JOHANNES GUTENBERG UNIVERSITÄT MAINZ

Protein structure prediction

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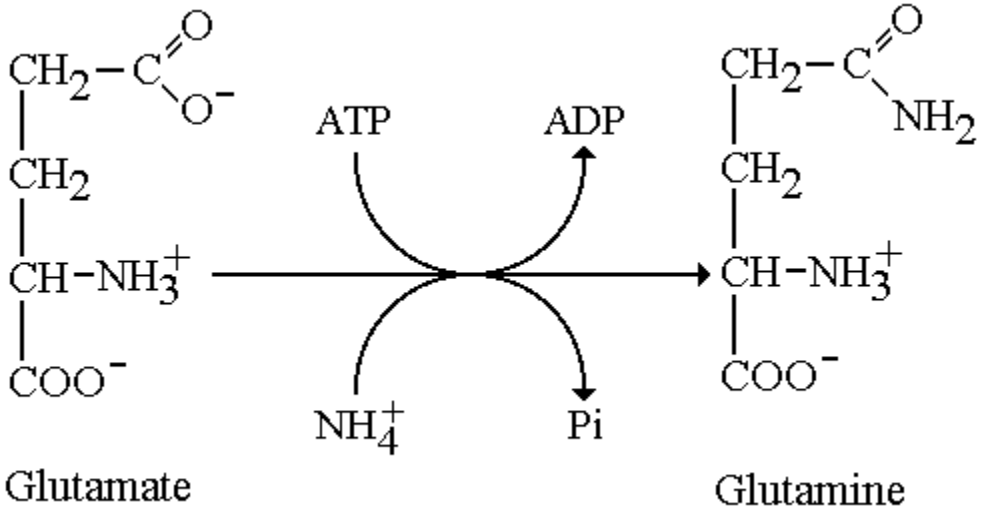
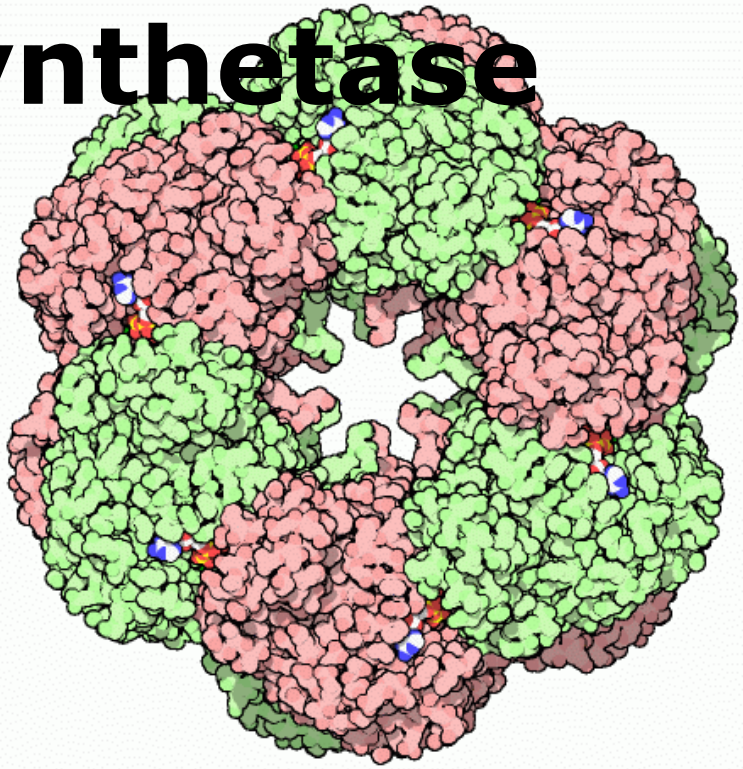
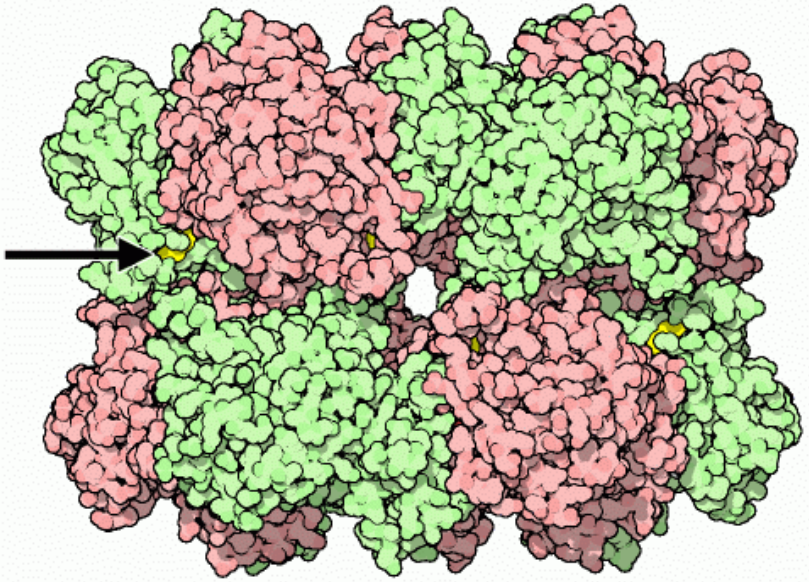
Mount Everest

