Multiple Sequence Alignment

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Examples

Sequence Similarity

Which pair of sequences are the closest?

$$S_1 = AAAAA \qquad S_2 = AGAGA \qquad S_3 = GGAAA$$

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Orthology Detection

$$\left(\begin{array}{c|cccccccccc} S_1 & L & A & S & T & F & A & - & T & C & A & T \\ S_2 & L & A & S & T & C & A & - & T & - & - & - \\ S_3 & V & E & R & Y & F & A & S & T & C & A & T \\ S_4 & - & - & - & - & F & A & - & T & C & A & T \end{array}\right)$$

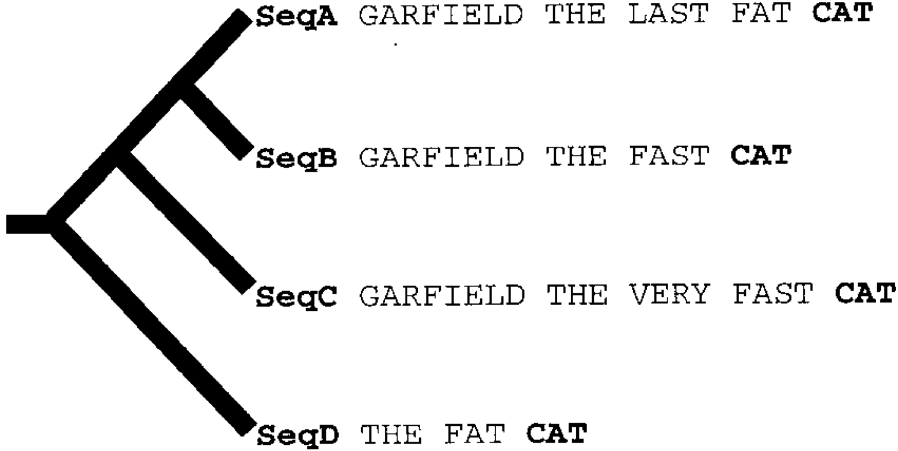
Examples

Orthology Detection

$$\left(\begin{array}{c|cccc|cccc|ccc} S_1 & L & A & S & T & F & A & - & T & C & A & T \\ S_2 & L & A & S & T & - & - & - & - & C & A & T \\ S_3 & V & E & R & Y & F & A & S & T & C & A & T \\ S_4 & - & - & - & - & F & A & - & T & C & A & T \end{array}\right)$$

Examples

Phylogeny Estimation



Examples

Scheduling

Outline

Multiple Sequence Alignment

Problem (Multiple Sequence Alignment)

Given a set of sequences $S_1, \ldots, S_n$, and a scoring function d, find a *multiple sequence alignment* $\mathscr{A} = (a_{ij})_{1 \leq i \leq n}$ minimizing

$$\sum_j d(a_{1j}, \ldots, a_{nj})$$

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$$d(\cdot) = \begin{cases} +\infty & \text{if number unique characters is more than 1} \\ 1 & \text{otherwise} \end{cases}$$

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$$d(\cdot) = \begin{cases} 0 & \text{if number unique characters is more than 1} \\ -1 & \text{otherwise} \end{cases}$$

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$$d(\cdot) = \#\text{ dashes} + \#\text{ unique} - 1$$

Pariwise Alignments

Edit Distance

	λ	T	T	A	A	G	C
λ	**0**	1	2	3	4	5	6
A	**1**	1	2	2	3	4	5
A	**2**	2	2	2	2	3	4
T	3	**2**	2	3	3	3	4
T	4	3	**2**	2	3	4	4
A	5	4	3	**2**	2	3	4
A	6	5	4	3	**2**	3	4
G	7	6	5	4	3	**2**	**3**

Alignment Graphs

Definition (Alignment Graph)

Given a set of sequences $S_1, \ldots, S_n$, and a scoring function d, and a set of *pairwise* alignments $A_1, \ldots, A_k$, construct $G = (V, E, \prec)$, where

1. For each sequence, for each *site* s_{ij}, create a vertex
2. For each alignment A_i, for each *homology* s_{ij}, s_{kl}, add weight $d(s_{ij}, s_{kl})$ to the edge
3. For each pair site s_{ij} and $s_{ij'}$ where $j' > j$, add $s_{ij}, s_{ij'}$ to $\prec$

