Repliement des protéines

avec la révolution d'AlphaFold2

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BFA, Université Paris Cité

octobre 2024

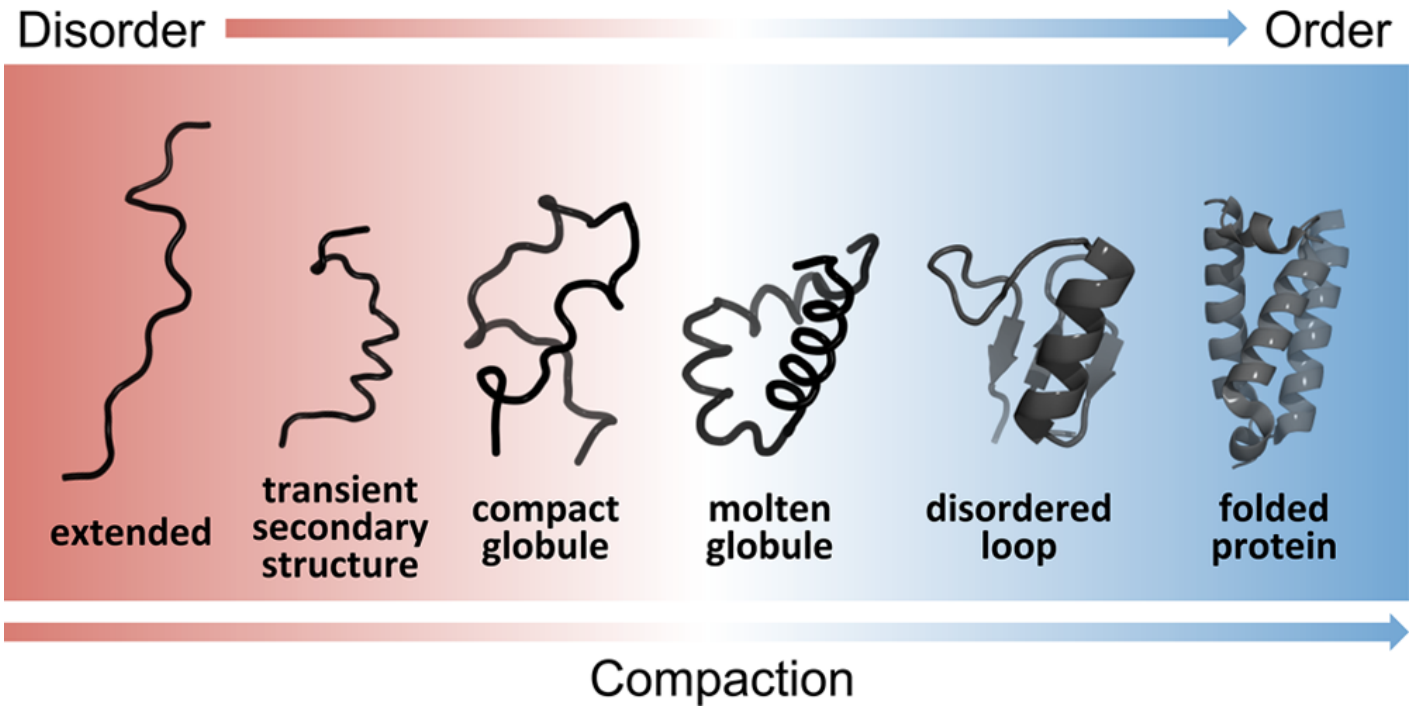
Plan

1. Processus du repliement
2. Techniques expérimentales
3. Repliement par dynamique moléculaire
4. Repliement in vivo
5. Protein design
6. Alpha Fold
7. Bibliographie

1. Processus du repliement
   - Processus du repliement
   - État déplié

Processus du repliement

Repliement



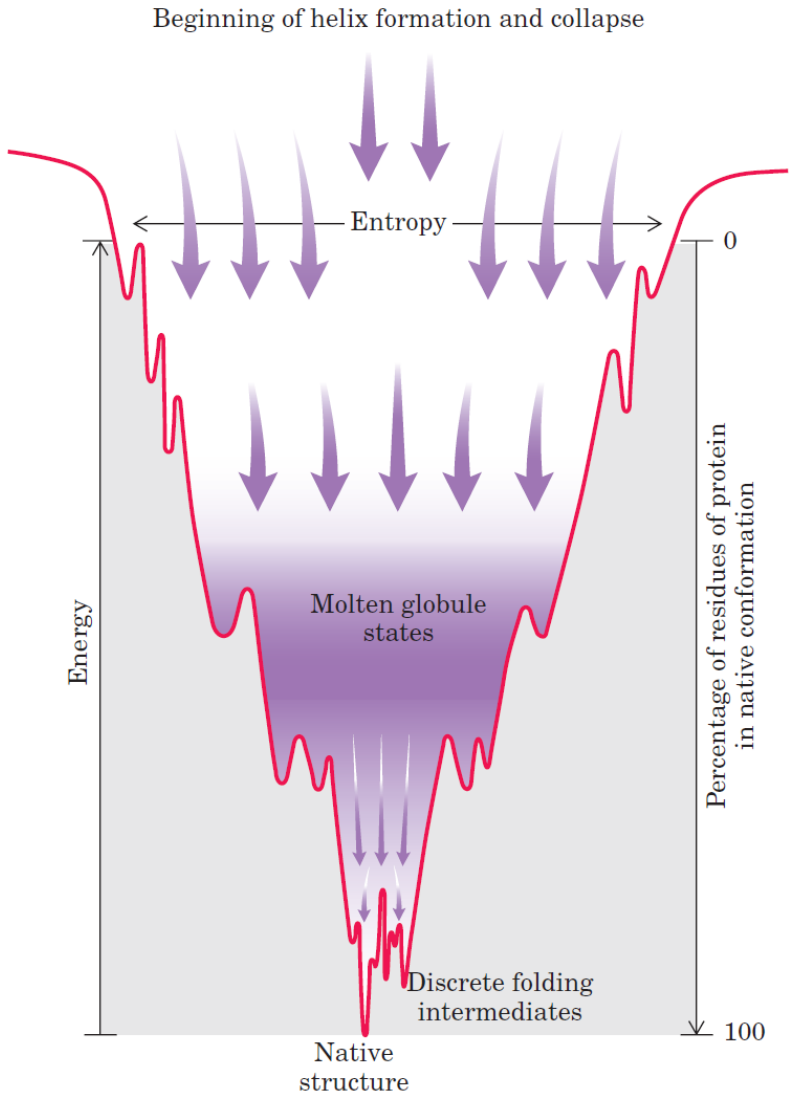
- Molten globule: interactions hydrophobes
- Structure 3D finale: liaisons hydrogènes