Age: 60M years

Glutamine synthetase



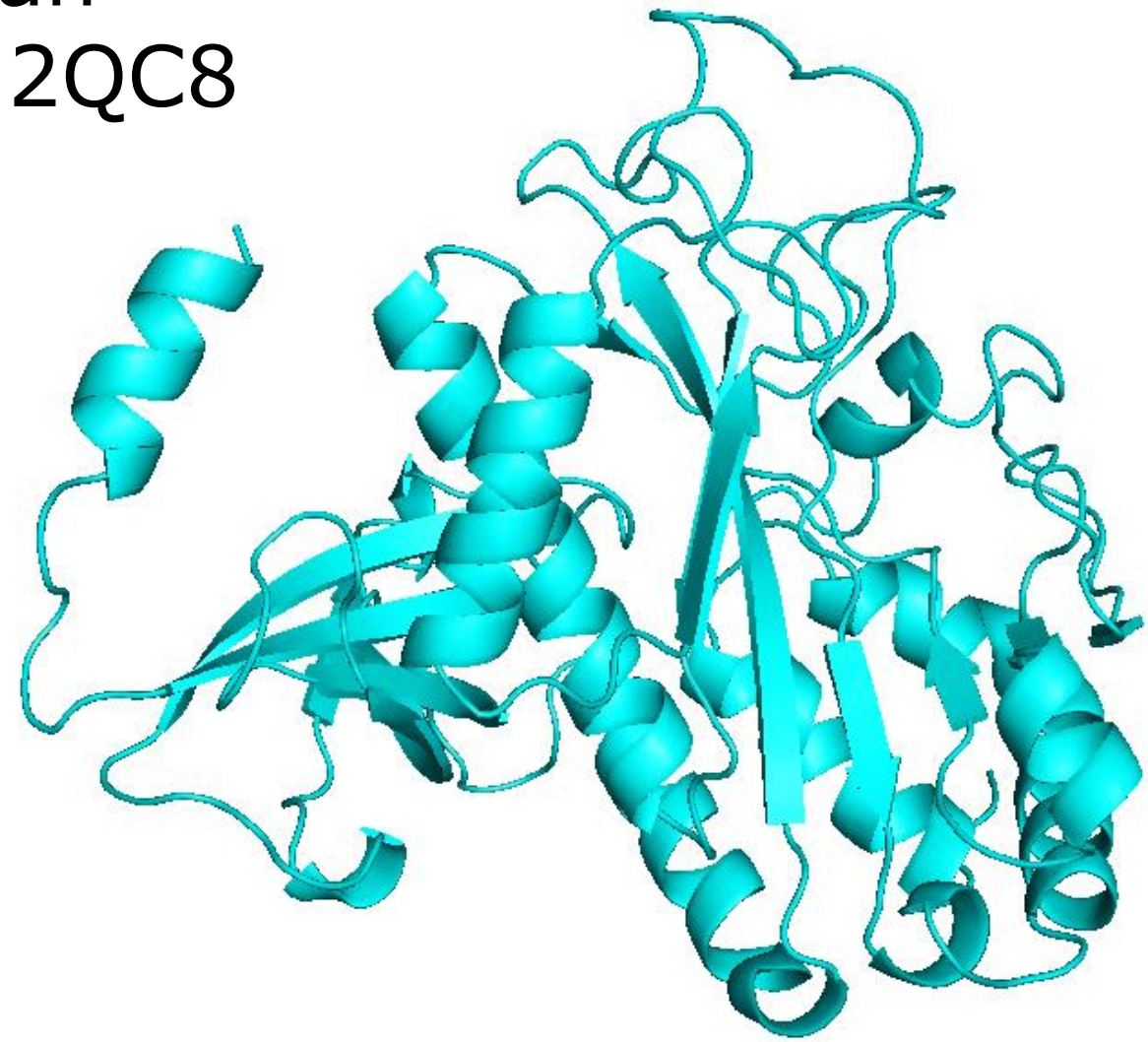




Age: +3500M years

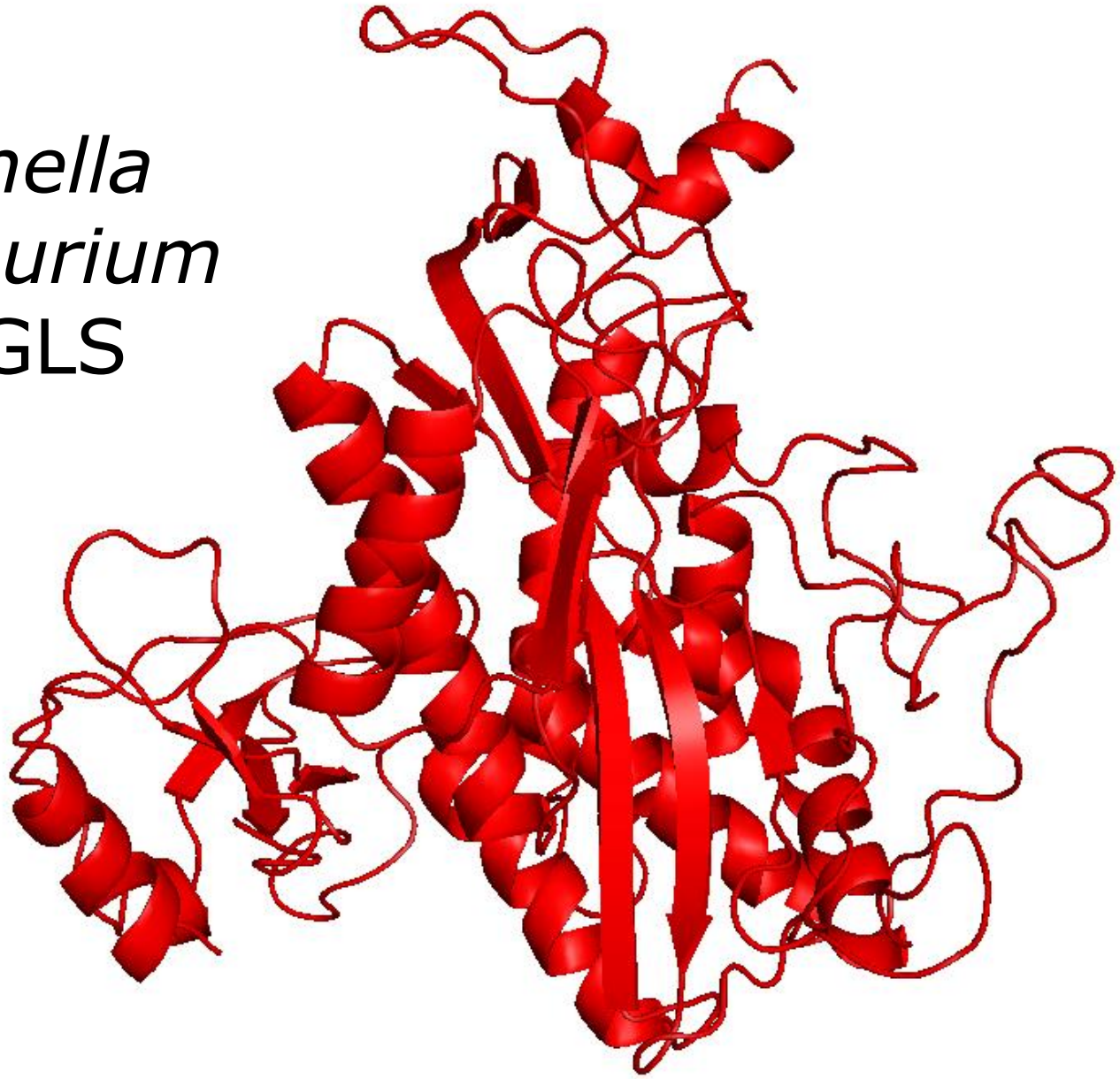
Glutamine synthetase

Human
PDB:2QC8

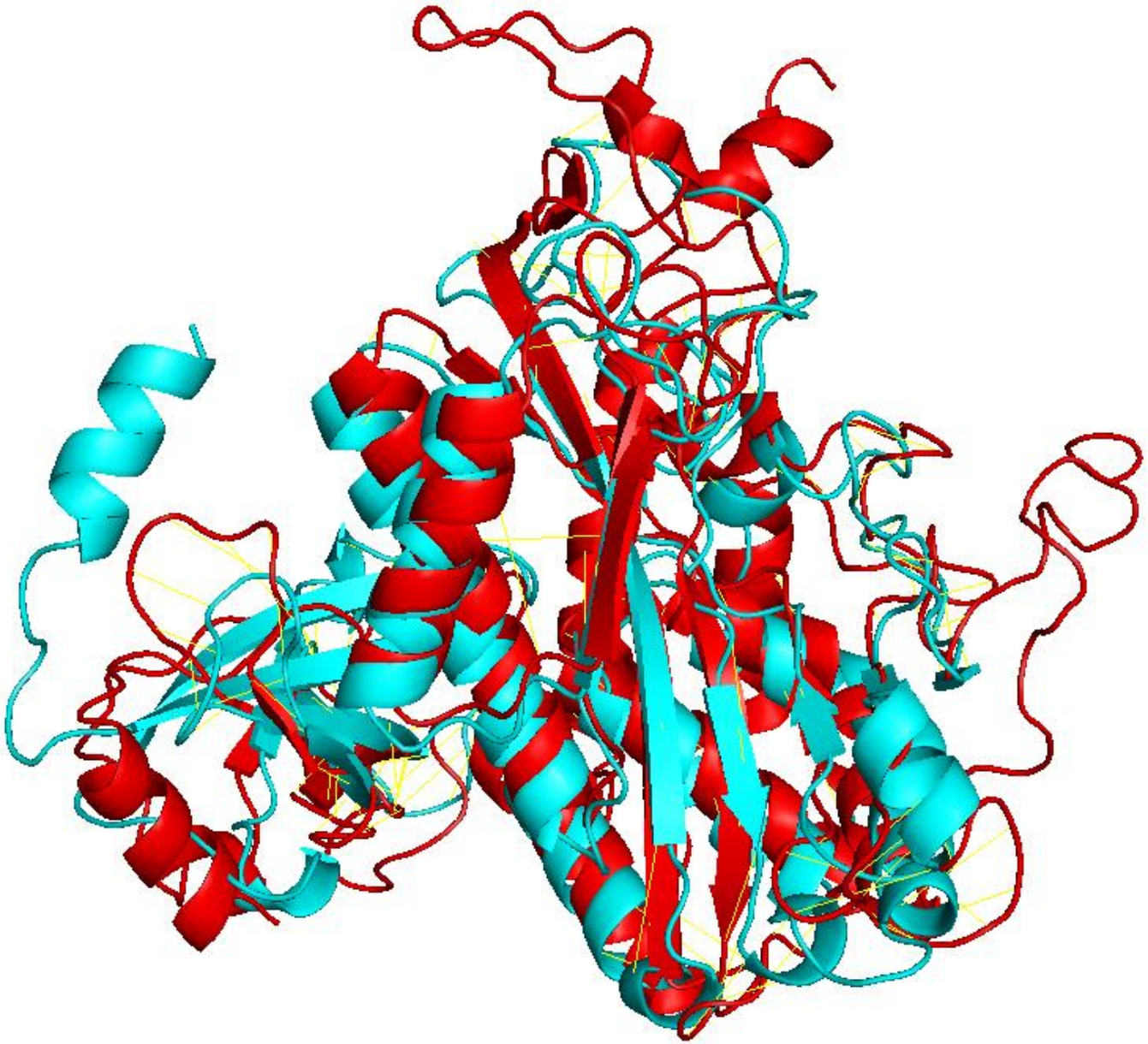


Glutamine synthetase

Salmonella typhimurium
PDB:2GLS



Glutamine synthetase



Time line

Earth: 4.6 By

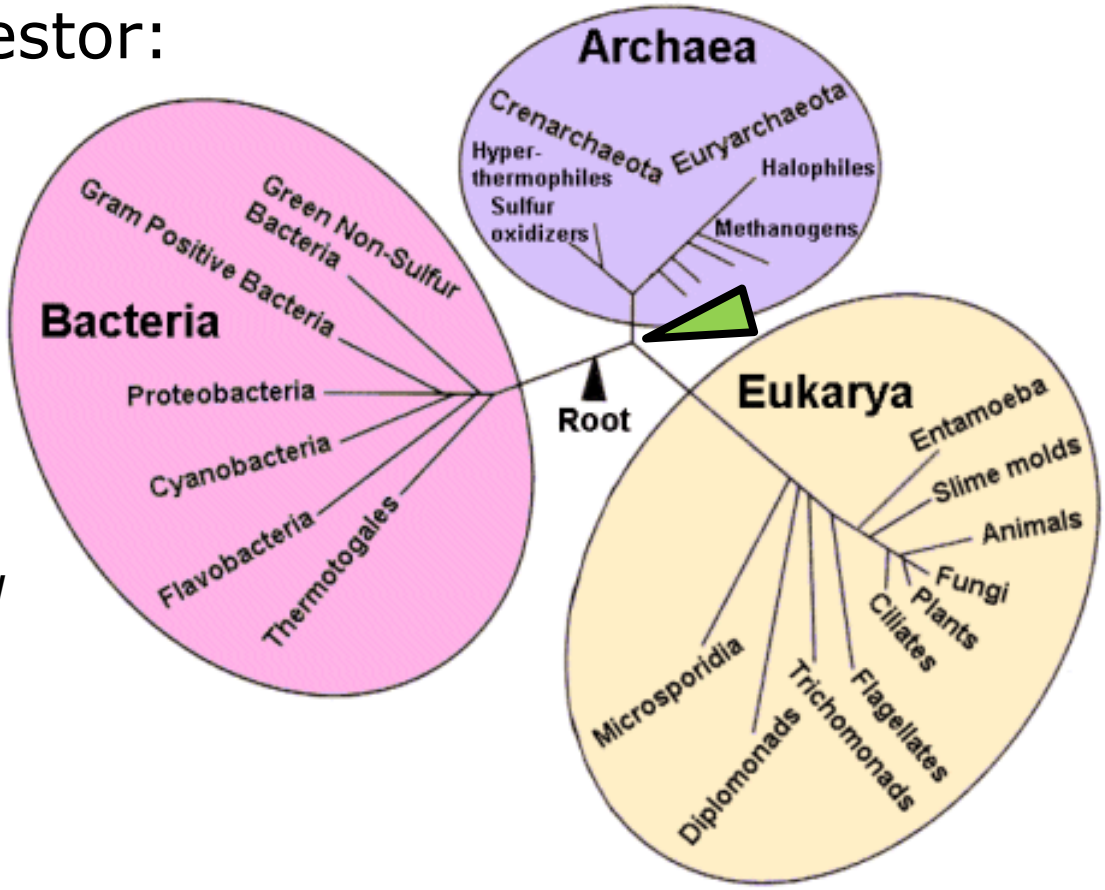
Origin of life: 3.9 By – 3.5 By

Last Common Ancestor:
3.5 – 3.8 By

Glansdorff & Labedan (2008) *Biology Direct*

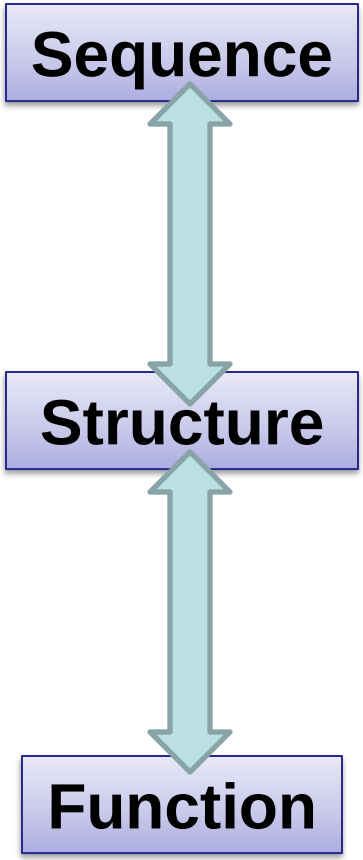
4.29 By

Sheridan *et al.* (2003) *Geomicrobiology Journal*



Sequence and function

Evolutionary constraints



MTQDELKKAVGWAALQYVQ

PG DA