Alignment Graphs

$$S_1 = ABCD \qquad S_2 = BAC \qquad S_3 = AAD$$

$$A_1 = \left(\begin{array}{c|ccccc} S_1 & A & B & - & C & D \\ S_2 & - & B & A & C & - \end{array}\right)$$

$$A_2 = \left(\begin{array}{c|cccc} S_2 & B & A & - & C \\ S_3 & - & A & A & D \end{array}\right)$$

$$A_3 = \left(\begin{array}{c|ccccc} S_1 & - & A & B & C & D \\ S_3 & A & A & - & - & D \end{array}\right)$$

Alignment Graphs

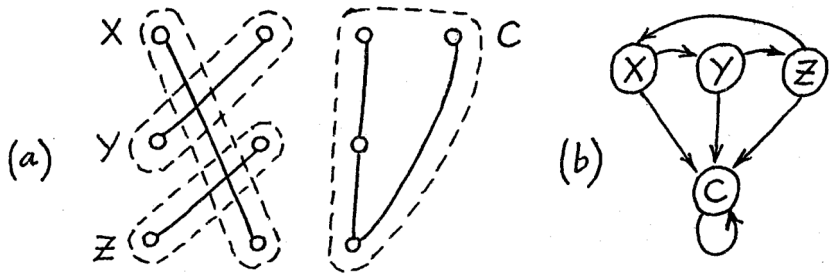

Fig. 1. (a) An alignment graph on three sequences. We use the convention of drawing the characters in a sequence horizontally left to right. (b) Relation $\prec^*$ on its connected components.

Alignment Graphs

Definition (Trace)

A *trace* of an alignment graph $G = (V, E, \prec)$ is a subset of the $T \subset E$ where $G^* = (V, T, \prec^*)$ is acyclic.

$$\mathcal{A}_1 = \left(\begin{array}{c|cccccc} S_1 & A & B & - & C & - & D \\ S_2 & - & B & A & C & - & - \\ S_3 & - & - & A & - & A & D \end{array}\right)$$

$$\mathcal{A}_2 = \left(\begin{array}{c|cccccc} S_1 & - & - & A & B & C & D \\ S_2 & B & A & - & - & C & - \\ S_3 & - & A & A & - & - & D \end{array}\right)$$

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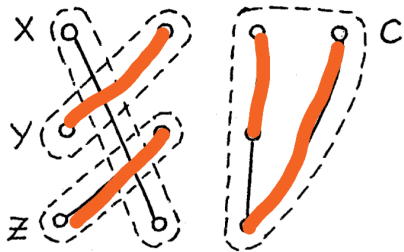

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Maximum Weight Trace

Problem (Maximum Weight Trace (MWT))

Given an alignment graph $G = (V, E, \prec)$, find the trace T that *maximizes*

$$\sum_{e \in T} w(e)$$

Maximum Weight Trace

Theorem (Kececioglu'93)

Maximum Weight Trace is NP-Hard

Proof.

Consider an instance $G = (V, E)$ and integer k of *Feedback Set*.

1. For every vertex v, create sequence $S_v = v$
2. For every edge $u \to v$, create sequence $S_{uv} = uv$
3. Create pairwise alignments

$$\begin{pmatrix} S_u & u & - \\ S_{uv} & u & v \end{pmatrix} \qquad \begin{pmatrix} S_v & - & v \\ S_{uv} & u & v \end{pmatrix}$$

4. Check if MWT is at least $2k$.

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Maximum Weight Trace

Let $D(x_1, \ldots, x_n)$ denote maximum weight trace over *prefixes* $S_i[1 : x_i]$. Then,

$$D(\vec{x}) = \max_{\vec{b} \in [2]^n} \{ D(\vec{x} - \vec{b}) + d(\vec{S}^{\vec{b}}) \}$$

Thus, MWT can be solved in $O((2k)^n \mathit{poly}(n))$.

Using the *Branch-and-Bound* paradigm, this can be fast.

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From Fa'24 CS 374 homework 14, this can be improved to $O(nk^n poly(n))$.

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T-COFFEE

1. Aggregate pairwise alignments through triples
2. Calculate new pairwise distances
3. Compute guide trees
4. Progressively align using guide tree

T-COFFEE

b)Primary Library

```
SeqA GARFIELD THE LAST FAT CAT
SeqB GARFIELD THE FAST CAT ---
```
Prim. Weight = 88

```
SeqA GARFIELD THE LAST FA-T CAT
SeqC GARFIELD THE VERY FAST CAT
```
Prim. Weight = 77

```
SeqA GARFIELD THE LAST FAT CAT
SeqD ------- THE ---- FAT CAT
```
Prim. Weight =100

```
SeqB GARFIELD THE ---- FAST CAT
SeqC GARFIELD THE VERY FAST CAT
```
Prim Weight = 100

```
SeqB GARFIELD THE FAST CAT
SeqD ------- THE FA-T CAT
```
Prim. Weight = 100

