Scoring Alignments

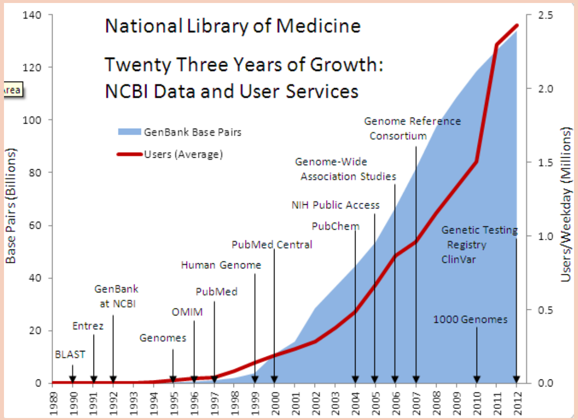
Genome 373

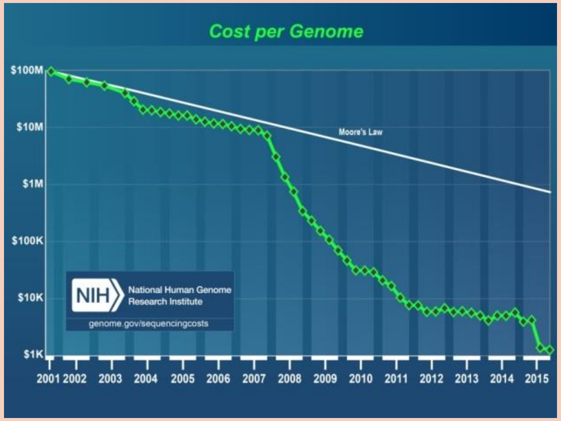
Genomic Informatics

Elhanan Borenstein

A quick review

- The computational bottleneck
  - Scale of biological data





  - Complexity of tasks

```
G  –  A  A  T  T  C  A  G  T  T  A
|        |     |  |     |        |
G  G  –  A  –  T  C  –  G  -  -  A
```

A quick review: Informatic challenges

- Sequence comparison:
  - Find the best alignment of two sequences
  - Find the best match (alignment) of a given sequence in a large dataset of sequences
  - Find the best alignment of multiple sequences
- Motif and gene finding
- Relationship between sequences
  - Phylogeny
- Clustering and classification
- Many many many more …

