1	1	1	2	0
S2		2	3	1
S3			2	1
S4				2

- Align each sequence to Sc according to an edit distance between Sc and each other string

SP score (Mc)=15

Theorem 1.

SP score(Mc)/SP score (M*)<2

Proof

For simplicity, let's consider values in all cells of the pairwise distance table. They are directly proportional to SP-score

(1). SP score (Mc)= $\sum_{i=1}^{k} \sum_{j=1}^{k}$ ED(Si, Sj)

(2). ED(Si, Sj) <=ED(Si, Sc)+ED(Sc,Sj)
(triangle inequality)

(3). $\forall$i ED(Si, Sc)=ED (Sc, Si) (symmetry)

(4). From (1) & (2) =>
SP score (Mc)<= $\sum_{i=1}^{k} \sum_{j=1}^{k}$ [ED(Si, Sc)+ED(Sc,Sj)]=
= $\sum_{i=1}^{k} \sum_{j=1}^{k}$ ED(Si, Sc)+ $\sum_{i=1}^{k} \sum_{j=1}^{k}$ ED(Sc, Sj) =
=k $\sum_{j=1}^{k}$ ED(Si, Sc)+ k $\sum_{j=1}^{k}$ ED(Sc, Sj)}=
=2*k $\sum_{j=1}^{k}$ ED(Si, Sc)

	S1	S2	S3	S4	S5
S1	0	1	1	2	0
S2	1	0	2	3	1
S3	1	2	0	2	1
S4	2	3	2	0	2
S5	0	1	1	2	0

Distance table for central star algorithm: total score Mc

$$\boxed{\text{SPScore (Mc)<= } 2k \sum_{i=1}^{k} \text{ED(Si, Sc)} \quad \text{(I)}}$$

Theorem 1.

SP score(Mc)/SP score (M*)<2
Proof(cont.)

(5) SP score (M*)= $\sum_{i=1}^{k} \sum_{j=1}^{k}$ D*(Si, Sj)

(6) $\forall$ i $\sum_{j=1}^{k}$ D(Si, Sj)>= $\sum_{j=1}^{k}$ ED(Sc, Sj) (from the choice of Sc to minimize this sum)

(7). From (5) and (6) =>
SP score (M*)>= k* $\sum_{j=1}^{k}$ ED(Sc, Sj)

and

1/ SP score (M*) <= k* $\sum_{j=1}^{k}$ ED(Sc, Sj)

	S1	S2	S3	S4	S5
S1	0	1	1	2	0
S2	1	0	2	2	1
S3	1	2	0	2	1
S4	2	2	2	0	2
S5	0	1	1	2	0

This is total distance table for optimal (minimal) scores between each pair – the alignment is unknown. Let’s call this unknown optimal alignment M*

$$\boxed{1/\text{ SP score (M*)} <= \sum_{j=1}^{k} \text{ED(Sc, Sj)} \quad \text{(II)}}$$

Theorem 1.

SP score(Mc)/SP score (M*)<2
Proof (end)

(8). From (I) and (II) =>

SP score(Mc)/SP score (M*)<=2 ■

For simplicity, we proved an upper bound which is not tight.

It can be shown that the tighter upper bound is 2(k-1)/k = 2 – 2/k.

Thus, the upper bound for k=3 is 4/3=1.33, for k=4 the upper bound is 1.5 and for k=6 (a problem size considered to be too large for efficient DP solution with strings of length 200) the bound is still only 1.67

How we can use this approximation for a better exact solution

- An approximate solution for the SP alignment can be used in order to cut off the number of DP table cells to be computed
- If we estimated the total SP-score to be not more than D, we can consider only the cells in the tunnel of diameter not more than D around the main diagonal of the multi-dimensional DP table

MSA program. The Carrillo-Lipman algorithm

- The around-the-main diagonal idea is used in the MSA algorithm and its implementation
- It is able to optimally align (on a large server)
  - 20 Phospholipase A2 sequences (approximately 130 residues),
  - 14 Cytochrome C sequences (approximately 110 residues),
  - 6 Aspartal proteases (approximately 350 residues),
  - 8 Lipid binding proteins (approximately 480 residues) on our supercomputers.

All of these problems **approached the limits** of the problems that can be solved optimally by the MSA program, which can compute an optimal multiple alignment for not more than 7 strings of length approximately 200 each

- There is no practical scalable solution to this problem

The meaning of MSA scores in terms of relationships between sequences

- In the SP-score based alignment we try to minimize the total number of edit operations between each pair – but that does no mean that each sequence was transformed into each other sequence by a series of these edit operations

- In consensus-score based alignment we try to align all sequences to their common ancestor –consensus sequence. The problem is that we cannot find this consensus ancestor by efficient computation

Phylogenetic multiple alignment

- Optimizes distance between more closely related sequences, as follows from the phylogenetic tree for these sequences
- Given an evolutionary phylogenetic tree with a distinct string labeling each leaf, a phylogenetic alignment is an assignment of one string to each internal node
- Each edge represents some mutational history (a series of edit operations), which transformed the ancestor string into its children
- The score of a phylogenetic alignment is the sum of scores of its edges
- Consensus is a phylogenetic alignment to a star-tree
- The problem of constructing a phylogenetic alignment with a minimal total score is NP-complete, in addition, the tree topology should be known in advance