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SeqC GARFIELD THE VERY FAST CAT
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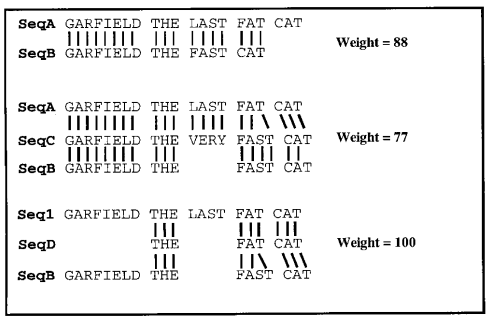
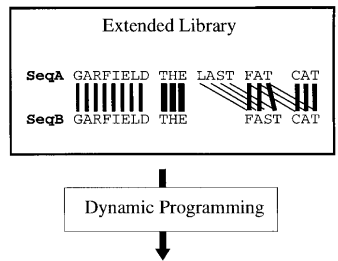
T-COFFEE

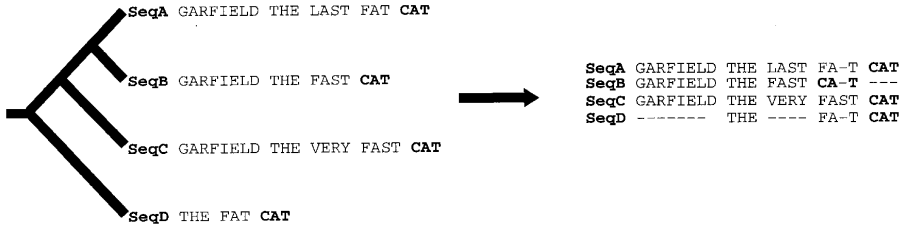
c)Extended Library for seq1 and seq2





T-COFFEE

a)Regular Progressive Alignment Strategy



MAGUS

1. Create alignment graph from backbone alignments
2. Cluster with Markov Clustering (MCL)
3. Break all clusters that violate ordering

MAGUS

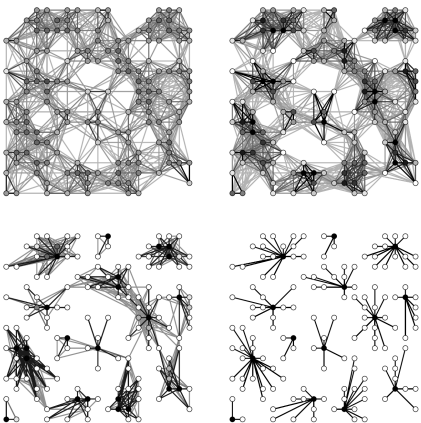


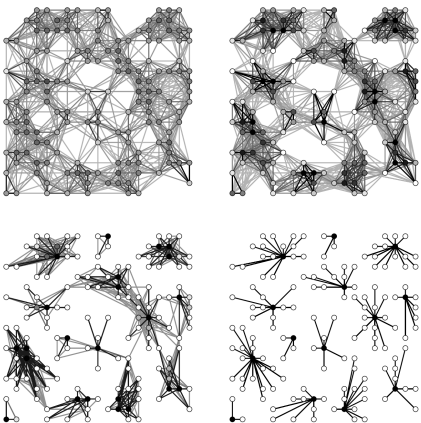
Figure 3. Successive stages of flow simulation by the *MCL* process.

Clusters have high *edge connectivity*. A random walk is *likely* to stay within the cluster.