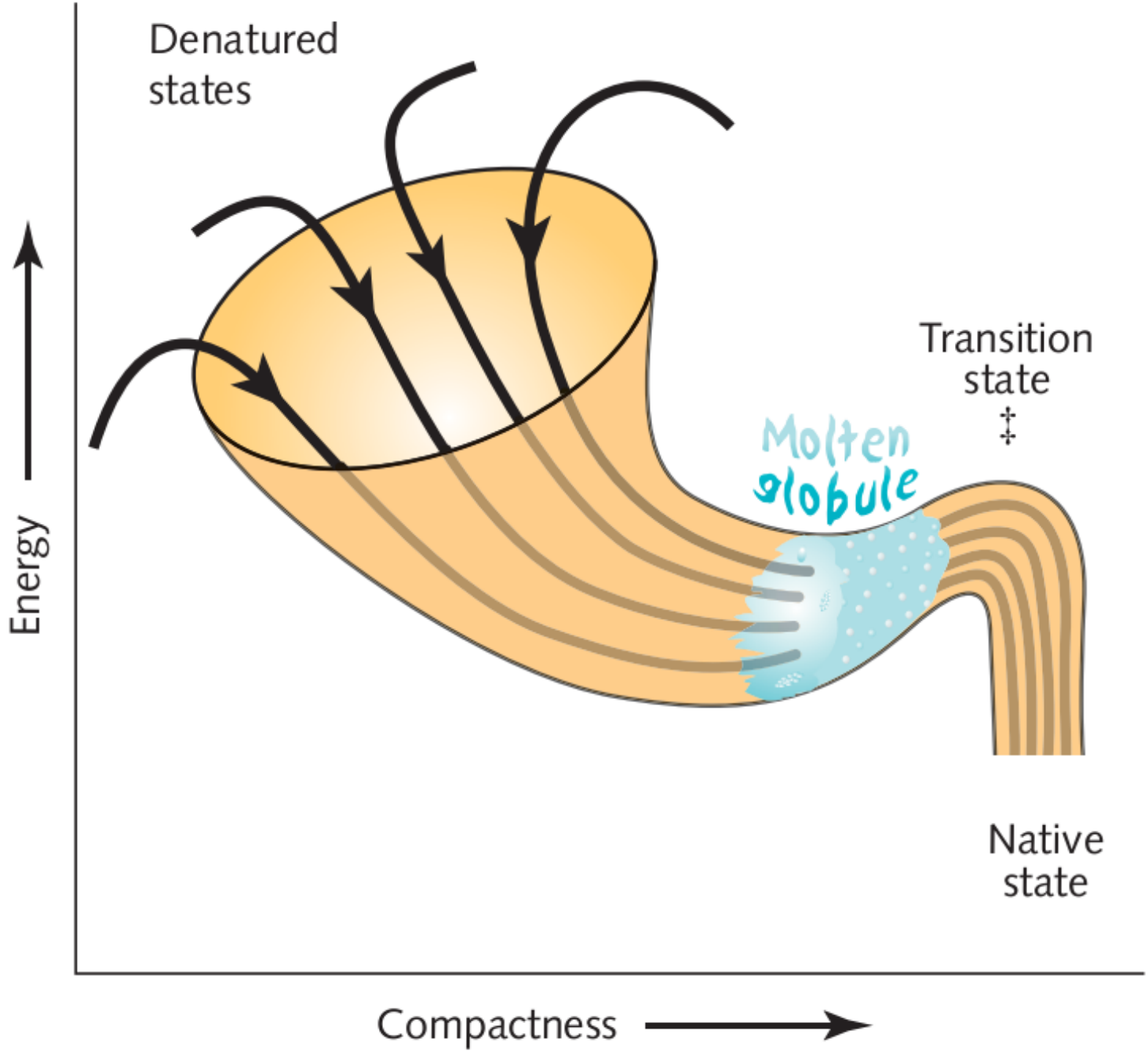
Pourquoi est le repliement des protéines si rapide ?

Paradoxe de Levinthal (1968):

- chaque résidu n'a que deux possibilité de conformation
- une protéine de 100 résidu aurait $2^{100} \approx 10^{30}$ conformations possibles.
- Conclusion: une protéine ne peut pas se replier par une recherche au hasard de la conformation native
- Elle doit suivre un chemin de repliement (*folding pathway*) plus efficace.

Entonnoir du repliement




Denatured states
Transition state
‡
Molten globule
Native state
Energy
Compactness

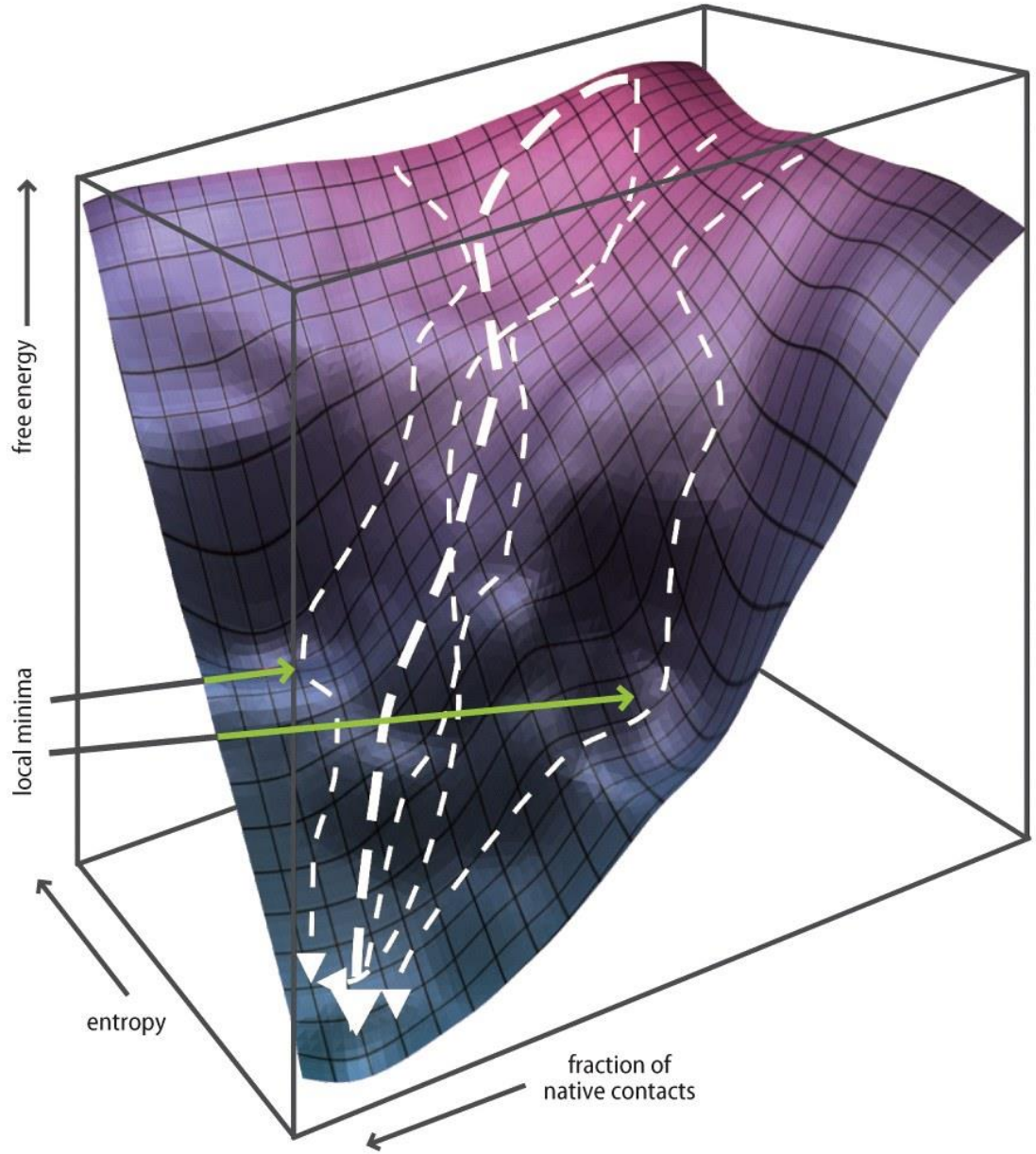
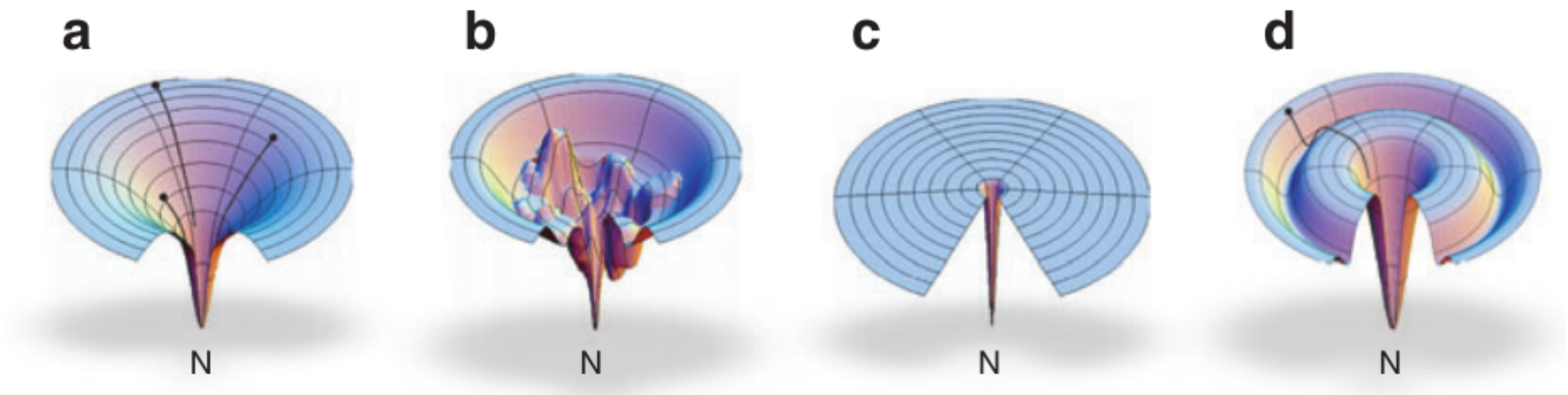