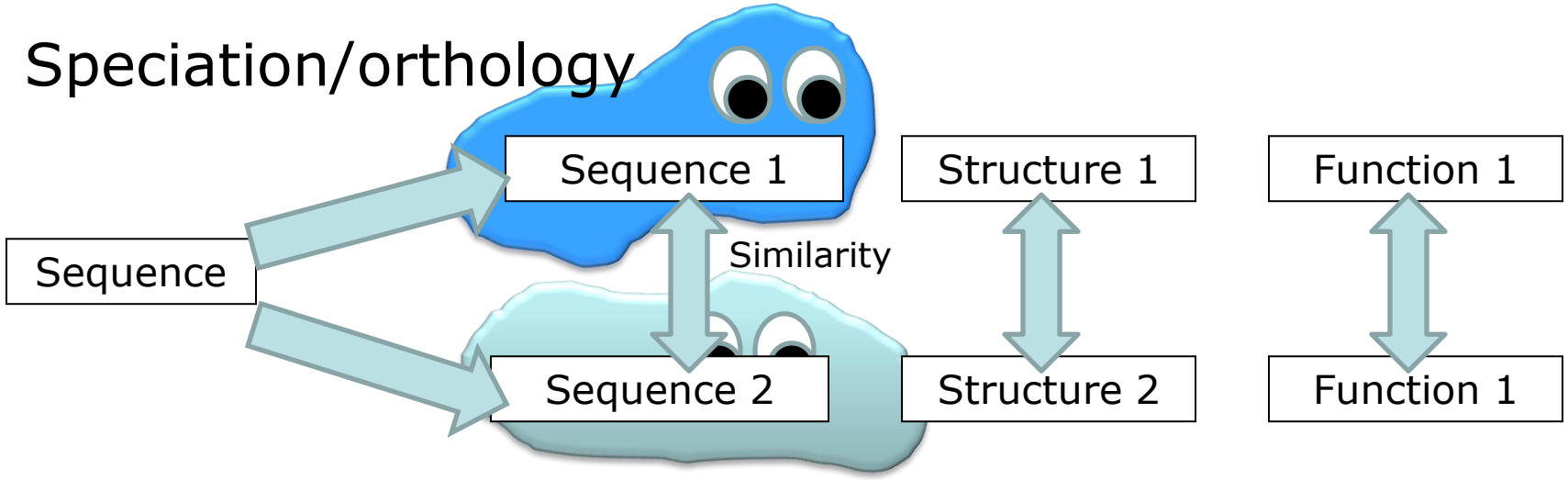
LG ST

EK

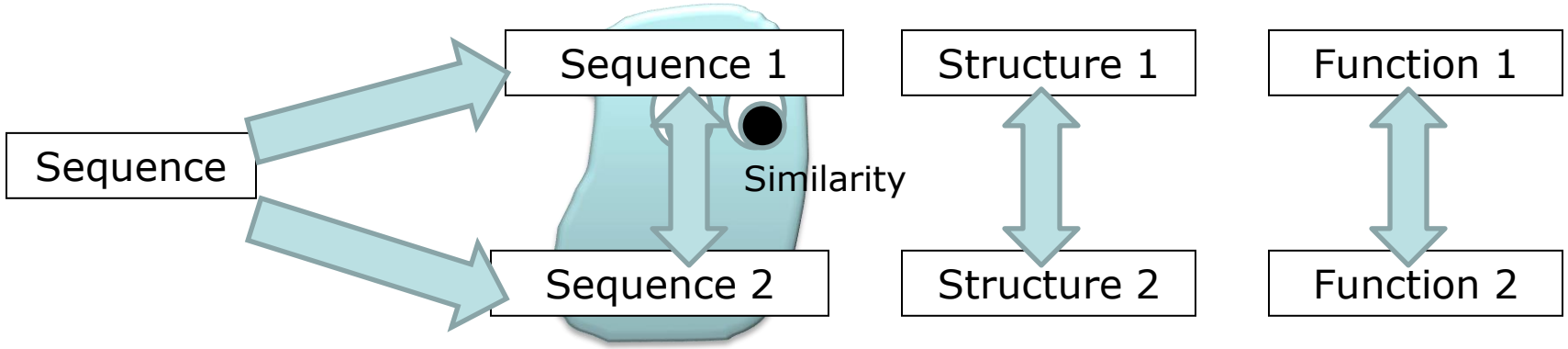
Sequence and function

Evolutionary constraints

Speciation/orthology



Gene duplication/paralogy



Sequence pairwise alignment

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GTDGHPFGWPSNGFPGPQGPYYCGVGADRAYGRDIVEAHYRACLYAGVKIAGTNAEVMPAQWEFQIGPCE
GISMGDHLWVARFILHRVCEDFGVIATFDPKPIPGNWNGAGCHTNFSTKAMREENGLKYIEEAIEKLSKR
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>gs_salmonella gi|16767272|ref|NP_462887.1| glutamine synthetase [Salmonella enterica subsp. enterica serovar Typhimurium str. LT2]
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Sequence pairwise alignment

