Sequence Alignment

Brief review from last class

- DNA is has direction, we will use only one (5’ -> 3’) and generate the opposite strand as needed.

- DNA is a 3D object (see lecture 1) but we will model it as a 2D object/string.

- Two generative models of sequences:
  - Multinomial: probability is equal to product of individual probabilities (no prior dependence)
  - Markov: probabity is equal to product of probabilities given a fixed number of preceding characters.

Today

- The next two weeks, we will discuss sequence alignment and all of its basic flavors.

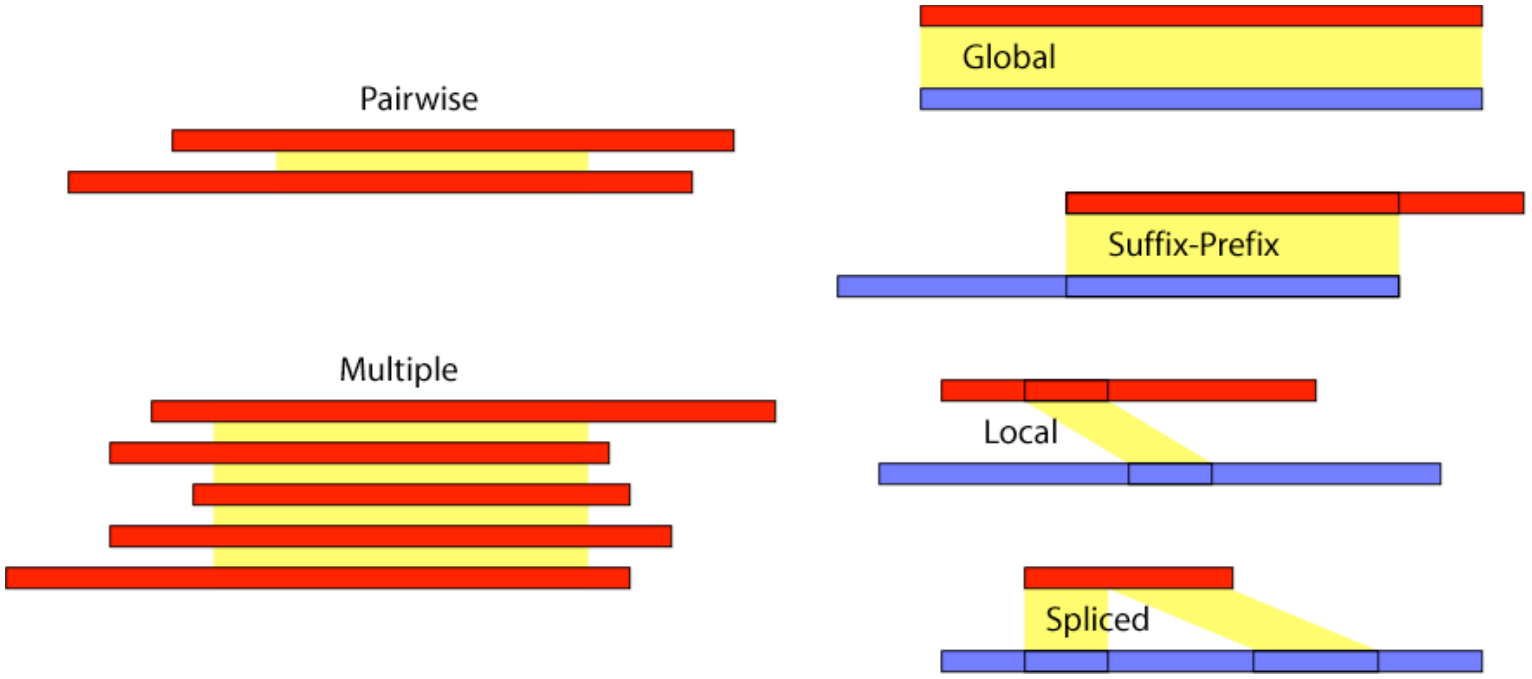
- Arguably one of the most important algorithms in bioinformatics; over 40 years old.

- The ultimate goal of alignment is to describe sequence similarity, or how closely two sequences match each other.
  - Can be a score (number)
  - Can also be an “alignment” (visual)

Similarity vs. biology

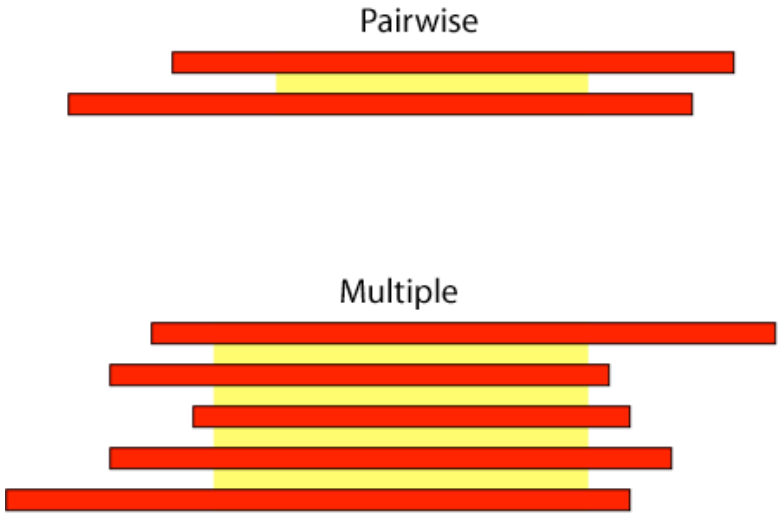
- Similarity (also called identity) is the number of matches / alignment length
- Homology, on the other hand, implies sequences came from a common ancestor
- Two kinds of homology:
  - Orthologous - speciation-based split
  - Paralogous - gene duplication-based split