Figure 6.15 How Proteins Work (©2012 Garland Science)

Entonnoir du repliement

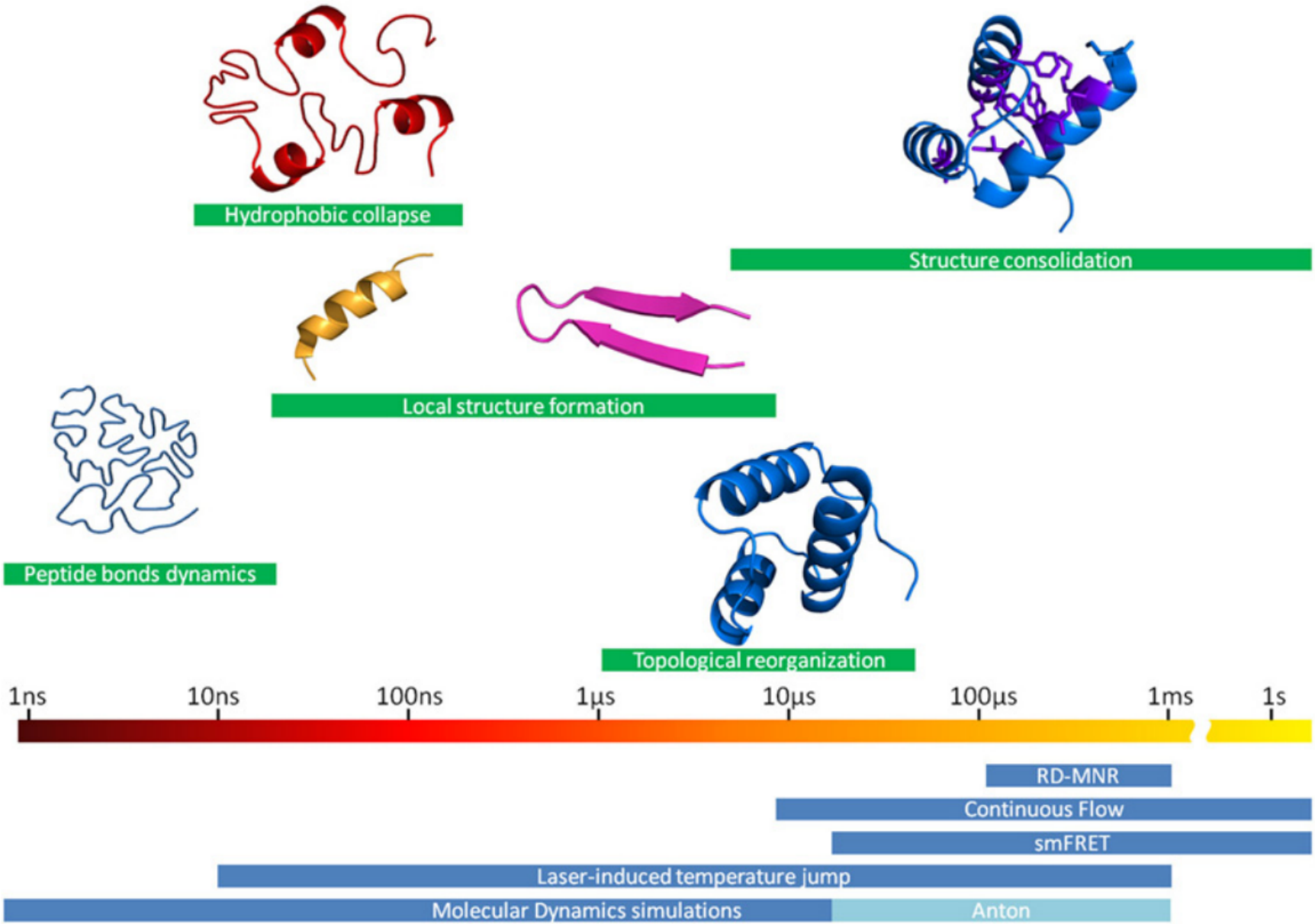


a: fast folder
b: kinetic traps
c: diffusional conformational search
d: obligatory intermediate

Ken A. Dill et al. (June 2008). en. In: *Annual Review of Biophysics* 37.1

Vitesses du repliement

- Hélices se forment plus rapidement que des feuillet
- Hélices <=> contacts proches dans la séquence
- Feuillets <=> contacts distants dans la séquence
- Hélices: 0.1 - 1 μs, β-hairpins: 1 - 10 μs
- La vitesse 'limite' du repliement: N/50 μs, N: nombre de résidus



Victor Muñoz and Michele Cerminara (Sept. 2016). In: *Biochem J* 473.17

État déplié

Rayon de giration

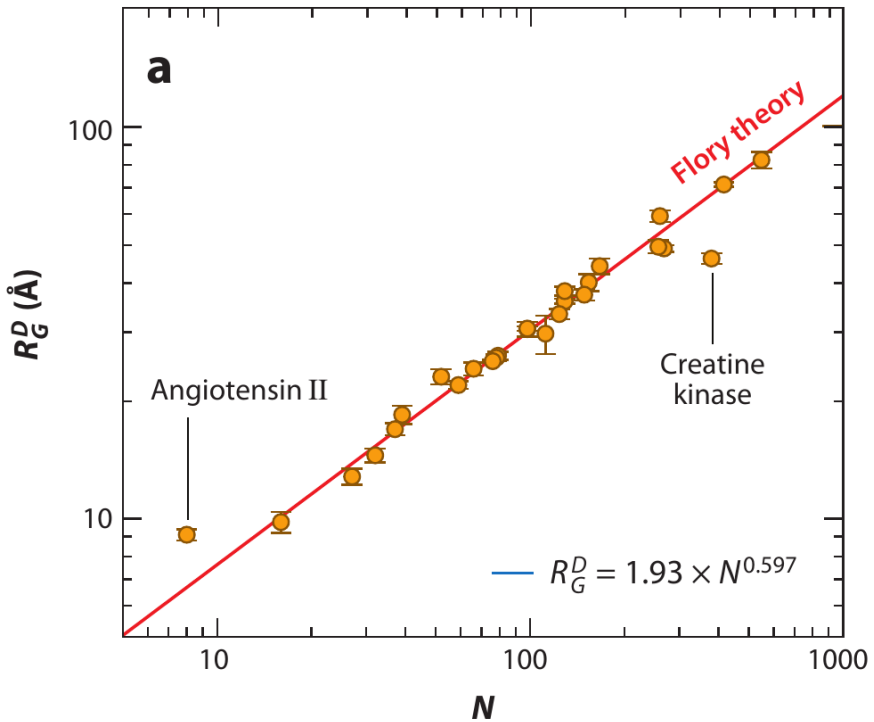
Protein	Number of residues	R_G (native)	R_G (denatured)	Ratio
PI3 kinase, SH3 domain	90	18.6	27.5	0.7
Horse heart cytochrome *c*	104	17.8	32.6	0.5
Hen egg white lysozyme	129	20.5	34.6	0.6
Yeast triose phosphate isomerase	247	29.7	49.7	0.6