A quick review: Informatic challenges

- **Sequence comparison:**
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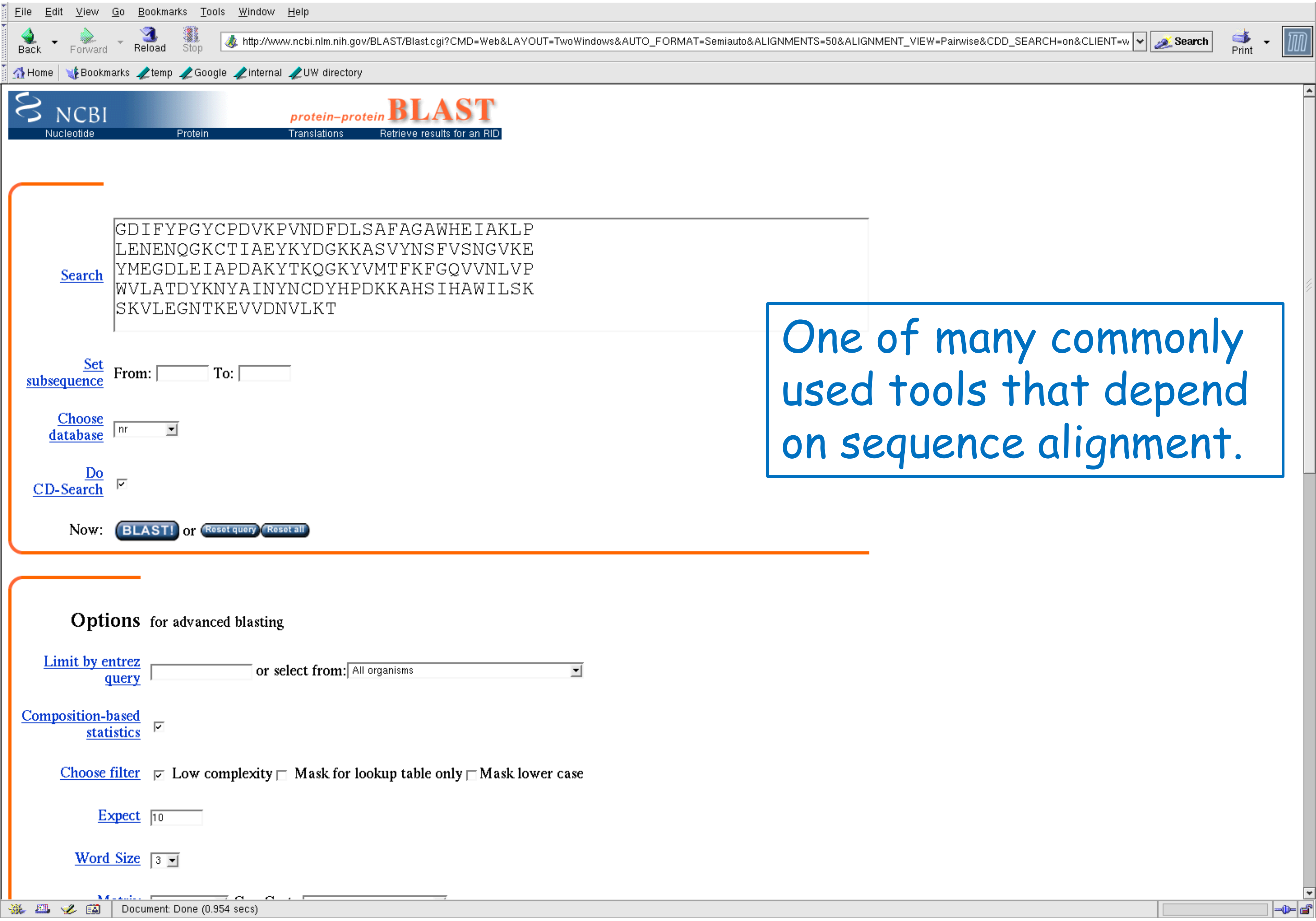
Motivation

- **Why compare two DNA or protein sequences?**

Motivation

- **Why compare two DNA or protein sequences?**
  - Determine whether they are descended from a common ancestor (homologous)
  - Infer a common function
  - Locate functional elements (motifs or domains)
  - Infer protein or RNA structure, if the structure of one of the sequences is known
  - Analyze sequence evolution
  - Infer the species from which a sequence originated
  - **Quantify abundance/coverage**



http://www.ncbi.nlm.nih.gov/BLAST/Blast.cgi?CMD=Web&LAYOUT=TwoWindows&AUTO_FORMAT=Semiauto&ALIGNMENTS=50&ALIGNMENT_VIEW=Pairwise&CDD_SEARCH=on&CLIENT=w

protein–protein BLAST

Search

```
GDIFYPGYCPDVKPVNDFDLSAFAGAWHEIAKLP
LENENQGKCTIAEYKYDGKKASVYNSFVSNGVKE
YMEGDLEIAPDAKYTKQGKYVMTFKFGQVVNLVP
WVLATDYKNYAINYNCDYHPDKKAHSIHAWILSK
SKVLEGNTKEVVDNVLKT
```

Set subsequence From: To:

Choose database: nr

Do CD-Search ☑

Now: BLAST! or Reset query Reset all

Options for advanced blasting

Limit by entrez query or select from: All organisms

Composition-based statistics ☑

Choose filter ☑ Low complexity □ Mask for lookup table only □ Mask lower case

Expect: 10

Word Size: 3

Document: Done (0.954 secs)

One of many commonly used tools that depend on sequence alignment.