BLAST (Altschul et al, 1990)

```
>lcl|39919 unnamed protein product
Length=469

 Score = 70.5 bits (171),  Expect = 1e-17, Method: Compositional matrix adjust.
 Identities = 102/363 (28%), Positives = 138/363 (38%), Gaps = 96/363 (26%)

Query  62   FDGSSTLQSEGSN-SDMYLVPAA--MFRDPFRKDPNKLVLCEVFK------YNRRP----  108
            FDGSS    +G N SDM L+P A     DPF  D   ++ C++ +      Y+R P
Sbjct  50   FDGSSIGGWKGINESDMVLMPDASTAVIDPFFADSTLIIRCDILEPGTLQGYDRDPRSIA  109

Query  109  --AETNLRHTCKRIMDMVSNQHPWFGMEQEYTLMGTDGHPFGWPSNGF-----------  154
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Sbjct  110  KRAEDYLRATG--IADTV-----LFGPEPEFFLF--DDIRFGASISGSHVAIDDIEGAWN  160

Query  155  ------------PGPQGPYYCGVGADRAYGRDI-------------VEAHYRACLYAG  187
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Sbjct  161  SSTKYEGGNKGHRPGVKGGYFPVPPVDSA--QDIRSEMCLVMEQMGLVVEAHHHEVATAG  218

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Query  247  WNGAGCHTNFSTKAMREENGLKYIEEAIEKLSKRHQYHIRAYDPKGGLDNA---------  297
              +G  CH + +       +G KY       LS++  Y+I           NA
Sbjct  266  GSGMHCHMSLAKNGTNLFSGDKY-----AGLSEQALYYIGGVIKHAKAINALANPTTNSY  320

Query  298  RRLTGFHETSNINDFSAGVANRSASIRIPRTVGQEKKGYFEDRRPSANCDPFSVTEALIR  357
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Sbjct  321  KRLVPGYEAPVMLAYSA--RNRSASIRIP-VVASPKARRIEVRFPDPAANPYLCFAALLM  377