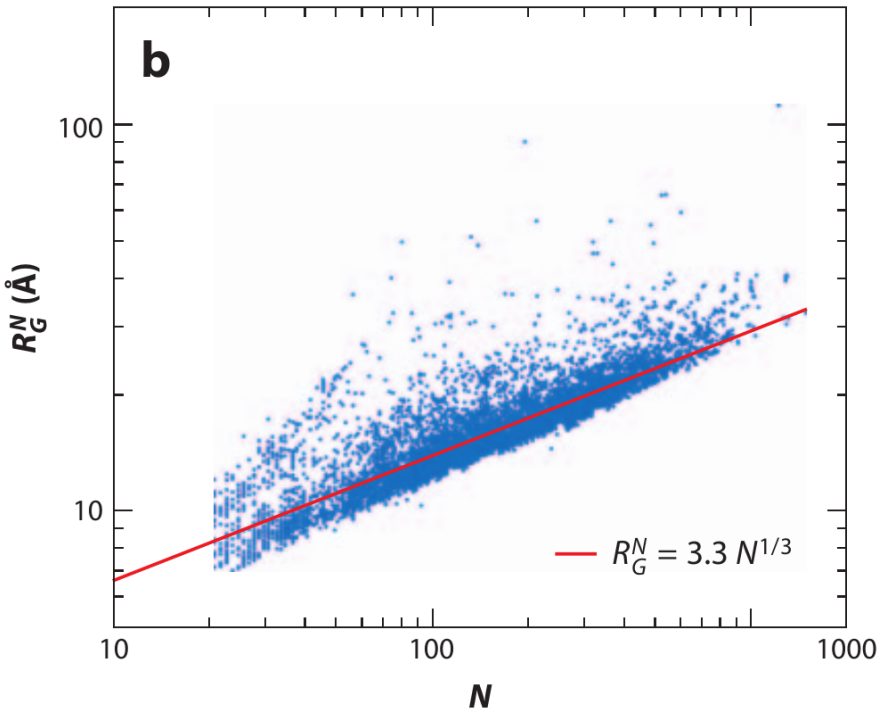
- Si centre de gravité à l'origine, alors:

$$R_G = \sqrt{\frac{\sum_i m_i(x_i^2 + y_i^2 + z_i^2)}{\sum_i m_i}}$$

- Random coil: $R_G \propto N^{0.6}$ avec N nombre de résidus
- Globule compacte: $R_G \propto N^{0.33}$

