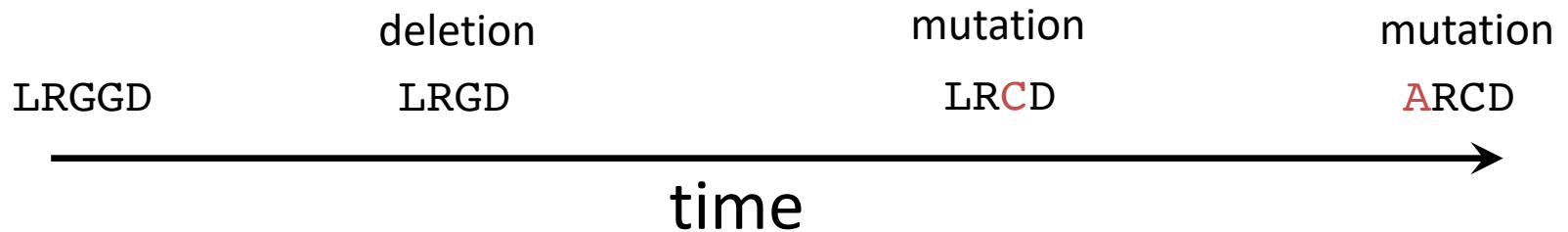


Sequence alignments

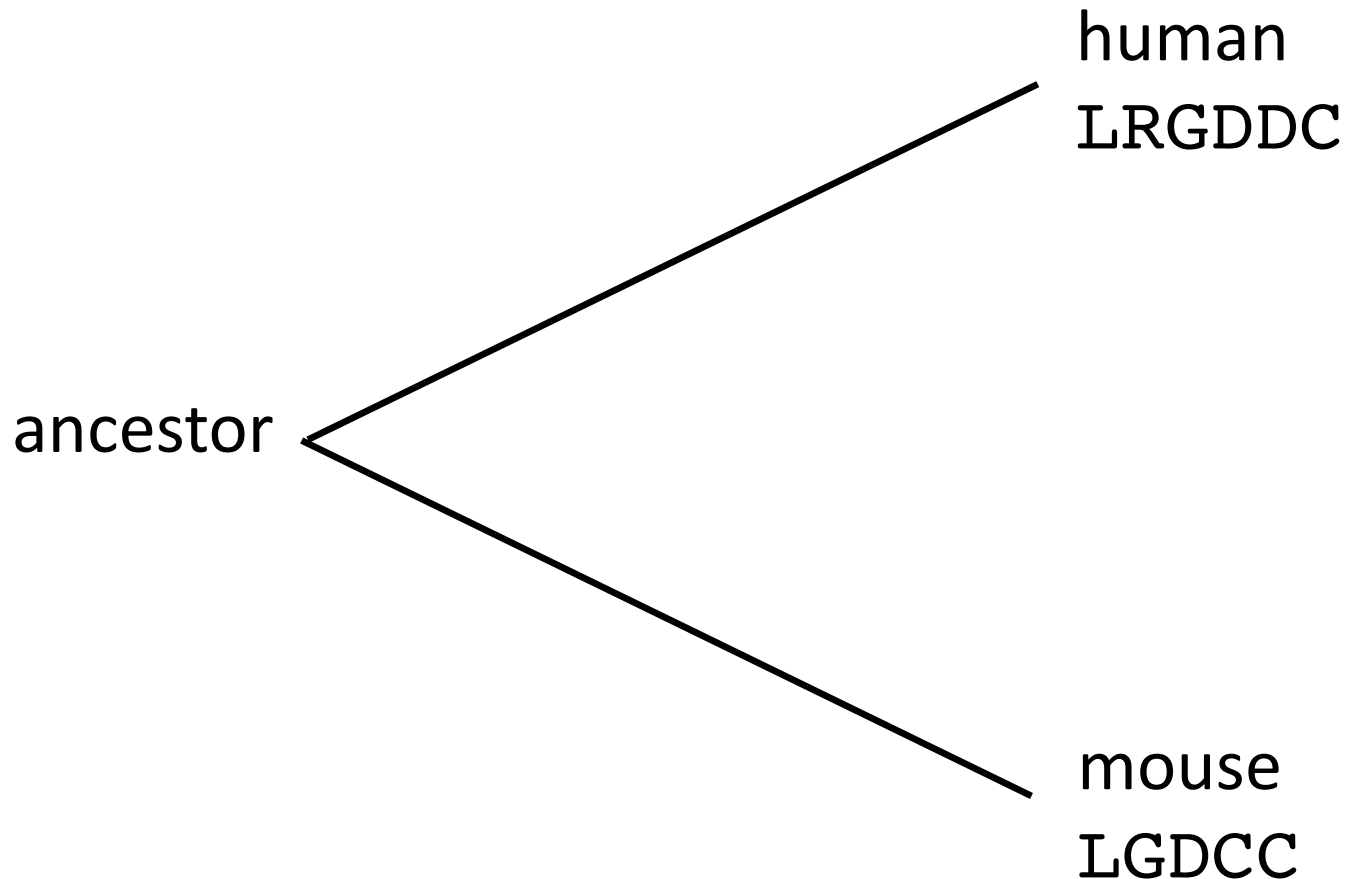
Genetic sequences change over time



Relationship between original and final sequence:

LRGGD LRGGD
AR-CD or ARC-D

In practice: we only know sequences from extant organisms



We need to align these sequences to compare them

human
LRGDDC

mouse
LGDCC

LRGDDC
L-GDCC

LRGDDC-
L-GD-CC

LRGDDC
-LGDCC

Which alignment is correct?

We need to score the alignment

Example:

- match = +1
- mismatch = -1
- gap = 0

LRGDDC score = 1+0+1+1-1+1
L-GDCC = 3

LRGDDC- score = 1+0+1+1+0+1+0
L-GD-CC = 4

LRGDDC score = 0-1+1+1-1+1
-LGDCC = 1

We need to score the alignment

Example:

- match = +1
- mismatch = -1
- gap = -2

LRGDDC	score = 1-2+1+1-1+1
L-GDCC	= 1

LRGDDC-	score = 1-2+1+1-2+1-2
L-GD-CC	= -2

LRGDDC	score = -2-1+1+1-1+1
-LGDC	= -1

We often score by amino-acid similarity

Ala	4																			
Arg	-1	5																		
Asn	-2	0	6																	
Asp	-2	-2	1	6																