Query  358  TCL  360
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Sbjct  378  AGL  380
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Multiple sequence alignment

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>gs_human gi|74271837|ref|NP_001028216.1| glutamine synthetase [Homo sapiens]
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>gs_salmonella gi|16767272|ref|NP_462887.1| glutamine synthetase [Salmonella
enterica subsp. enterica serovar Typhimurium str. LT2]
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DDVYGWPKGGYPAPQGPYYCGVGAGKVYARDMIEAHYRACLYAGLEISGINAEVMPSQWEFQVGPCTGID
MGDQLWMARYFLHRVAEEFGIKISFHPKPLKGDWNGAGCHTNVSTKEMRQPGGMKYIEQAIEKLSKRHAE
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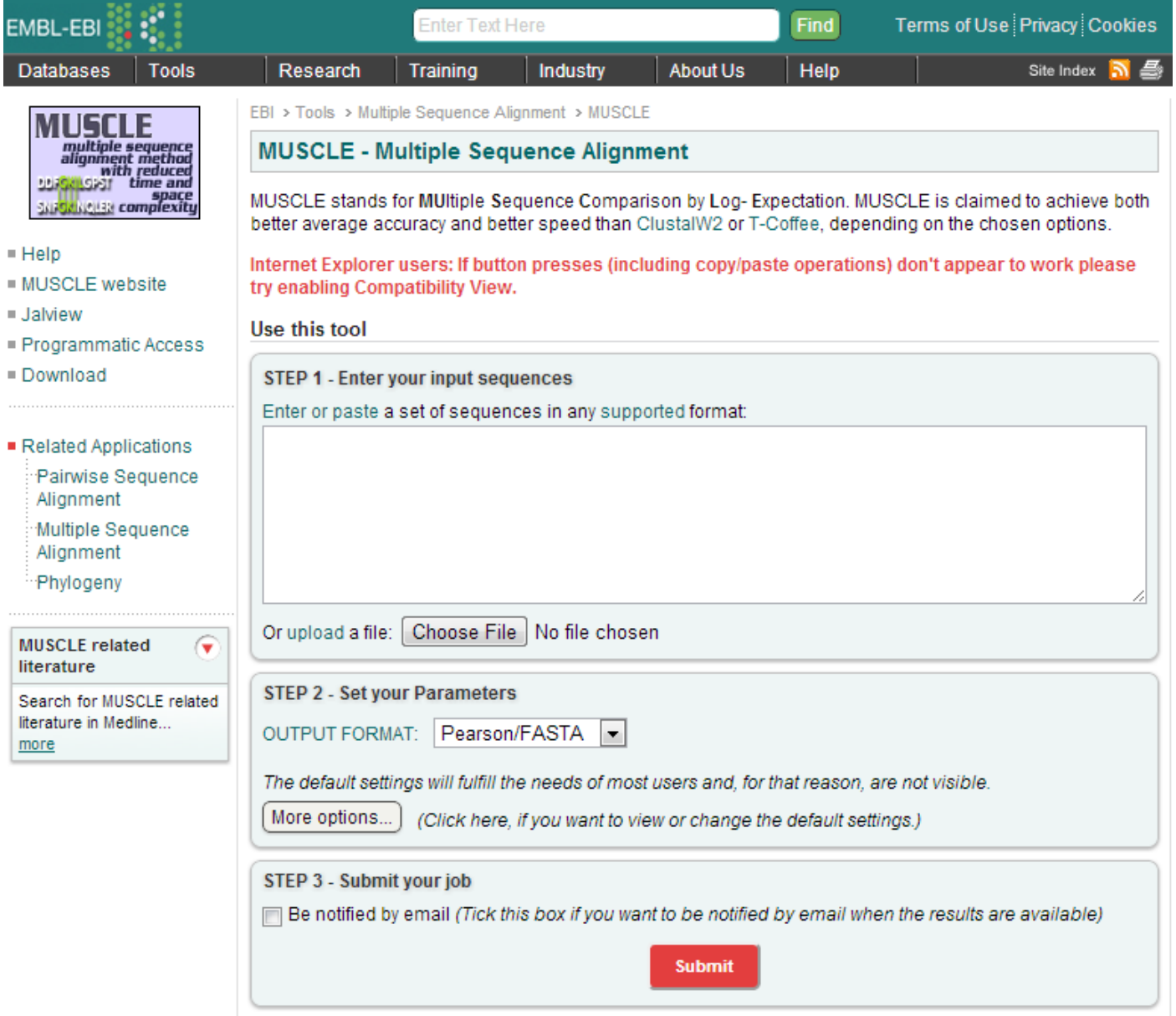

http://www.ebi.ac.uk/Tools/msa/muscle/

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>gs_yeast gi|330443748|ref|NP_015360.2| Gln1p [Saccharomyces cerevisiae S288c]
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KNLF-QSSTGKITEECGYAISAILSNARALSSFVAPLVNSYKRLVPHYEAPTRIVW--GY
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>gs_salmonella gi|16767272|ref|NP_462887.1| glutamine synthetase [Salmonella enterica
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TRFNTMTKKADEIQIYKYVVHNVAHRFGKTATFMPKPMFGD-NGSGMHCHMS---LAKNG