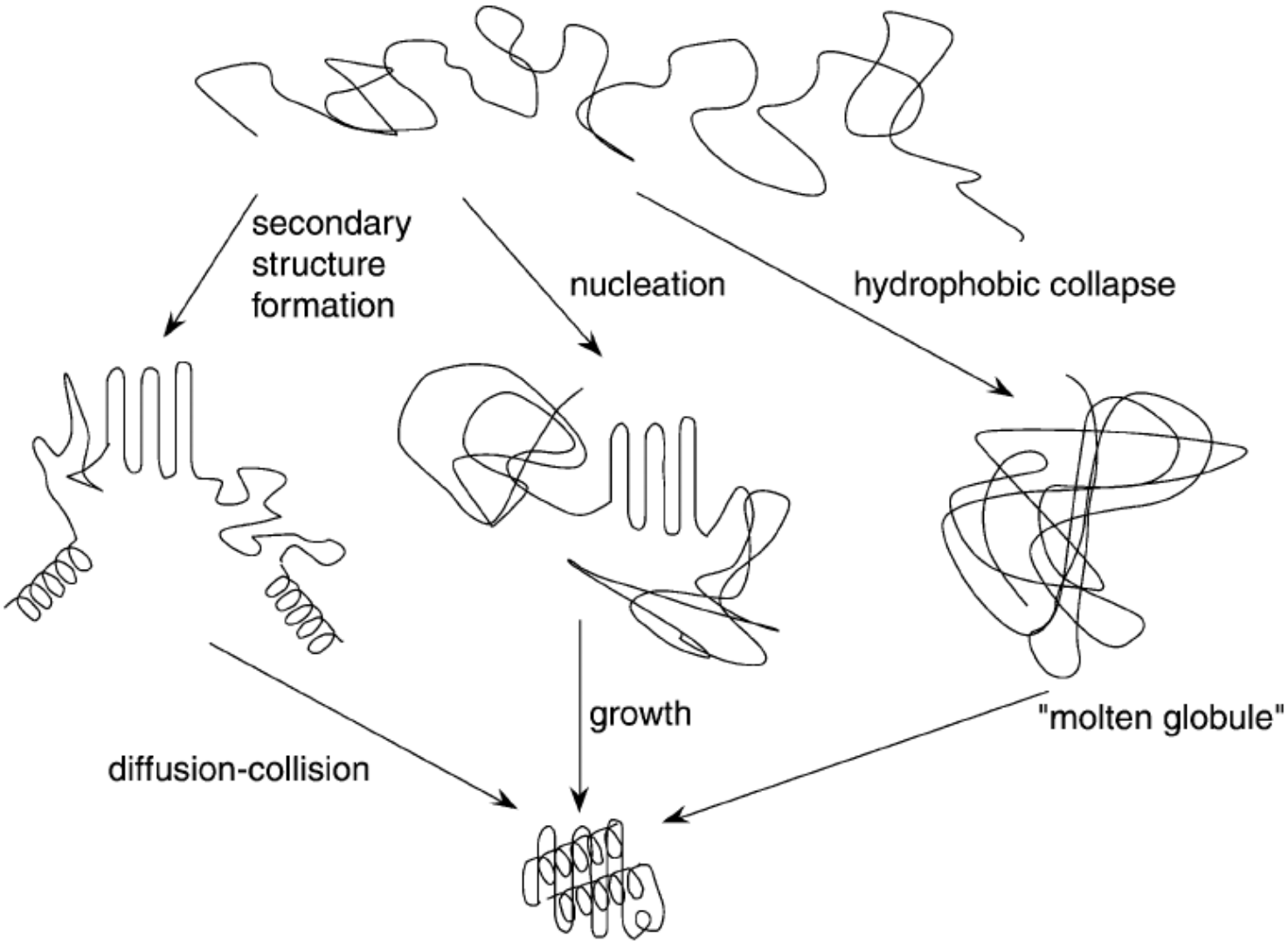
Rayon de giration





D. Thirumalai et al. (2010). In: *Annual Review of Biophysics* 39.1

Modèles classiques pour le repliement



à gauche: "framework" ou "hierarchical",

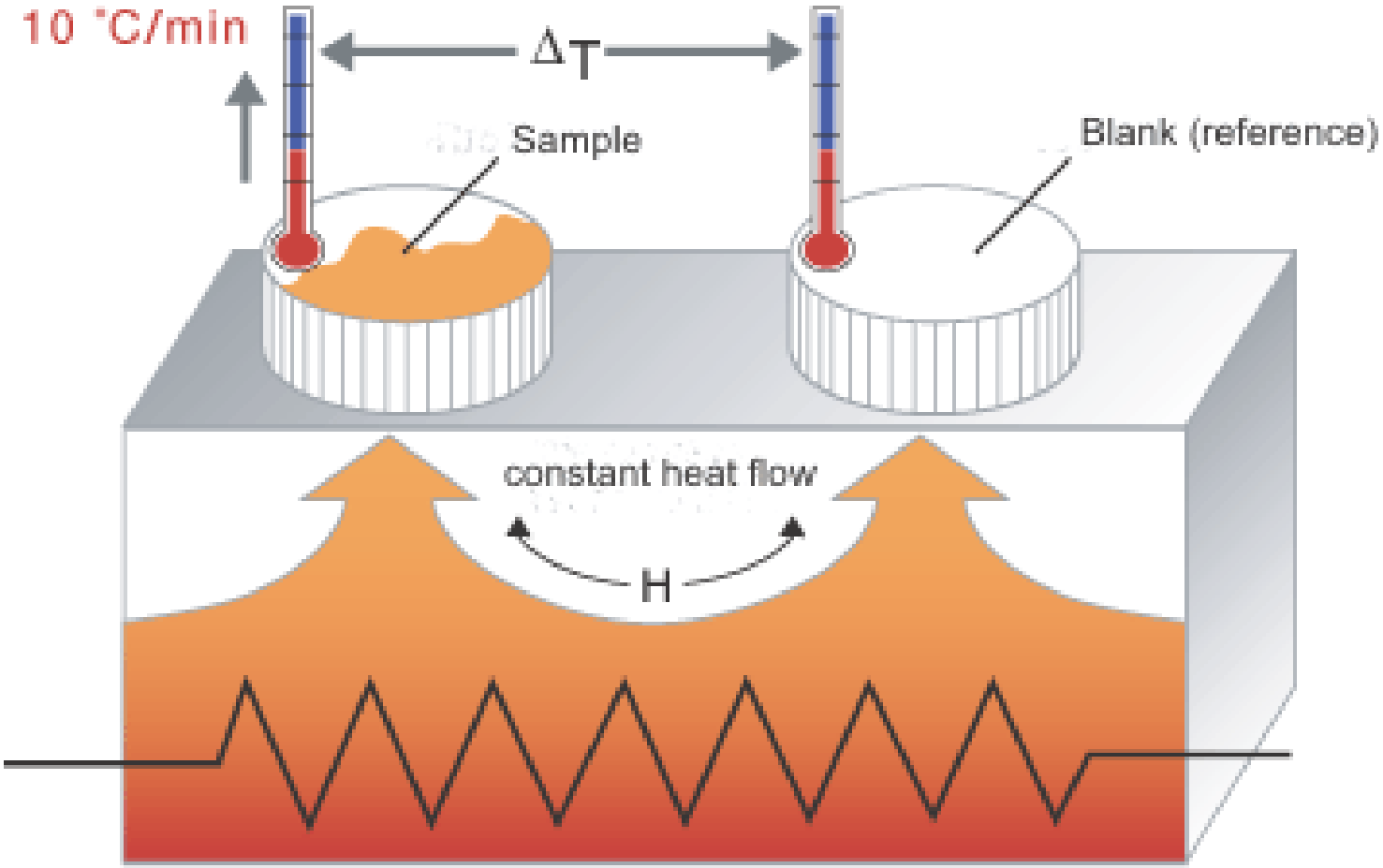
Alan R. Fersht and Valerie Daggett (2002). In: *Cell* 108.4

2 Techniques expérimentales

- Méthodes thérmodynamiques
- Techniques sur molécule unique
- RMN
- Méthodes pour cinétique "ultra rapide"

Méthodes thérmodynamiques

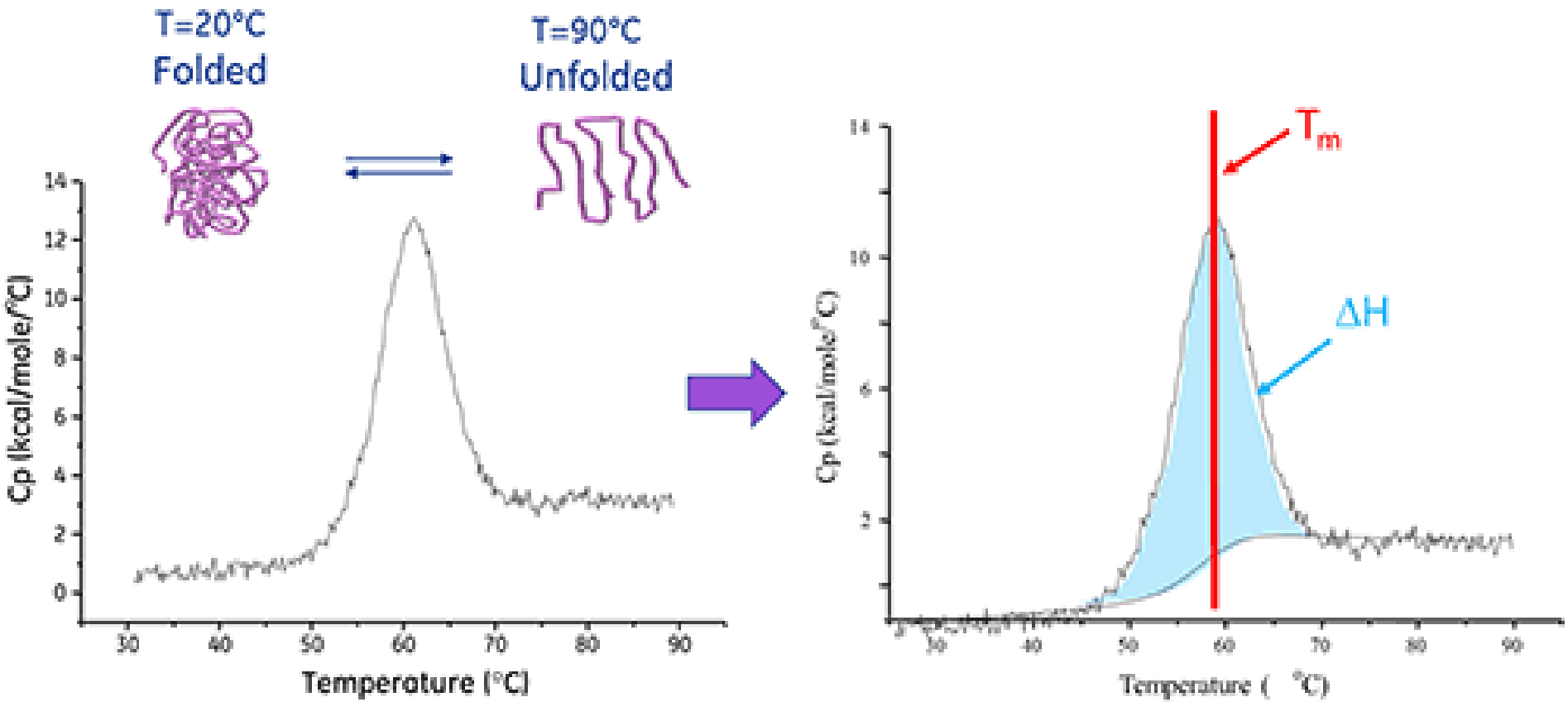
Differential scanning calorimetry (DSC)