Cys	0	-3	-3	-3	9															
Gln	-1	1	0	0	-3	5														
Glu	-1	0	0	2	-4	2	5													
Gly	0	-2	0	-1	-3	-2	-2	6												
His	-2	0	1	-1	-3	0	0	-2	8											
Ile	-1	-3	-3	-3	-1	-3	-3	-4	-3	4										
Leu	-1	-2	-3	-4	-1	-2	-3	-4	-3	2	4									
Lys	-1	2	0	-1	-3	1	1	-2	-1	-3	-2	5								
Met	-1	-1	-2	-3	-1	0	-2	-3	-2	1	2	-1	5							
Phe	-2	-3	-3	-3	-2	-3	-3	-3	-1	0	0	-3	0	6						
Pro	-1	-2	-2	-1	-3	-1	-1	-2	-2	-3	-3	-1	-2	-4	7					
Ser	1	-1	1	0	-1	0	0	0	-1	-2	-2	0	-1	-2	-1	4				
Thr	0	-1	0	-1	-1	-1	-1	-2	-2	-1	-1	-1	-1	-2	-1	1	5			
Trp	-3	-3	-4	-4	-2	-2	-3	-2	-2	-3	-2	-3	-1	1	-4	-3	-2	11		
Tyr	-2	-2	-2	-3	-2	-1	-2	-3	2	-1	-1	-2	-1	3	-3	-2	-2	2	7	
Val	0	-3	-3	-3	-1	-2	-2	-3	-3	3	1	-2	1	-1	-2	-2	0	-3	-1	4
Ala	Arg	Asn	Asp	Cys	Gln	Glu	Gly	His	Ile	Leu	Lys	Met	Phe	Pro	Ser	Thr	Trp	Tyr	Val	

BLOSUM62 Matrix

$$score = \log \frac{p_{ij}}{p_i p_j}$$

Gaps in alignments are called “indels”

LRGDDC
L-GDCC
↑
indel

How do we find the best alignment given a scoring system?