Sequence Comparison Challenges

- Find the best *global* alignment of two sequences

- Find the best *global* alignment of multiple sequences

- Find the best *local (partial)* alignment of two sequences

- Find the best match (alignment) of a given sequence in a longer dataset of sequences

Sequence Comparison Challenges

✓ Find the best *global* alignment of two sequences

✗ Find the best *global* alignment of multiple sequences

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✓✗ Find the best match (alignment) of a given sequence in a longer dataset of sequences

Global Alignment Mission:

Find the best global alignment between two sequences.

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Find the best alignment of GAATC and CATAC:

```
GAATC
CATAC
```

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GAAT-C
C-ATAC
```

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-GAAT-C
C-A-TAC
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GAATC-
CA-TAC
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```
GAAT-C
CA-TAC
```

```
GA-ATC
CATA-C
```

```
GAAT-C
CA-TAC
```

(some of a very large number of possibilities)

Global Alignment Mission:

Find the best global alignment between two sequences.

Find the best alignment of GAATC and CATAC:

```
-GAAT-C
C-A-TAC
```

Conceptually:

- What does a “correct” alignment mean?
- Correct vs. Best

Global Alignment Mission:

Find the best global alignment between two sequences.

Find the best alignment of GAATC and CATAC:

```
-GAAT-C
C-A-TAC
```

Technically:

- This is a search (optimization) problem!!
- What do we need to solve this problem?

Global Alignment Mission:

Find the best global alignment between two sequences.

An algorithm for finding the alignment with the best score

A method for scoring alignments

Scoring Principles

```
GAATC
CATAC
```

- Score each locus independently.
- The alignment score will be the sum of the scores in all loci.
- Perfect Matches will get a positive (good) score.
- What about mismatches?

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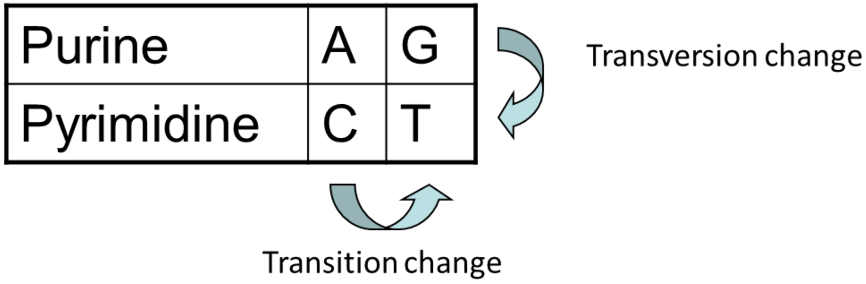


Purine	A	G
Pyrimidine	C	T

Transversion change

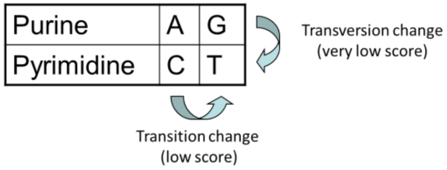
Transition change

(transitions are typically about 2x as frequent as transversions in real sequences)

Scoring Aligned Bases

- A reasonable **substitution matrix:**

	A	C	G	T
A	10	-5	0	-5
C	-5	10	-5	0
G	0	-5	10	-5
T	-5	0	-5	10



Purine	A	G
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What about gaps?

```
GAATC
CATAC
```

-5 + 10 + -5 + -5 + 10 = 5

What About Gaps?