Principe: référence et échantillion sont chauffés simultanément
Protéine absorbe de la chaleur lors du dépliement => ΔT

www.itc.tu-bs.de/Abteilungen/Makro/Methods/dsc.htm

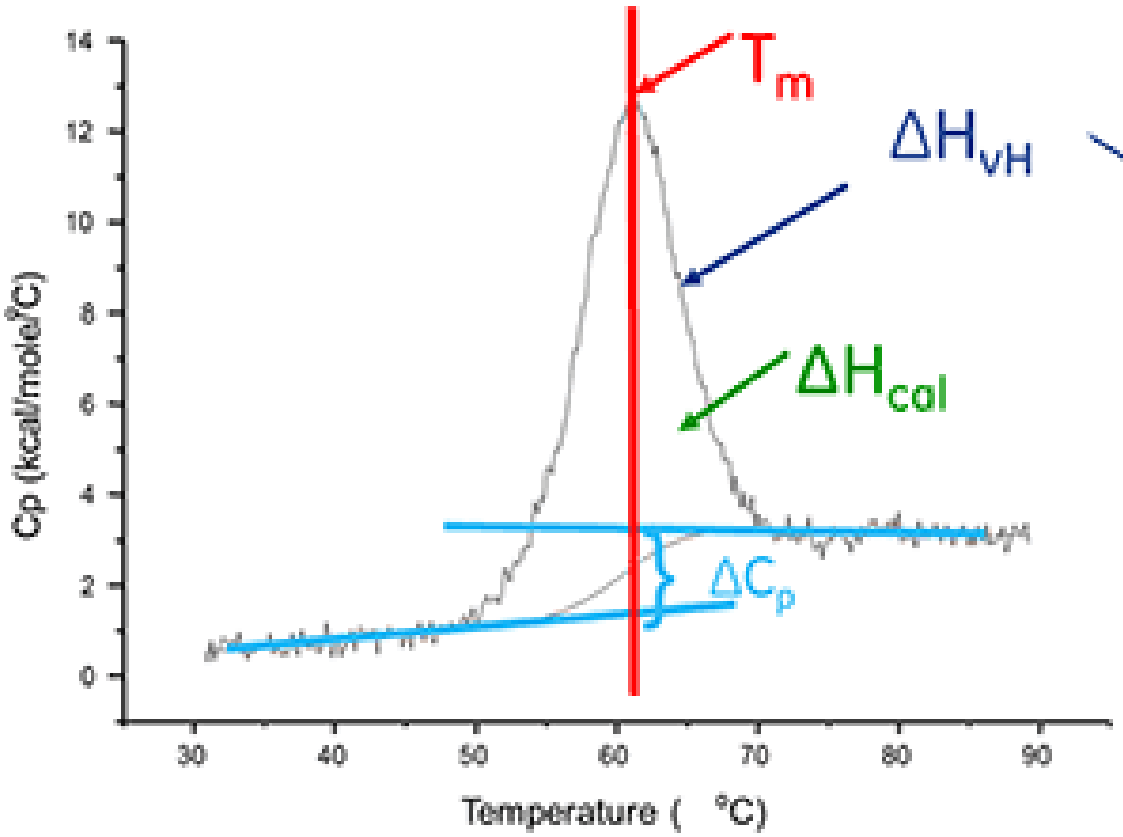
Differential scanning calorimetry (DSC)

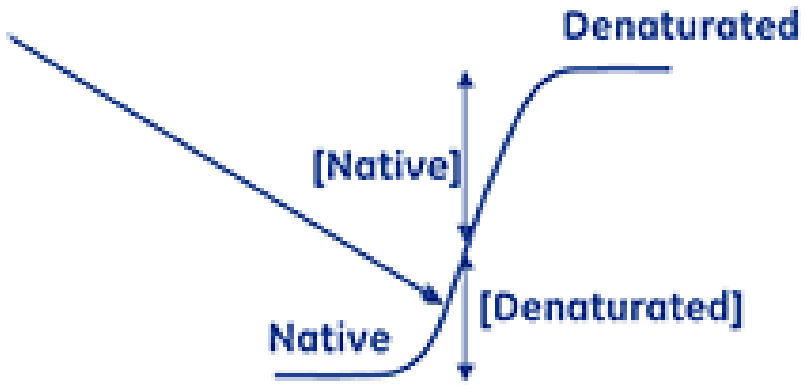


Tm: température médiane de transistion thermique (thermal transition midpoint)
Cp: capacité calorifique (heat capacity)
H: enthalpie

www.malvern.com/fr/products/technology/differential-scanning-calorimetry

Differential scanning calorimetry (DSC)

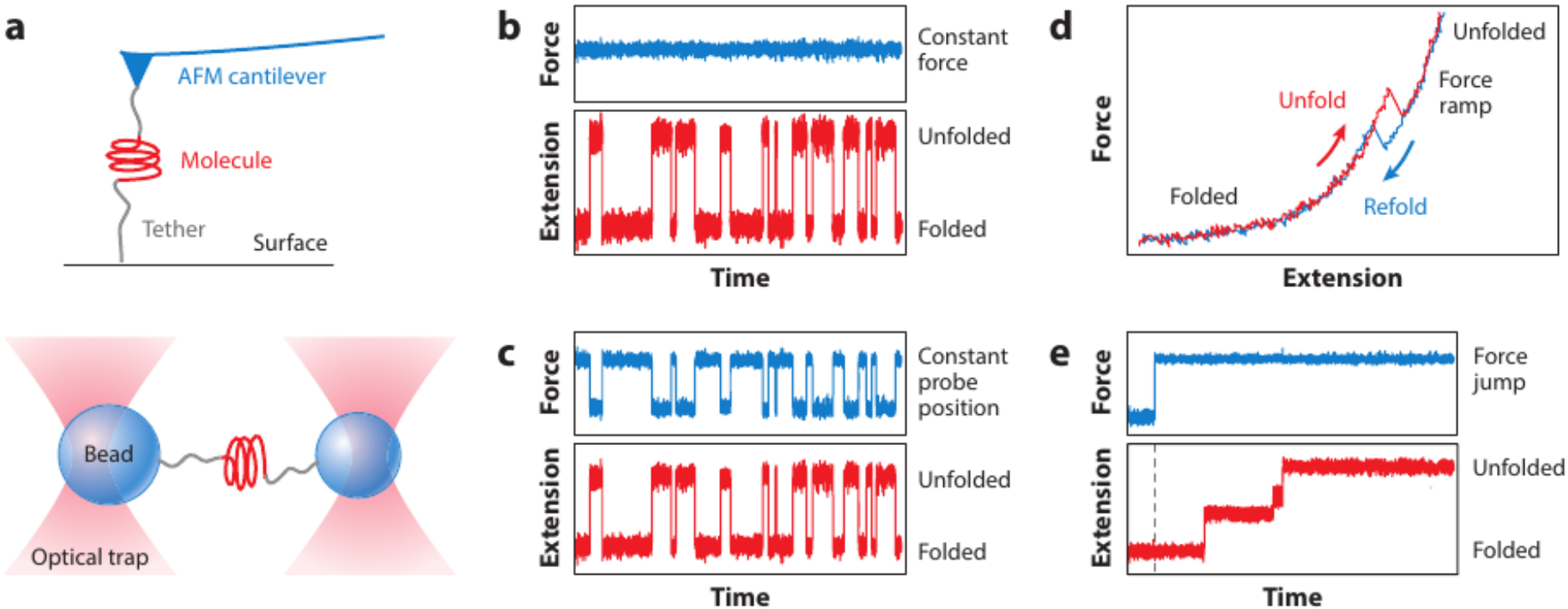




Tm: température médiane de transistion thermique (thermal transition midpoint)
Cp: capacité calorifique (heat capacity)
H: enthalpie
www.malvern.com/fr/products/technology/differential-scanning-calorimetry