Various types



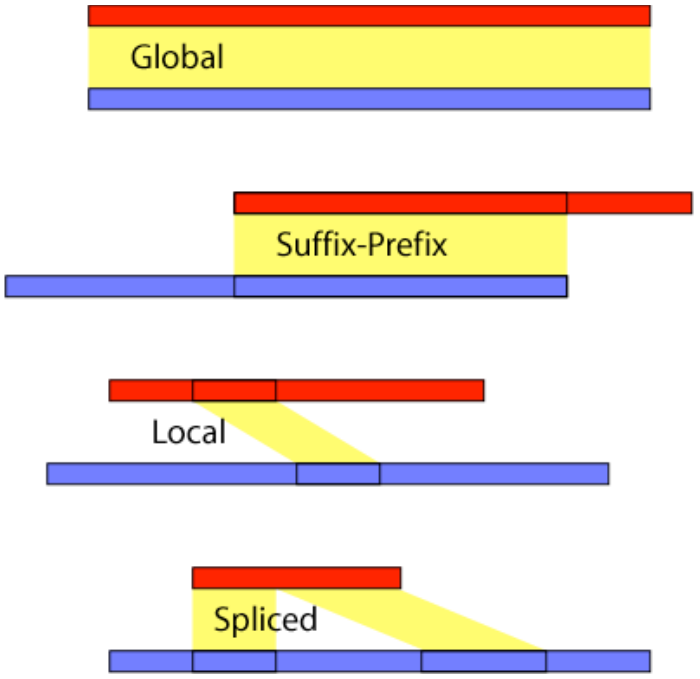
Examples

Database search



Phylogeny

Protein structure



Gene finding

Applications

- Prediction on function
  - Commonalities among sequences can imply similar functions
- Database searching (BLAST)
  - Find interesting genes in a new genome
- Sequence divergence
  - Look at evolutionary relationships
- Sequence assembly
  - Making a big sequence from a bunch of small ones

Global alignment

- Also called a *pairwise alignment.*

- Intuitive goal: related sequences will share many (most?) characters. To maximize this we introduce gaps represented by "-"

Two simple rules

- Rule #1:
  - A gap must be aligned to a nongap, i.e., "-" can not align to "-"
- Rule #2:
  - To distinguish good alignment from not so good ones, we introduce a scoring function *E*. Some functions have biological meaning, some are arbitrary.
- Consequence #1:
  - Alignment length can be no longer than sum of two sequences!

Example from text

- How do we align these proteins:
  - VIVALASVEGAS
  - VIVADAVIS

Alignments

- Show one sequence placed above another such that similarity is revealed.
- Alignments can be longer than either string!

Example:

```
A: C  A  T  -  T  C  A  -  C
B: C  -  T  C  G  C  A  G  C
```

Improving readability

Example:

```
A: C  A  T  -  T  C  A  -  C
   |     |     |     |     |
B: C  -  T  C  G  G  A  G  C
```

Scoring functions

- Here is a basic scoring function that rewards 1 for a match and -1 for a mismatch gap

$$E(-,a) = E(a,-) = E(a,b) = -1 \quad \forall a \neq b$$
$$E(a,b) = 1 \quad \forall a = b$$

- Can also be represented as a substitution matrix.

In class example

S: CATCAC
T: CTCCAGC

$$E(-,a) = E(a,-) = E(a,b) = -1 \quad \forall a \neq b$$
$$E(a,b) = 1 \quad \forall a = b$$

Measuring similarity

Score: A measure of alignment quality

```
  C  A  T  -  T  C  A  -  C
  C  -  T  C  G  C  A  G  C
-------------------------------
 10 -5 10 -5 -2 10 10 -5 10

Total =  33

Scored as E(C,C) E(A,-), E(T,T), E(-,C),
  etc.
```

Alignment overview

- Computationally, naïve alignments grow exponentially with n : not good
  - There are 10^{17} alignments for two length 30 sequences.

- Luckily, a tried and true method for solving similar problems (we'll provide an overview today) comes to the rescue.

- First efficient algorithm published in 1970 by Needleman and Wunch, improved by Smith and Waterman in 1981.