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RNRSASIRIPV-VASPKARRIEVRFPDPAANPYLCFAALLMAGLDGIKNKIHPGEAMDKN
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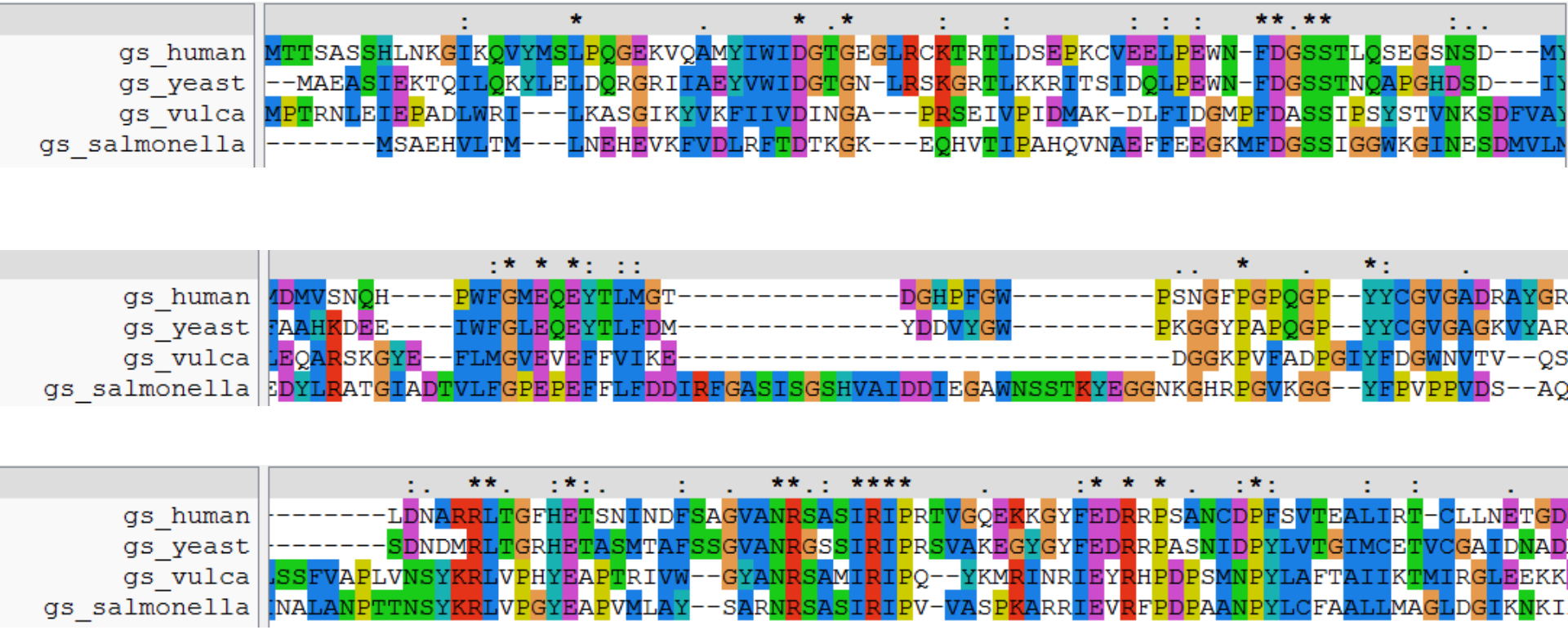

ClustalW, JalView

Determination of protein structure

X-ray crystallography (179K in PDB)

- need crystals

Nuclear Magnetic Resonance (NMR) (14K)

- proteins in solution
- lower size limit (600 aa)

Electron microscopy (17K)

- Recently resolution improving a lot!

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X-ray crystallography (179K in PDB)

- need crystals

Nuclear Magnetic
(14K)

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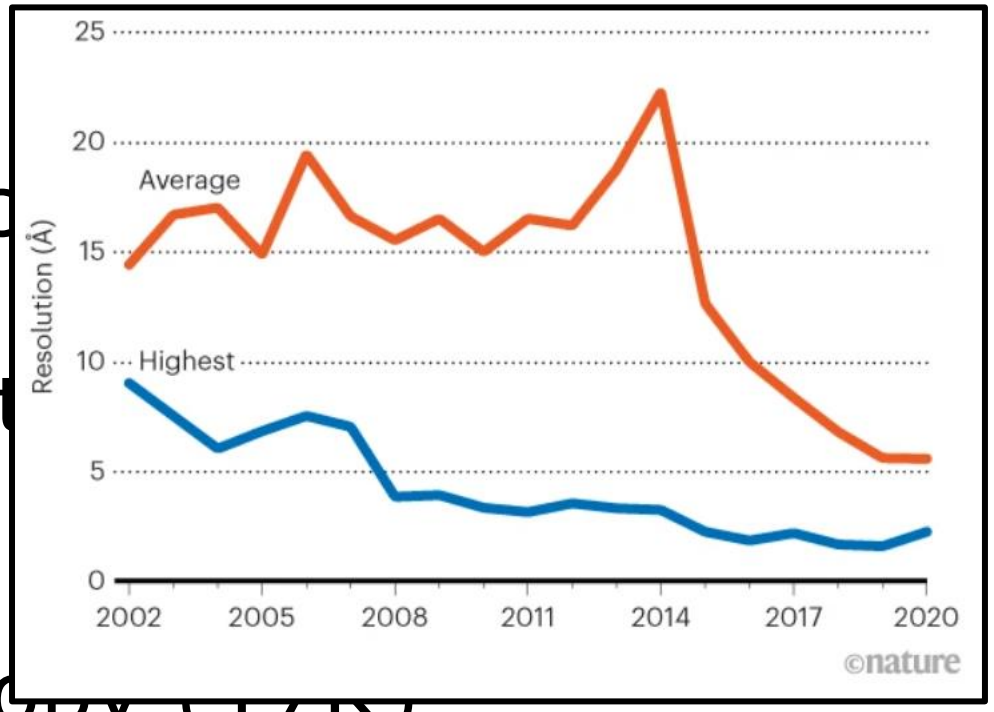
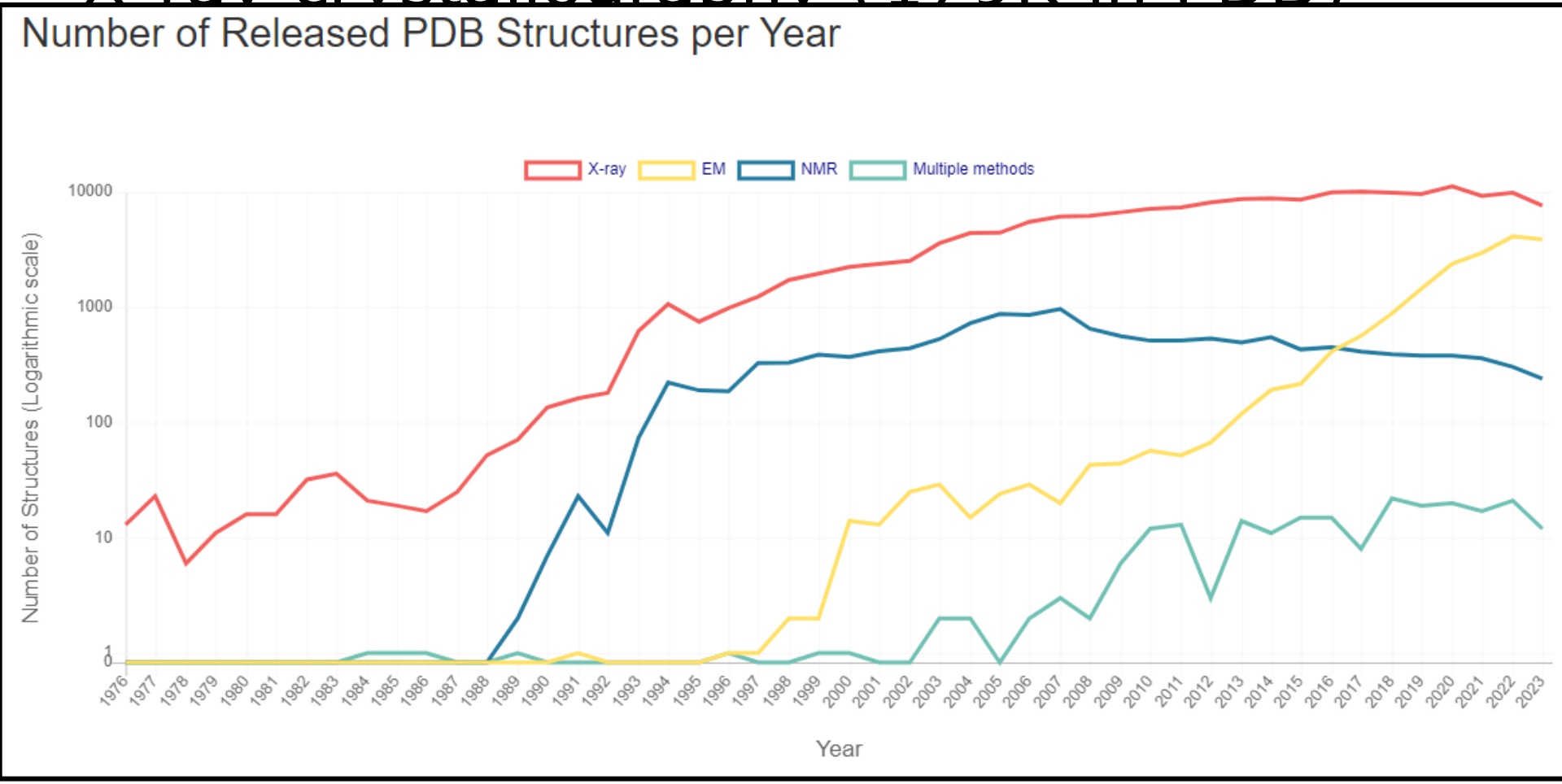
- Recently resolution improving a lot!

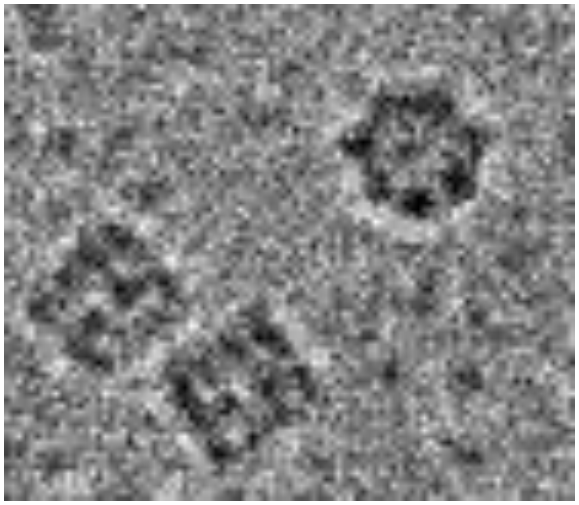


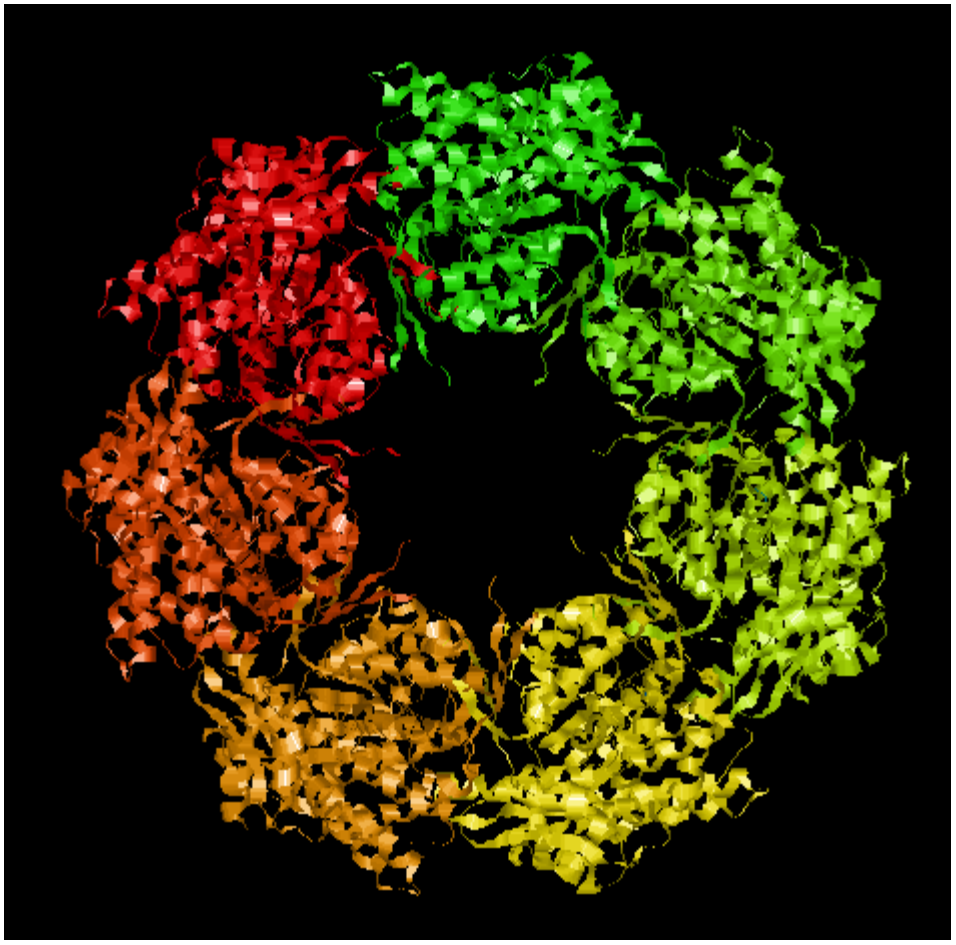
Determination of protein structure

X-ray crystallography (179K in PDB)



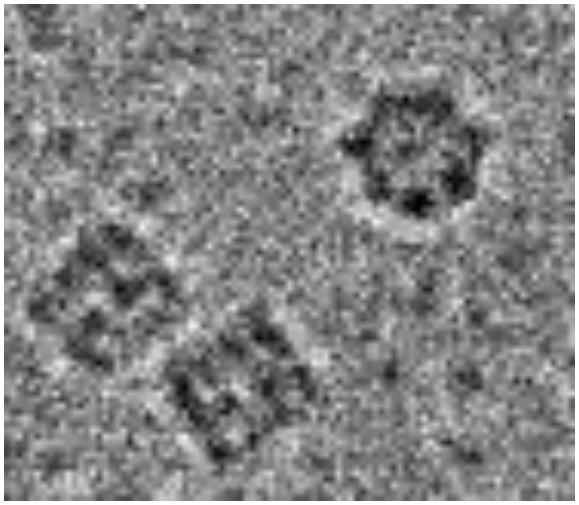
Determination of protein structure

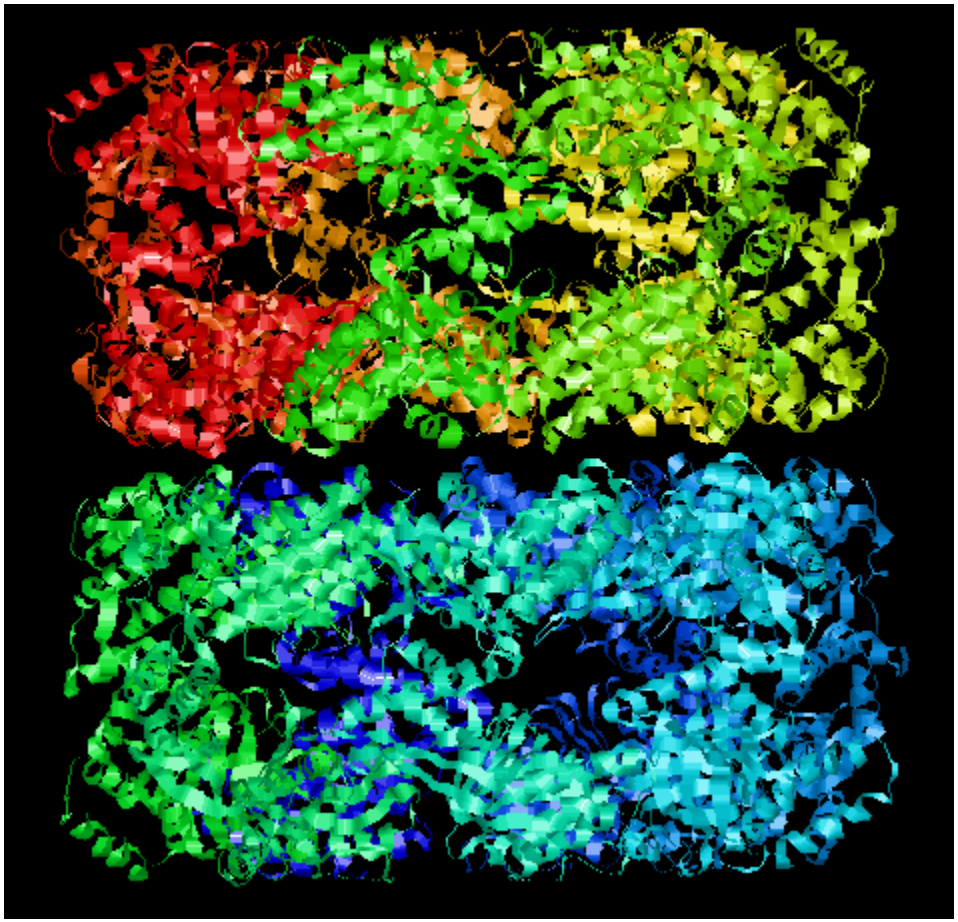




resolution 2.4 A

Determination of protein structure



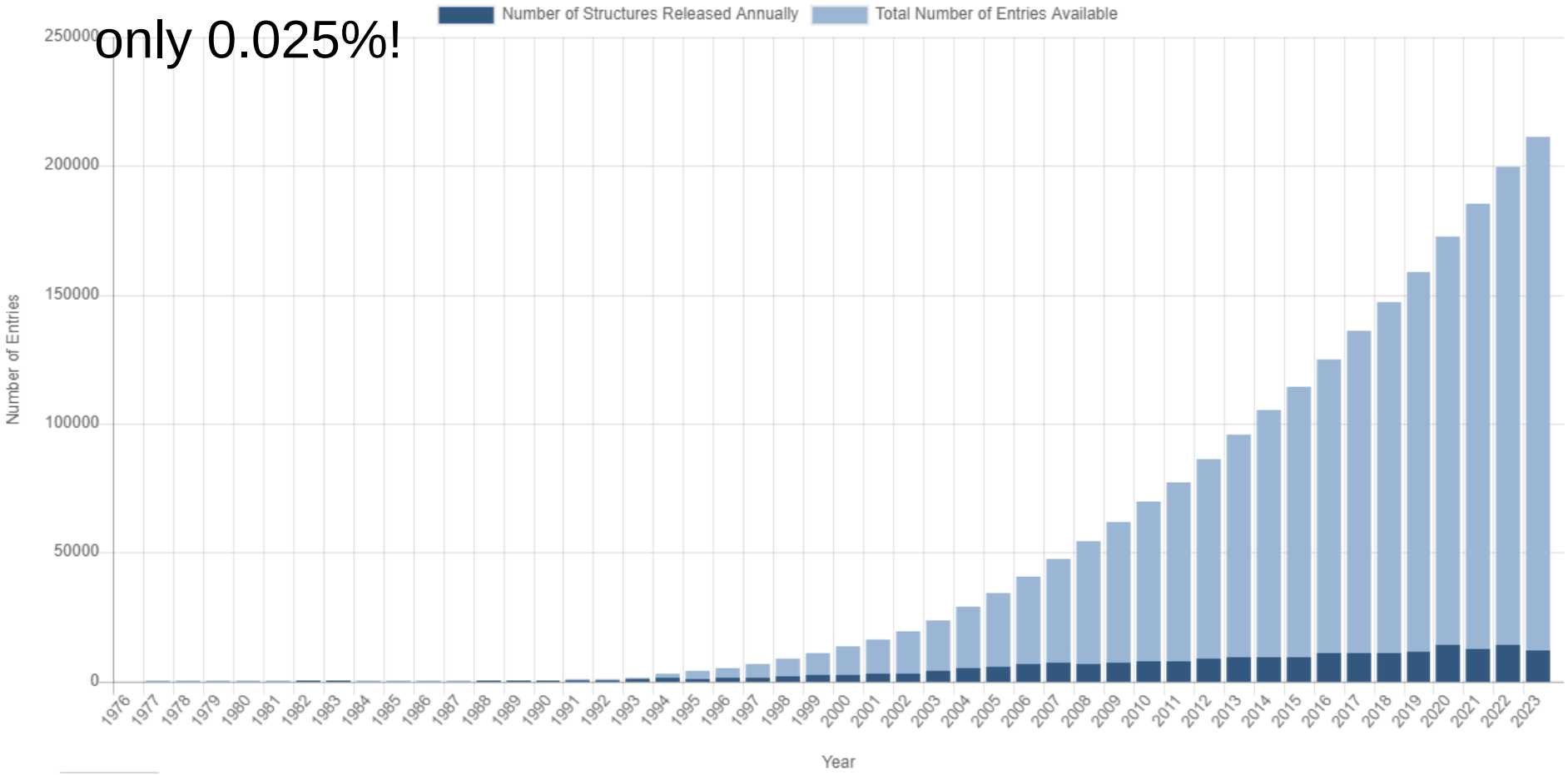


resolution 2.4 A

Structural genomics

Currently: 211K protein 3D structures
from around 62K sequences in UniProt (how do I know?)
252M sequences in UniProt