Techniques sur molécule unique

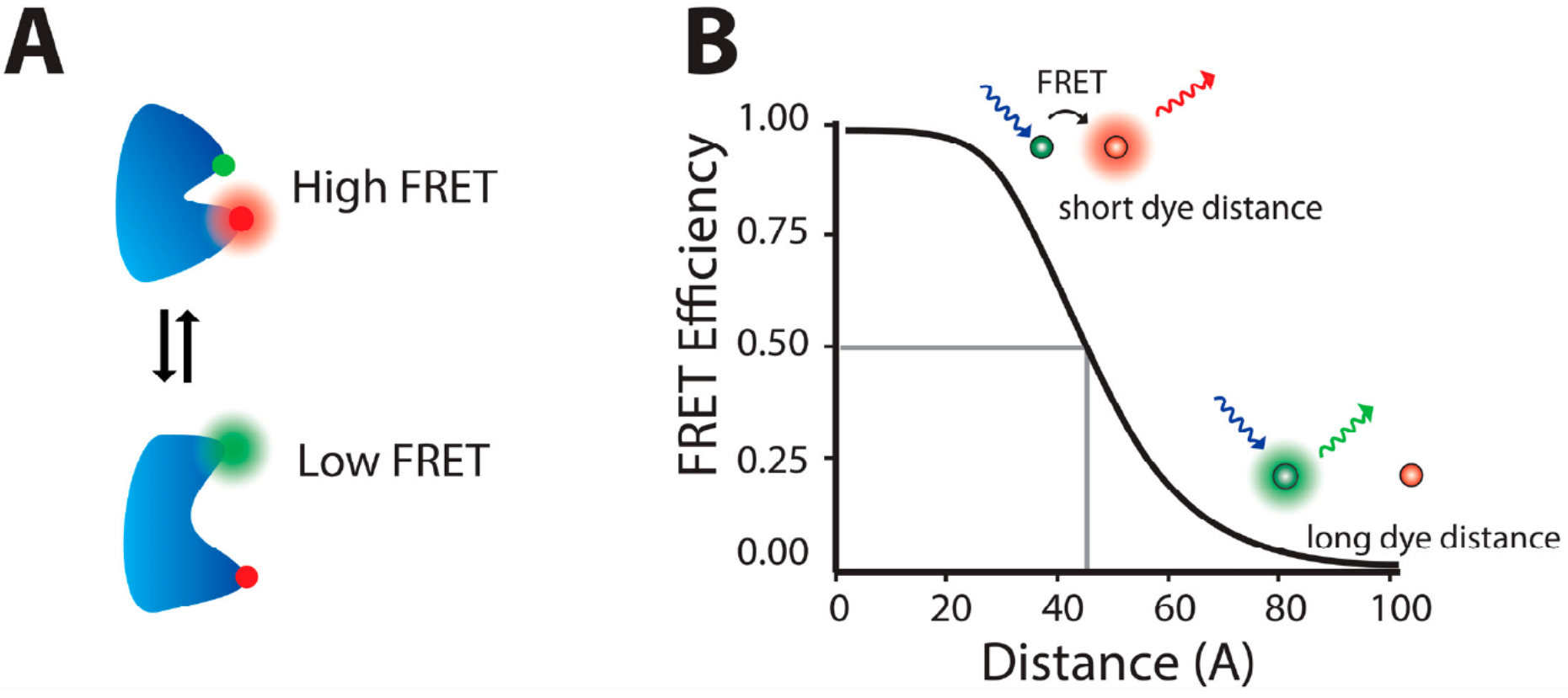
Single molecule force spectroscopy (SMFS)



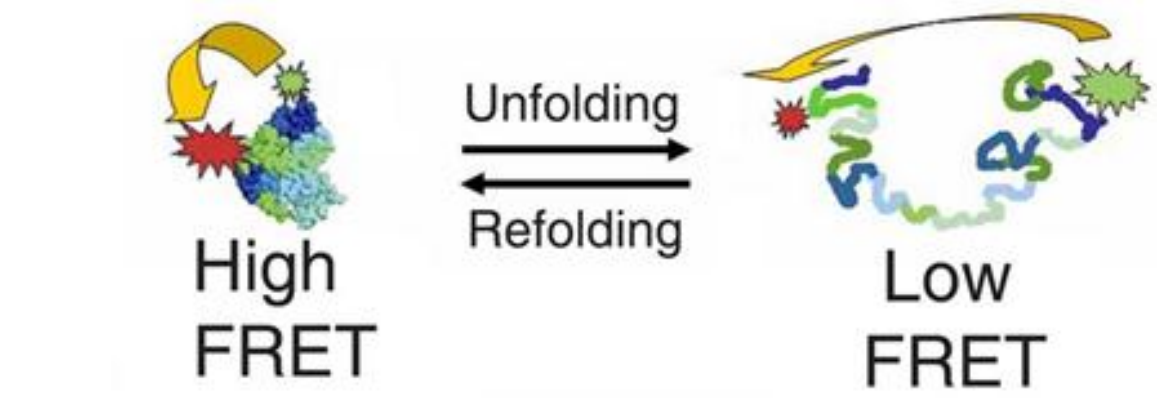
a) Atomic force microscopy (AFM) and Opticial tweezers
b) Constant force mode: extension fluctuates
c) Constant probe position: force and extension fluctuate
d) Force-ramp mode: elastic stretching is interrupted by a "rip", hysteresis indicates a nonequilibrium process
e) Force-jump mode: extension changes in steps

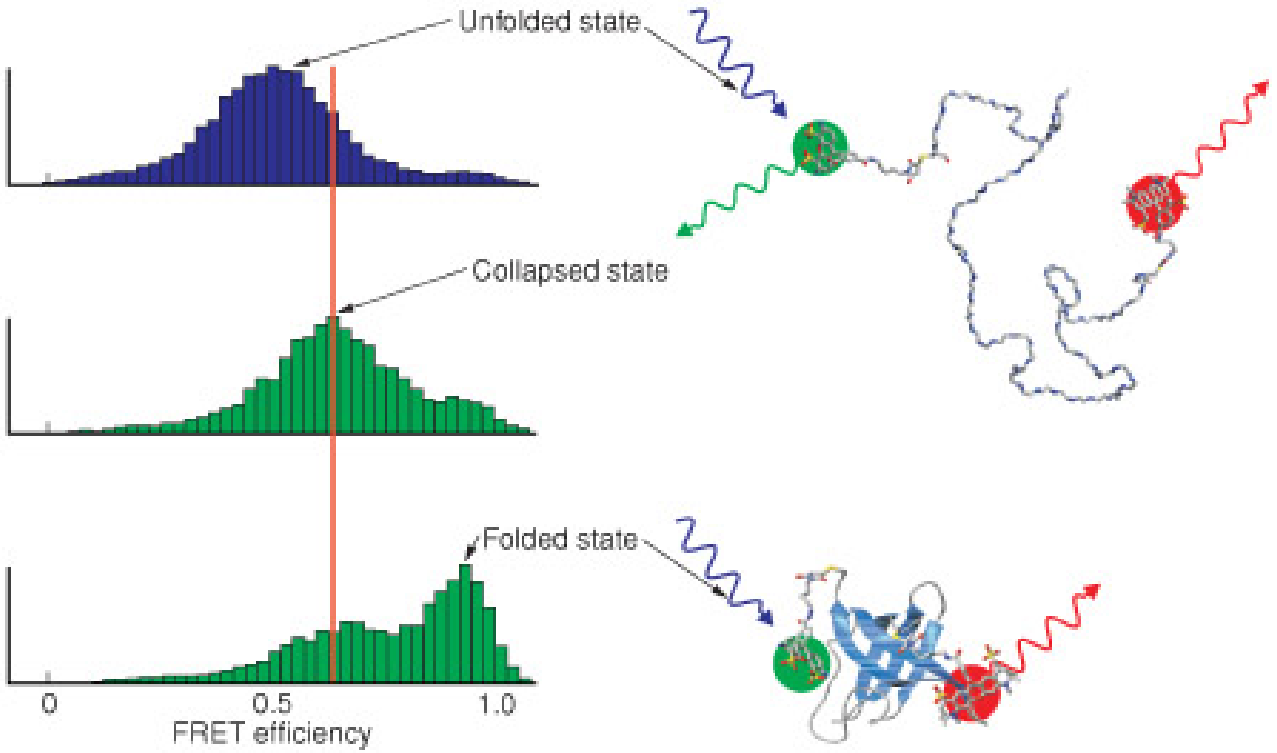
Michael T. Woodside and Steven M. Block (2014). In: *Annual Review of Biophysics* 43.1