Global alignment: Needleman-Wunsch algorithm

Example: align GCAT and GAT

Scoring: match = 1, mismatch = -1, gap = -1

	-	G	C	A	T
-					
G					
A					
T					

How do we find the best alignment given a scoring system?

Global alignment: Needleman-Wunsch algorithm

Example: align GCAT and GAT

Scoring: match = 1, mismatch = -1, gap = -1

	-	G	C	A	T
-	0				
G					
A					
T					

Alignment:

-

-

How do we find the best alignment given a scoring system?

Global alignment: Needleman-Wunsch algorithm

Example: align GCAT and GAT

Scoring: match = 1, mismatch = -1, gap = -1

	-	G	C	A	T
-	0	-1			
G					
A					
T					

Alignment:

-G

--

How do we find the best alignment given a scoring system?

Global alignment: Needleman-Wunsch algorithm

Example: align GCAT and GAT

Scoring: match = 1, mismatch = -1, gap = -1

	-	G	C	A	T
-	0	-1	-2		
G					
A					
T					

Alignment:

-GC

How do we find the best alignment given a scoring system?

Global alignment: Needleman-Wunsch algorithm

Example: align GCAT and GAT

Scoring: match = 1, mismatch = -1, gap = -1

	-	G	C	A	T
-	0	-1	-2	-3	-4
G					
A					
T					

Alignment:

-GCAT

How do we find the best alignment given a scoring system?

Global alignment: Needleman-Wunsch algorithm

Example: align GCAT and GAT

Scoring: match = 1, mismatch = -1, gap = -1

	-	G	C	A	T
-	0	-1	-2	-3	-4
G	-1				
A					
T					

Alignment:

--
-G

How do we find the best alignment given a scoring system?

Global alignment: Needleman-Wunsch algorithm

Example: align GCAT and GAT

Scoring: match = 1, mismatch = -1, gap = -1

	-	G	C	A	T
-	0	-1	-2	-3	-4
G	-1				
A	-2				
T	-3				

Alignment:

-GAT

How do we find the best alignment given a scoring system?

Global alignment: Needleman-Wunsch algorithm

Example: align GCAT and GAT

Scoring: match = 1, mismatch = -1, gap = -1

	-	G	C	A	T
-	0	-1	-2	-3	-4
G	-1	?			
A	-2				
T	-3				

How do we find the best alignment given a scoring system?

Global alignment: Needleman-Wunsch algorithm

Example: align GCAT and GAT

Scoring: match = 1, mismatch = -1, gap = -1

	-	G	C	A	T
-	0	-1	-2	-3	-4
G	-1	-2			
A	-2				
T	-3				

Alignment:

-G-

--G

How do we find the best alignment given a scoring system?

Global alignment: Needleman-Wunsch algorithm

Example: align GCAT and GAT

Scoring: match = 1, mismatch = -1, gap = -1

	-	G	C	A	T
-	0	-1	-2	-3	-4
G	-1	-2			
A	-2				
T	-3				

Alignment:

--G

-G-

How do we find the best alignment given a scoring system?

Global alignment: Needleman-Wunsch algorithm

Example: align GCAT and GAT

Scoring: match = 1, mismatch = -1, gap = -1

	-	G	C	A	T
-	0	-1	-2	-3	-4
G	-1	1			
A	-2				
T	-3				

Alignment:

-G

-G

How do we find the best alignment given a scoring system?