- A reasonable **substitution matrix:**

	A	C	G	T
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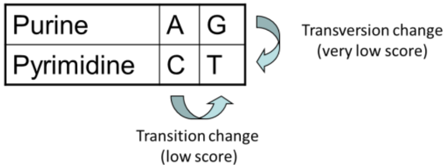
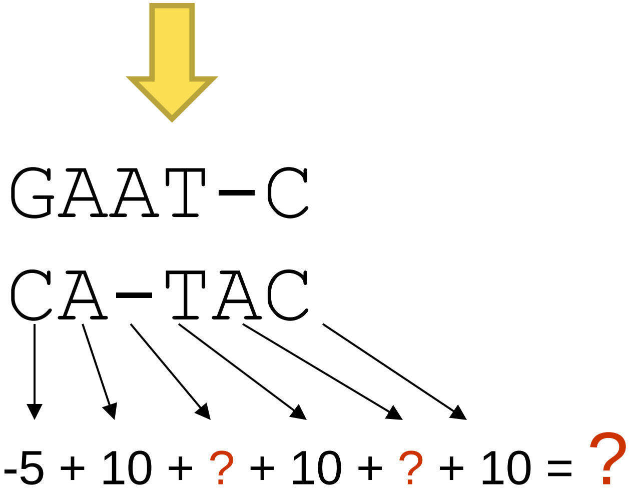


Purine	A	G
Pyrimidine	C	T

Transversion change (very low score)
Transition change (low score)

What do gaps mean?

What if gaps have no penalty?



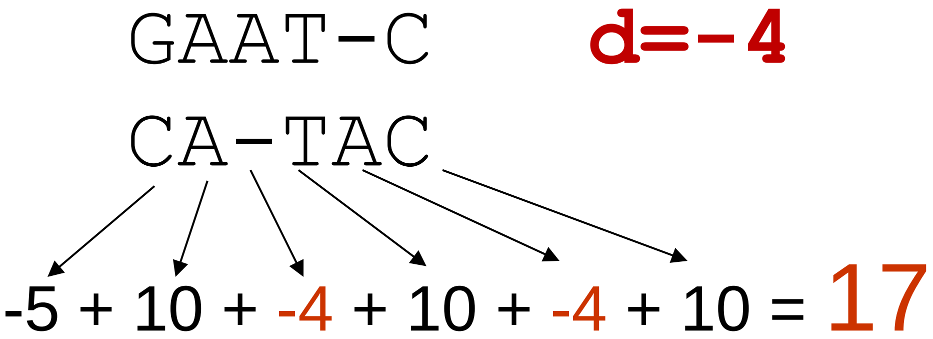
GAAT-C

CA-TAC

-5 + 10 + ? + 10 + ? + 10 = ?

Scoring Gaps?

- **Linear** gap penalty: every gap receives a score of **d**:

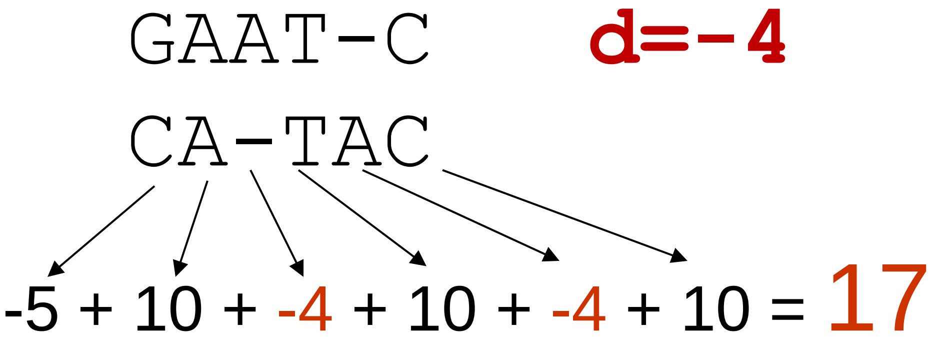


```
GAAT-C      d=-4
CA-TAC
```

-5 + 10 + -4 + 10 + -4 + 10 = 17

Scoring Gaps?