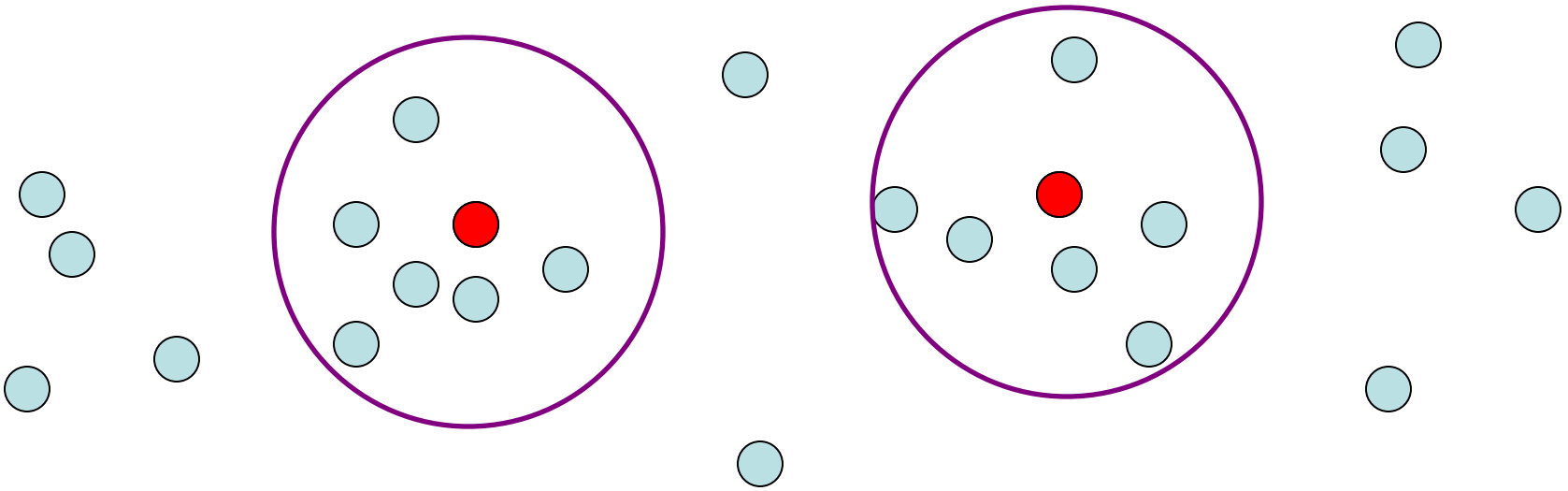
only 0.025%!



Structural genomics

Currently: 187K protein 3D structures
from around 55K sequences in UniProt (how do I know?)
226M sequences in UniProt

only 0.024%!



50% sequences covered (25% in 1995)

Protein structure prediction
Ab initio

Explore conformational space

Limit the number of atoms

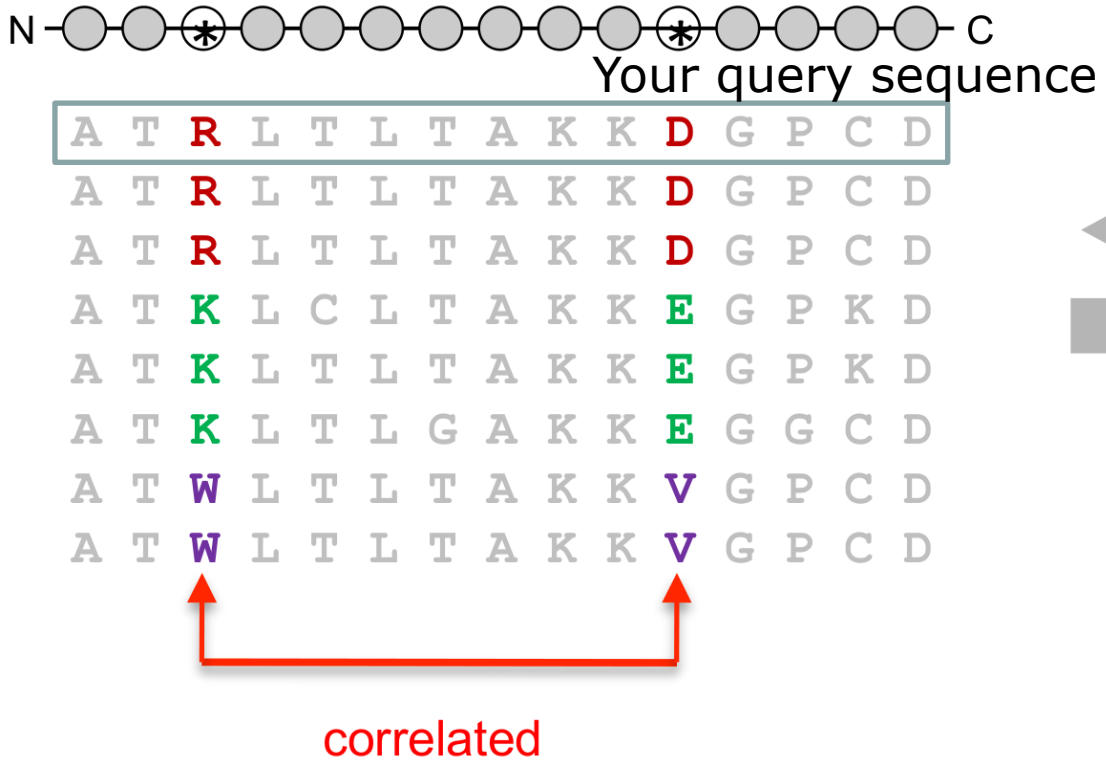
Break the problem into fragments of sequence

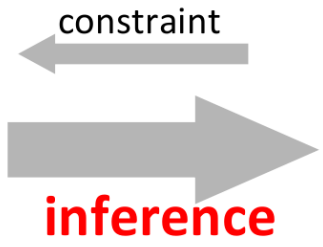
Optimize hydrophobic residue burial and pairing of beta-strands

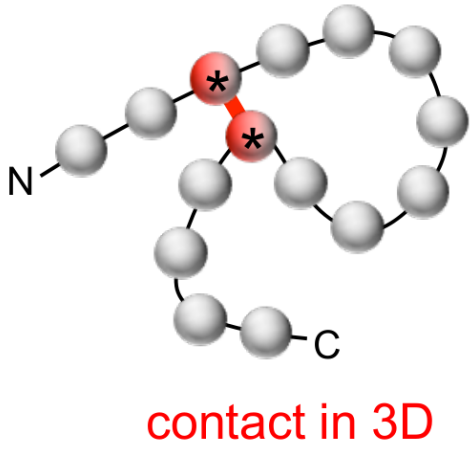
Limited success…

Protein structure prediction

Correlated mutations







https://commons.wikimedia.org/wiki/File:Correlated_mutation.png

Protein structure prediction
Combined

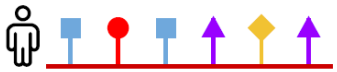
Homology to solved structures

Correlated sequence variation in homologs

Generation of a structure following physical constraints

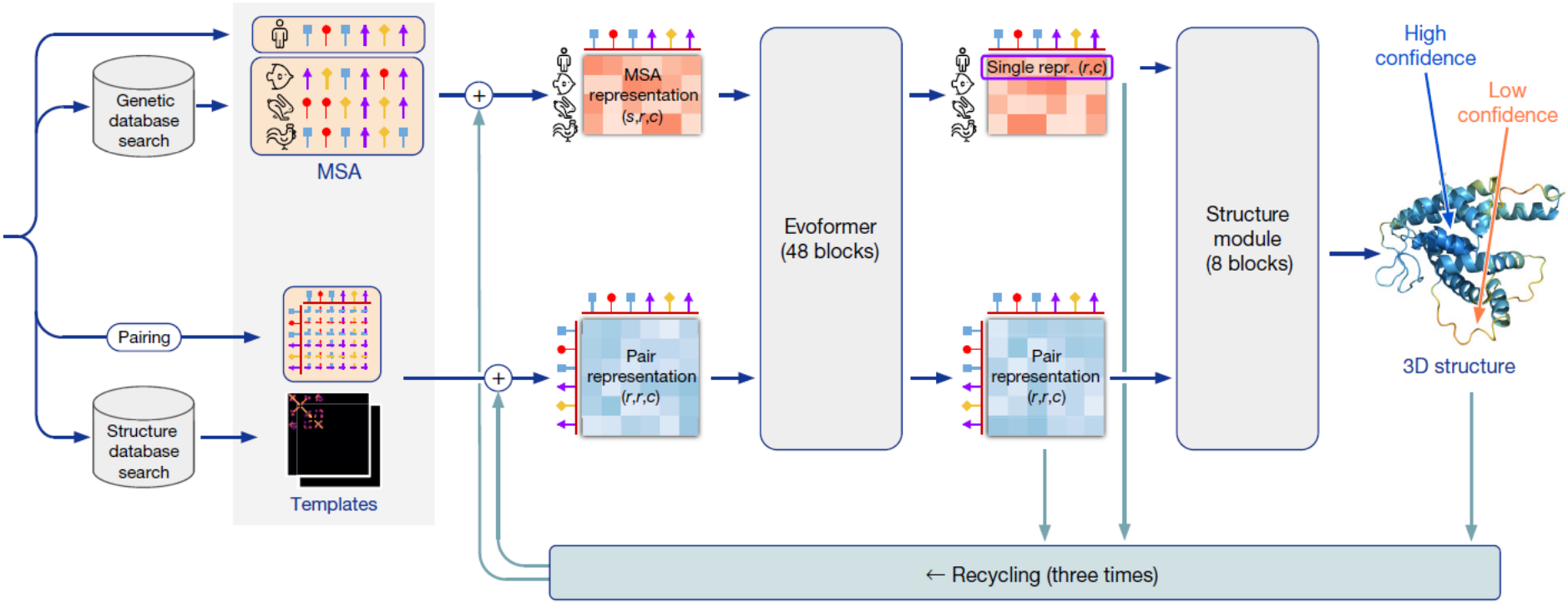
Protein structure prediction

AlphaFold: DeepMind, Google



Input sequence

Demis Hassabis



Jumper et al (2021) Nature

Protein structure prediction

AlphaFold: DeepMind, Google

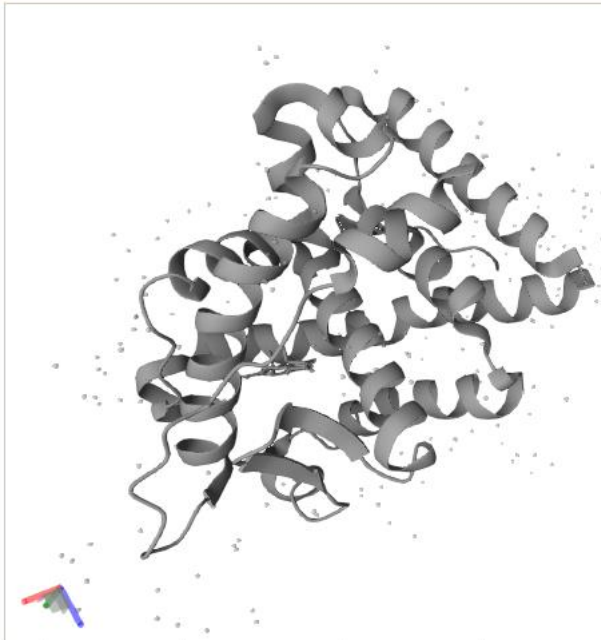
UniProtKB - P08235 (MCR_HUMAN)

Display | Help video | BLAST | Align | Format | Add to basket | History

- Entry
- Publications
- Feature viewer
- Feature table