Global alignment: Needleman-Wunsch algorithm

Example: align GCAT and GAT

Scoring: match = 1, mismatch = -1, gap = -1

	-	G	C	A	T
-	0	-1	-2	-3	-4
G	-1	1	0		
A	-2				
T	-3				

Alignment:

-GC

-G-

How do we find the best alignment given a scoring system?

Global alignment: Needleman-Wunsch algorithm

Example: align GCAT and GAT

Scoring: match = 1, mismatch = -1, gap = -1

	-	G	C	A	T
-	0	-1	-2	-3	-4
G	-1	1	0		
A	-2	0			
T	-3				

Alignment:

-G-

-GA

How do we find the best alignment given a scoring system?

Global alignment: Needleman-Wunsch algorithm

Example: align GCAT and GAT

Scoring: match = 1, mismatch = -1, gap = -1

	-	G	C	A	T
-	0	-1	-2	-3	-4
G	-1	1	0		
A	-2	0	-1		
T	-3				

Alignment:

-GC-

-G-A

How do we find the best alignment given a scoring system?

Global alignment: Needleman-Wunsch algorithm

Example: align GCAT and GAT

Scoring: match = 1, mismatch = -1, gap = -1

	-	G	C	A	T
-	0	-1	-2	-3	-4
G	-1	1	0		
A	-2	0	-1		
T	-3				

Alignment:

-G-C

-GA-

How do we find the best alignment given a scoring system?

Global alignment: Needleman-Wunsch algorithm

Example: align GCAT and GAT

Scoring: match = 1, mismatch = -1, gap = -1

	-	G	C	A	T
-	0	-1	-2	-3	-4
G	-1	1	0		
A	-2	0	0		
T	-3				

Alignment:

-GC

-GA

How do we find the best alignment given a scoring system?

Global alignment: Needleman-Wunsch algorithm

Example: align GCAT and GAT

Scoring: match = 1, mismatch = -1, gap = -1

	-	G	C	A	T
-	0	-1	-2	-3	-4
G	-1	1	0	-1	-2
A	-2	0	0	1	0
T	-3	-1	-1	0	2

How do we find the best alignment given a scoring system?

Global alignment: Needleman-Wunsch algorithm

Example: align GCAT and GAT

Scoring: match = 1, mismatch = -1, gap = -1

	-	G	C	A	T
-	0	-1	-2	-3	-4
G	-1	1	0	-1	-2
A	-2	0	0	1	0
T	-3	-1	-1	0	2

Alignment:

-GCAT

-G-A-T

Needleman-Wunsch algorithm, mathematical form

$$\mathbf{M}(0, j) = j \times p \quad \text{first row, } p = \text{gap penalty}$$

$$\mathbf{M}(i, 0) = i \times p \quad \text{first column}$$

$$\mathbf{M}(i, j) = \max \left(\begin{array}{l} \mathbf{M}(i-1, j) + p \\ \mathbf{M}(i, j-1) + p \\ \mathbf{M}(i-1, j-1) + s(a_j, b_i) \end{array} \right) \begin{array}{l} \text{top} \\ \text{left} \\ \text{diagonal} \end{array}$$

$s(a_j, b_i)$ = match/mismatch score for sites j and i
in sequences a and b

Now try on your own

Align ATGCT and ATTACA

Scoring: match = 1, mismatch = -1, gap = -1

	-	A	T	T	A	C	A
-							
A							
T							
G							
C							
T							

Multiple sequence alignment (MSA)