- **Linear** gap penalty: every gap receives a score of **d**:



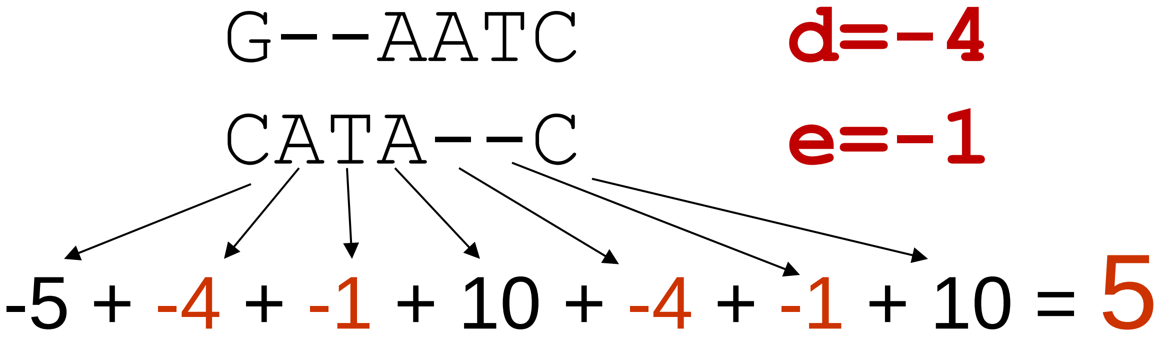
GAAT-C

CA-TAC

d=-4

-5 + 10 + -4 + 10 + -4 + 10 = 17

- **Affine** gap penalty: opening a gap receives a score of **d**; extending a gap receives a score of **e**:



G--AATC

CATA--C

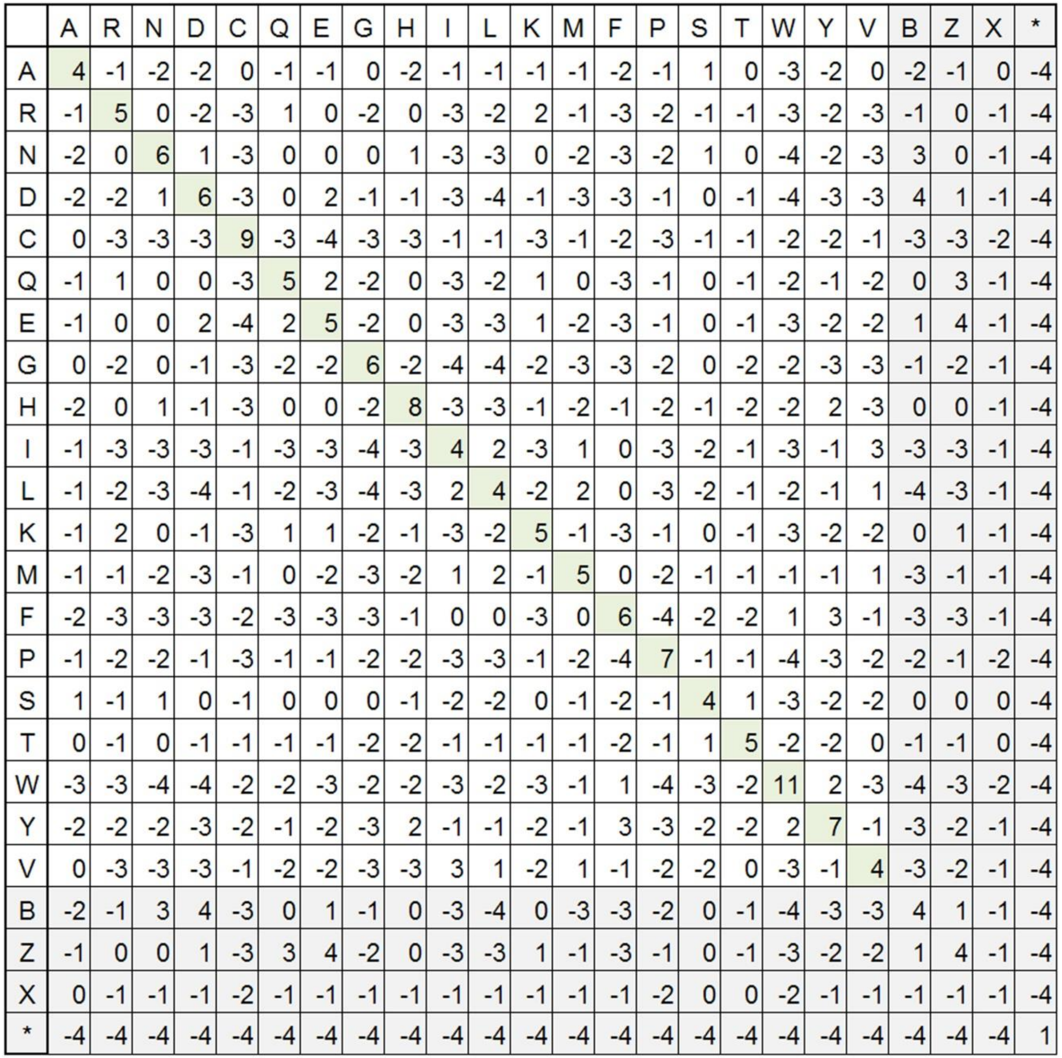
d=-4

e=-1

-5 + -4 + -1 + 10 + -4 + -1 + 10 = 5

Same Method Applies to AA

BLOSUM62 Score Matrix



	A	R	N	D	C	Q	E	G	H	I	L	K	M	F	P	S	T	W	Y	V	B	Z	X	*
A	4	-1	-2	-2	0	-1	-1	0	-2	-1	-1	-1	-1	-2	-1	1	0	-3	-2	0	-2	-1	0	-4
R	-1	5	0	-2	-3	1	0	-2	0	-3	-2	2	-1	-3	-2	-1	-1	-3	-2	-3	-1	0	-1	-4
N	-2	0	6	1	-3	0	0	0	1	-3	-3	0	-2	-3	-2	1	0	-4	-2	-3	3	0	-1	-4
D	-2	-2	1	6	-3	0	2	-1	-1	-3	-4	-1	-3	-3	-1	0	-1	-4	-3	-3	4	1	-1	-4
C	0	-3	-3	-3	9	-3	-4	-3	-3	-1	-1	-3	-1	-2	-3	-1	-1	-2	-2	-1	-3	-3	-2	-4
Q	-1	1	0	0	-3	5	2	-2	0	-3	-2	1	0	-3	-1	0	-1	-2	-1	-2	0	3	-1	-4
E	-1	0	0	2	-4	2	5	-2	0	-3	-3	1	-2	-3	-1	0	-1	-3	-2	-2	1	4	-1	-4
G	0	-2	0	-1	-3	-2	-2	6	-2	-4	-4	-2	-3	-3	-2	0	-2	-2	-3	-3	-1	-2	-1	-4
H	-2	0	1	-1	-3	0	0	-2	8	-3	-3	-1	-2	-1	-2	-1	-2	-2	2	-3	0	0	-1	-4
I	-1	-3	-3	-3	-1	-3	-3	-4	-3	4	2	-3	1	0	-3	-2	-1	-3	-1	3	-3	-3	-1	-4
L	-1	-2	-3	-4	-1	-2	-3	-4	-3	2	4	-2	2	0	-3	-2	-1	-2	-1	1	-4	-3	-1	-4
K	-1	2	0	-1	-3	1	1	-2	-1	-3	-2	5	-1	-3	-1	0	-1	-3	-2	-2	0	1	-1	-4
M	-1	-1	-2	-3	-1	0	-2	-3	-2	1	2	-1	5	0	-2	-1	-1	-1	-1	1	-3	-1	-1	-4
F	-2	-3	-3	-3	-2	-3	-3	-3	-1	0	0	-3	0	6	-4	-2	-2	1	3	-1	-3	-3	-1	-4
P	-1	-2	-2	-1	-3	-1	-1	-2	-2	-3	-3	-1	-2	-4	7	-1	-1	-4	-3	-2	-2	-1	-2	-4
S	1	-1	1	0	-1	0	0	0	-1	-2	-2	0	-1	-2	-1	4	1	-3	-2	-2	0	0	0	-4
T	0	-1	0	-1	-1	-1	-1	-2	-2	-1	-1	-1	-1	-2	-1	1	5	-2	-2	0	-1	-1	0	-4
W	-3	-3	-4	-4	-2	-2	-3	-2	-2	-3	-2	-3	-1	1	-4	-3	-2	11	2	-3	-4	-3	-2	-4
Y	-2	-2	-2	-3	-2	-1	-2	-3	2	-1	-1	-2	-1	3	-3	-2	-2	2	7	-1	-3	-2	-1	-4
V	0	-3	-3	-3	-1	-2	-2	-3	-3	3	1	-2	1	-1	-2	-2	0	-3	-1	4	-3	-2	-1	-4
B	-2	-1	3	4	-3	0	1	-1	0	-3	-4	0	-3	-3	-2	0	-1	-4	-3	-3	4	1	-1	-4
Z	-1	0	0	1	-3	3	4	-2	0	-3	-3	1	-1	-3	-1	0	-1	-3	-2	-2	1	4	-1	-4
X	0	-1	-1	-1	-2	-1	-1	-1	-1	-1	-1	-1	-1	-1	-2	0	0	-2	-1	-1	-1	-1	-1	-4
*	-4	-4	-4	-4	-4	-4	-4	-4	-4	-4	-4	-4	-4	-4	-4	-4	-4	-4	-4	-4	-4	-4	-4	1