Markov Clustering Algorithm

1. Expansion (random walk)
2. Inflation (amplify probabilities)
3. Repeat

MAGUS



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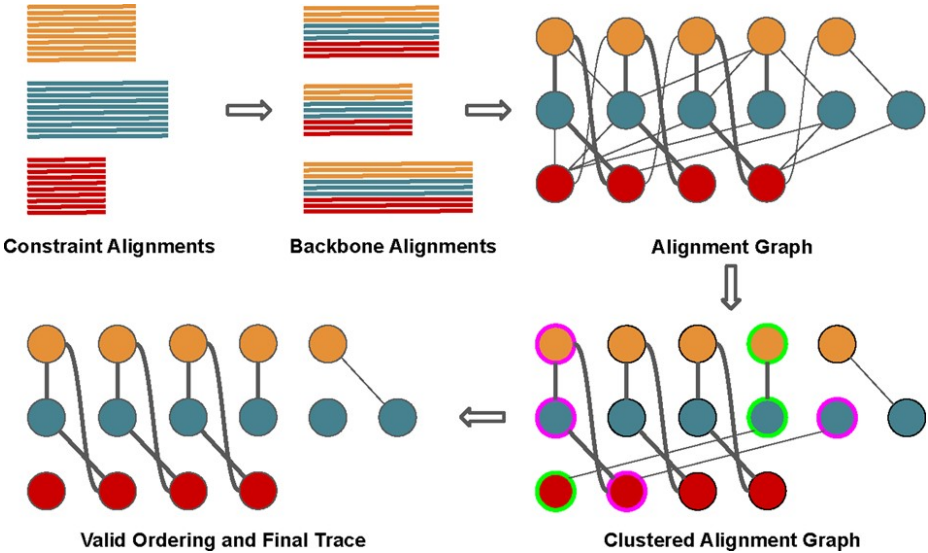
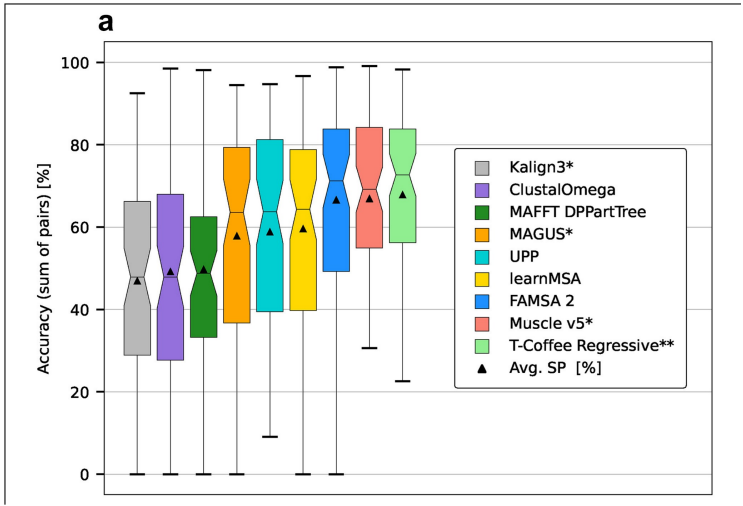
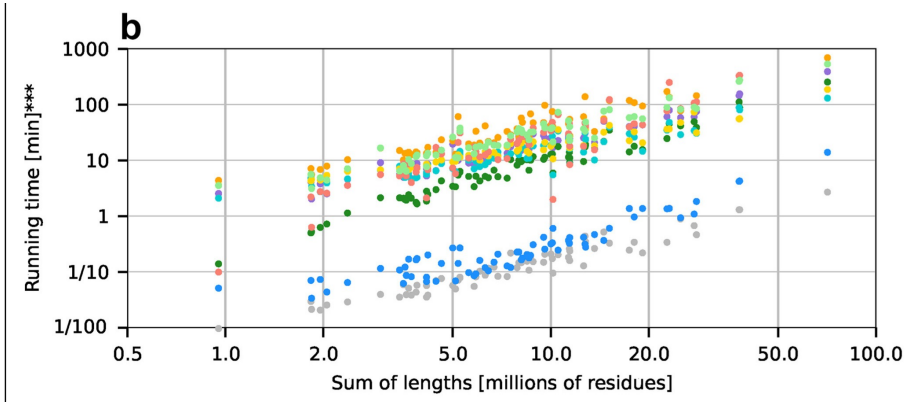

Constraint Alignments
Backbone Alignments
Alignment Graph
Clustered Alignment Graph
Valid Ordering and Final Trace

Results


a
Accuracy (sum of pairs) [%]
100
80
60
40
20
0
Kalign3*
ClustalOmega
MAFFT DPPartTree
MAGUS*
UPP
learnMSA
FAMSA 2
Muscle v5*
T-Coffee Regressive**
Avg. SP [%]

Results


b
1000
100
10
1
1/10
1/100
Running time [min]***
0.5
1.0
2.0
5.0
10.0
20.0
50.0
100.0
Sum of lengths [millions of residues]

Results

1. T-Coffee (Regressive) is the best
2. Consistency based methods are very good (MAGUS, T-Coffee, Muscle)
3. Single-stage aligners are bad (Kalign, ClustalOmega, ...)
4. Exception for FAMSA

Questions

1. How can we encode genome events into the alignment graph?
2. Can T-COFFEE perform better if we give it multiple sequence alignments (instead of pairwise) as input?
3. Do other clustering algorithms (beyond MCL) cluster the alignment graph better in MAGUS?

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