B9SI54 | B9SI54_RICCO_263_570
 Q01I60 | Q01I60_ORYSA_160_476
 C5Y8S2 | C5Y8S2_SORBI_153_466
 B4FRR6 | B4FRR6_MAIZE_154_469
 D7U4G4 | D7U4G4_VITVI_82_394
 D7M270 | D7M270_ARALY_263_574
 Q8L7Q7 | PME64_ARATH_283_601
 D8QSM2 | D8QSM2_SELML_242_541
 A9TZ89 | A9TZ89_PHYPA_262_575
 D8SH72 | D8SH72_SELML_209_529

```

-----DAVVAAD-----GSGQFKTIGEALNSYKLNK--GWYVIYVKAGVYNEHFVIFIS
--TLRAHATVCNAPSPATTQRCDYSTVQAAIDAAPNHTA--GHFVIKVAAGIYKENVVVIP
---IRPDATVCK--PNSGAEP CGYSTVQAAVDAAPNYTA--GHFVIAVAAGTYKENIVVIP
---IRPDATVCK--PNSGVKPCGYSTVQAAVDAAPNHTAGAGHFVIAVAGAGTYKENVVVIP
--SPQPNATVCKG-----GDGCKYKTVQEAUNAAPDNDS--SRKFFVIRIQEGVYEEETVRVP
-SGLKEDVTVCKD-----GKCGYKTVQDAVNAAPEDNG--MRKFFVIRISEGVYEEENVIVP
-SGLTEDVTVCKN----GGKDKYKTVQEAUNASAPDTNR--TVKFFVIRIREGVYEEETVRVP
-----SVV-----VGKSGSFKTIQEAIDSAPSNSK--ERFSIYIQEGYDERIYVS
----SPSVTVDI-----YSAFSSIQRAVDLAPDWST--QRYVIYIKTGVYNEVVRIP
ASLISPSAIVSRT--PDQPQLTIFTSIQAAVDHAPNHCT--ARYVIYIKAGVYAENVVRIP
      .                : : :   * : :           : * :   * * * : :
  
```

B9SI54 | B9SI54_RICCO_263_570
 Q01I60 | Q01I60_ORYSA_160_476
 C5Y8S2 | C5Y8S2_SORBI_153_466
 B4FRR6 | B4FRR6_MAIZE_154_469
 D7U4G4 | D7U4G4_VITVI_82_394
 D7M270 | D7M270_ARALY_263_574
 Q8L7Q7 | PME64_ARATH_283_601
 D8QSM2 | D8QSM2_SELML_242_541
 A9TZ89 | A9TZ89_PHYPA_262_575
 D8SH72 | D8SH72_SELML_209_529

```

RILTNVYMYGDGIDRTIISGSKHTM-DGLPAYRTATVAVLGDGFVCKSMTIQNSATSD-K
YEKTNILLVGDGIGATVITASRSVGDIGTYETATVAVIGDGFRAKDIFFENGAGAGAH
YEKTNILLMGEGMGATVITASRSVGDIGLTHEATVAVIGDGFRRARDITFENSAGARAH
YEKANILLMGEGMGATVITASRSVGDIGLGTYEATVVDVIGDGFRRARDITFENSAGAGAH
LEKKNVVFVLDGMGKTIVITGSLNVGQPGISTYNSATVGVAGDGFMAHDLTFQNTAGPDAH
FEKKNVVFVLDGMGKTIVITGSLNAGMPGITTYNTATVGVVGDGFMAHDLTFQNTAGPDAH
FEKKNVVFVLDGMGKTIVITGSLNVGQPGMTTFESATVGVLDGDFMARDLTIENTAGADAH
DSKSMIMLVGAGARKTIIISGNNYVR-EGVTTMDTATVVLVAGDGFVARDLTIIRNTAGPELH
KQKTNLMFLGDGTDKTIITGSLSDSQPGMITWATATVAVSGSGFIARGITFQNTAGPAGR
LQKSMMLFVGDGMDKTIIRGSMVS KGGTTTTFASATLAVNGKGF LARDLTVENTAGPEGH
      : : * *   * : * ..      * :   : * : * * * .   : * . * *   :
  
```

Software to generate MSAs

- MAFFT
(very good, very fast)
<http://mafft.cbrc.jp/alignment/software/>
- Clustal Omega
(very good, very fast)
<http://www.ebi.ac.uk/Tools/msa/clustalo/>
- PRANK
(extremely good, very slow)
<http://wasabiapp.org/software/prank/>