Protein: **Mineralocorticoid receptor**
Gene: **NR3C2**
Organism: *Homo sapiens (H*
Status: Reviewed - A

Structure



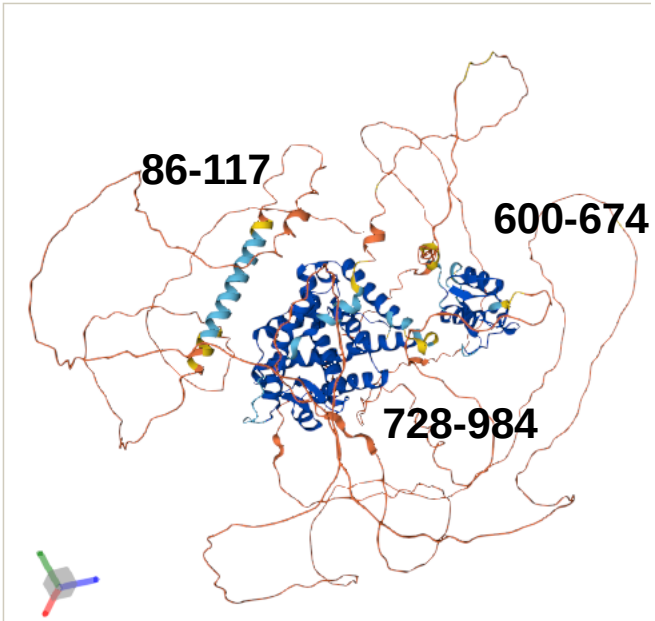
PDB	1Y9R	X-ray	1.96 Å	A/B	731-984

Structure

Model Confidence:

- Very high (pLDDT > 90)
- Confident (90 > pLDDT > 70)
- Low (70 > pLDDT > 50)
- Very low (pLDDT < 50)

AlphaFold produces a per-residue confidence score (pLDDT) between 0 and 100. Some regions with low pLDDT may be unstructured in isolation.



AlphaFold	AF-P08235-F1	Predicted			1-984

Protein structure prediction

AlphaFold: DeepMind, Google

Precomputed models:
UniProt
https://alphafold.ebi.ac.uk/
(limited to model organisms)

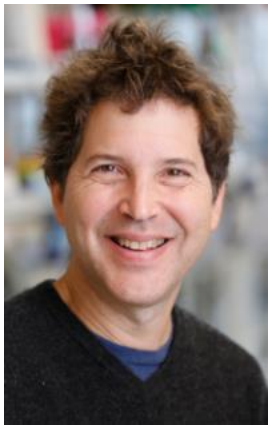
Colab notebook (simplified version / limited server / takes hours)

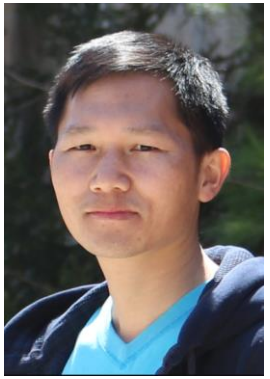
Source code (Needs 3 Tb disk space)

Protein structure prediction

trRosetta: David Baker & Jianyi Yang

Needs large multiple sequence alignments to predict contacts





Predictions available for all PFAM domains
Example:
https://www.ebi.ac.uk/interpro/entry/pfam/PF07887/rosettafold/

Run online at
https://yanglab.nankai.edu.cn/trRosetta/

Du et al (2021) *Nature Protocols*

Protein structure prediction

C-I-Tasser: Yang Zhang

Run online at
https://zhanggroup.org/C-I-TASSER/

Zheng et al (2021) *Cell Reports Methods*