regular 20 amino acids

ambiguity codes and stop

Y mutates to V receives -1
M mutates to L receives 2
E gets deleted receives -10
G gets deleted receives -10
D matches D receives 6

Total score = -13

```
YMEGDLEIAPDAK
VL--DKELSPDGT
```

Global Alignment Mission:

Find the best global alignment between two sequences.

An algorithm for finding the alignment with the best score

?

A method for scoring alignments

- **Substitution matrix:**

	A	C	G	T
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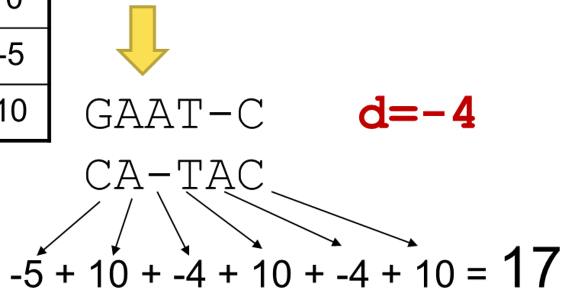
- **Gap penalty:**
  - **Linear** gap penalty
  - **Affine** gap penalty



```
GAAT-C
CA-TAC
```

d=-4

-5 + 10 + -4 + 10 + -4 + 10 = 17

A simple algorithm ….

- *Align the two sequences: GAATC and CATAC*

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CA-TAC
```

```
GAAT-C
CA-TAC
```

```
GA-ATC
CATA-C
```

```
GAAT-C
CA-TAC
```

<u>Simple (exhaustive search) algorithm</u>

1) Construct all possible alignments

2) Use the substitution matrix and gap penalty to score each alignment

3) Pick the alignment with the best score

How many possibilities?

- *Align the two sequences: GAATC and CATAC*

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```

- How many different possible alignments of two sequences of length ***n*** exist?

5	2.5x10^{2}
10	1.8x10^{5}
20	1.4x10^{11}
30	1.2x10^{17}
40	1.1x10^{23}

Mission:

Find the best alignment between two sequences.

A algorithm for finding the alignment with the best score

- Needleman–Wunsch Algorithm
- Dynamic programming

A method for scoring alignments

- **Substitution matrix:**

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