Basic intuition

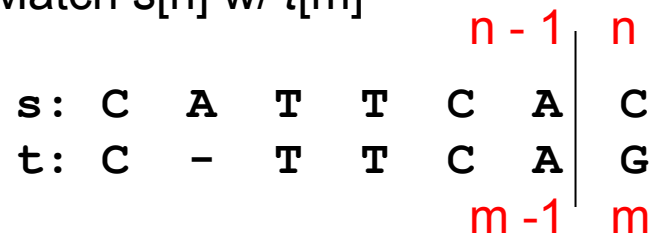
- Suppose we have an optimum alignment of size L. Is the following true?

- $A^* = A^*(s_1 \ldots s_i, t_1 \ldots t_j) + A^*(s_{i+1} \ldots s_n, t_{j+1} \ldots t_m)$
  - Where $|s| = n$ and $|t| = m$

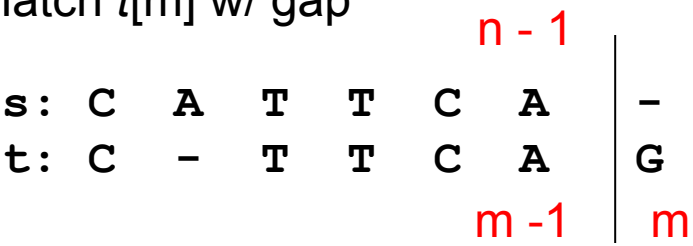
- If so, what would happen if $i = n - 1$ and $j = m - 1$?

Visualization

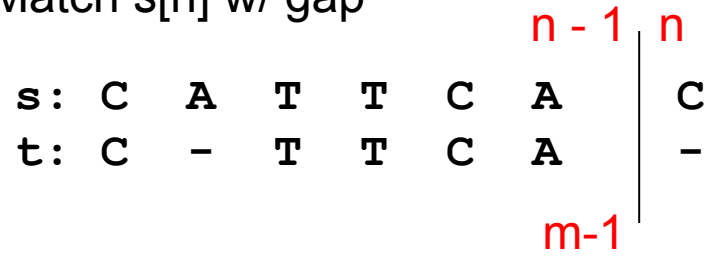
Case 1: Match *s*[n] w/ *t*[m]



Case 2: Match *t*[m] w/ gap



Case 3: Match *s*[n] w/ gap



Global alignment

- Dynamic programming (DP) will save the day!
- DP is a general technique used when a large problem can be broken into smaller, easier problems like this.
- To solve sequence alignment, we will fix two substrings and find the best way to add the next character from at least one string.

Notation from Jackson and Aluru

- S(i,j) = $E(Opt(A[0,i],B[0,j]))$
  - "S(i,j) is the evaluated score of the optimal alignment between the prefix of A ending at position i and the prefix of B ending at position j."

$$S(n-1,m-1)=\max\begin{cases} S(n-2,m-2)+E(A[n-1],B[m-1]) \\ S(n-1,m-2)+E('-',B[m-1]) \\ S(n-2,m-1)+E(A[n-1],'-') \end{cases}$$

$$S(i,j)=\max\begin{cases} S(i-1,j-1)+E(A[i],B[j]) \\ S(i,j-1)+E('-',B[j]) \\ S(i-1,j)+E(A[i],'-') \end{cases}$$

Requirements

- We will need four things to compute a global alignment:
  1. Substitution matrix (parameters)
  2. Recurrence relation
  3. Filling up a table
  4. Traceback

Pairwise Global Alignment

$T[i,j]$ = Score of optimally aligning first i bases of s with first j bases of t.

$$T[i,j] = \max \begin{cases} T[i-1,j-1] + score(s[i], t[j]) \\ T[i-1,j] + g \\ T[i,j-1] + g \end{cases}$$

	λ	C	T	C	G	C	A	G	C
λ	0	-5	-10	-15	-20	-25	-30	-35	-40
C	-5	10	5						
A	-10								
T	-15								
T	-20								
C	-25								
A	-30								
C	-35								

+10 for match, -2 for mismatch, -5 for space (rowwise)

	λ	C	T	C	G	C	A	G	C
λ	0	-5	-10	-15	-20	-25	-30	-35	-40
C	-5	10	5	0	-5	-10	-15	-20	-25
A	-10	5	8	3	-2	-7	0	-5	-10
T	-15	0	15	10	5 *	0	-5	-2	-7
T	-20	-5	10 *	13	8	3	-2	-7	-4
C	-25	-10	5	20	15	18	13	8	3
A	-30	-15	0	15	18	13	28	23	18
C	-35	-20	-5	10	13	28	23	26	33

Traceback yields both optimal alignments in this example

Some Results

- Most pairwise sequence alignment problems can be solved in $O(mn)$ time. Some speedups exist, most notably the Four Russians technique.

- Space requirement can be reduced to $O(m+n)$, while keeping run-time fixed [Myers88].

- Two highly similar sequences can be aligned in $O(dn)$ time, where d is a measure of the distance between the sequences [Landau86].

Pairwise Sequence Alignment

Variations for future classes:

- Given two sequences, find if parts of them are similar (local alignment).
- Given a large sequence and a short sequence, find if the short sequence is similar to a stretch of the long sequence.
- Cool fact is these are easy to do once we learn the basics of global alignment!