Fluorescence resonance energy transfer (FRET)

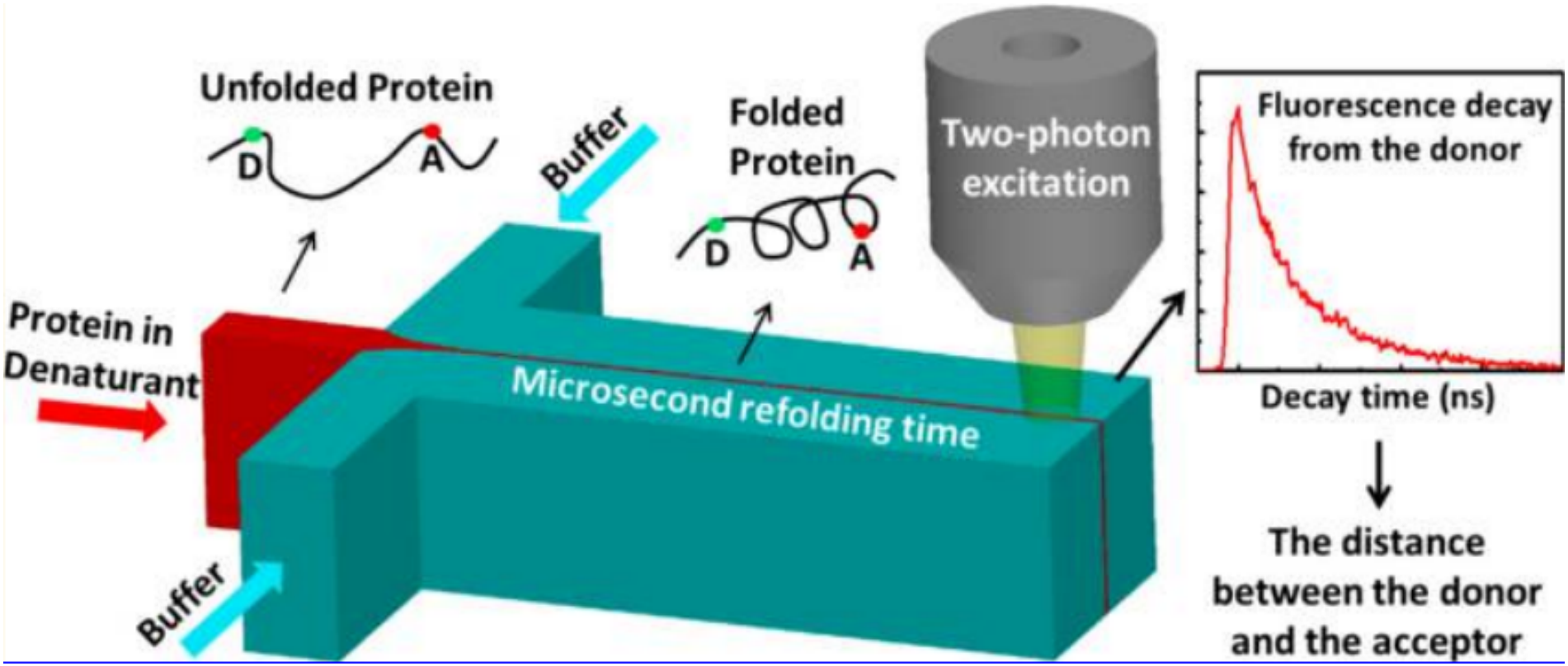


Fluorescence resonance energy transfer (FRET)

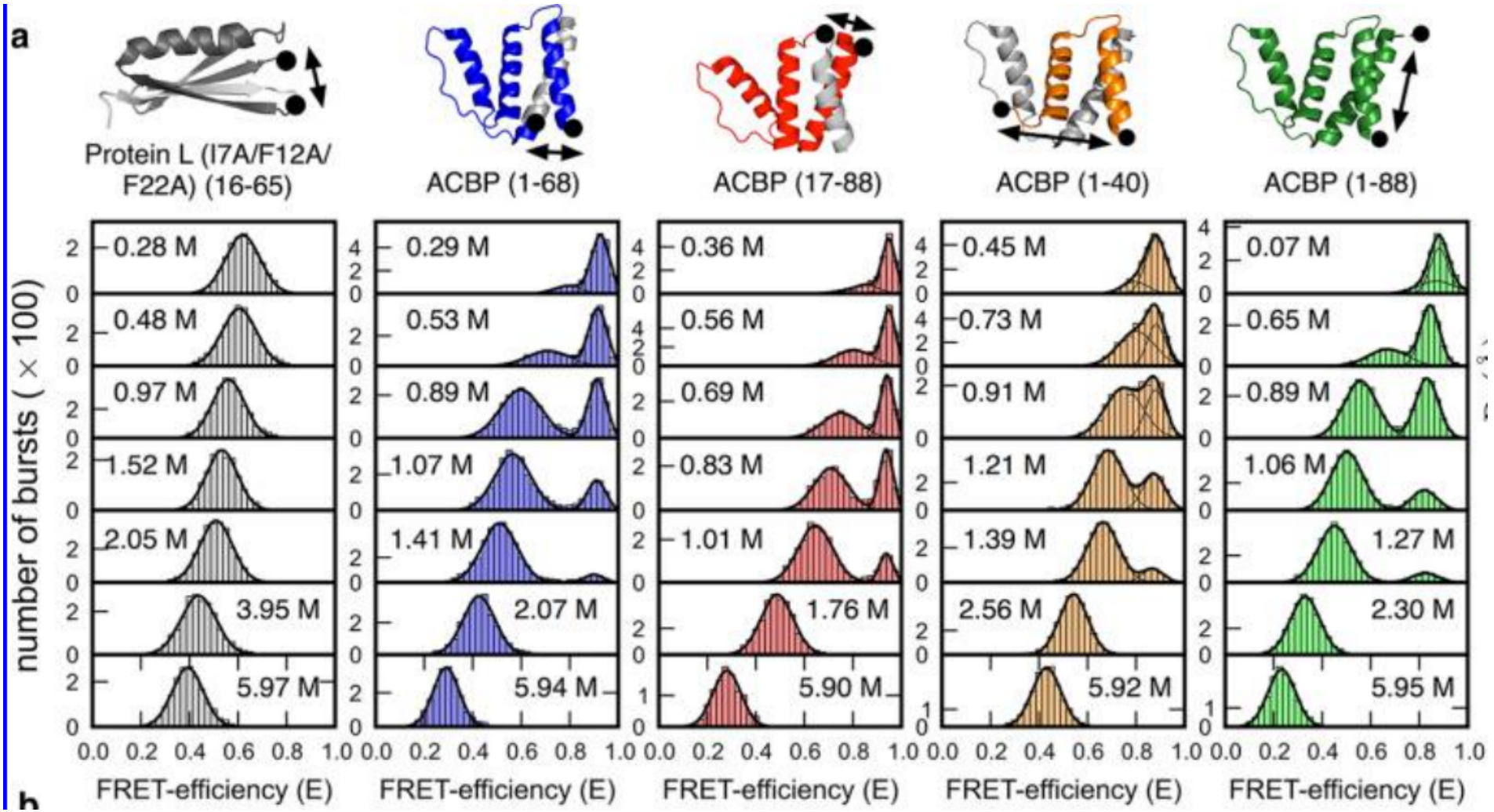




Time-resolved FRET (TR-FRET)



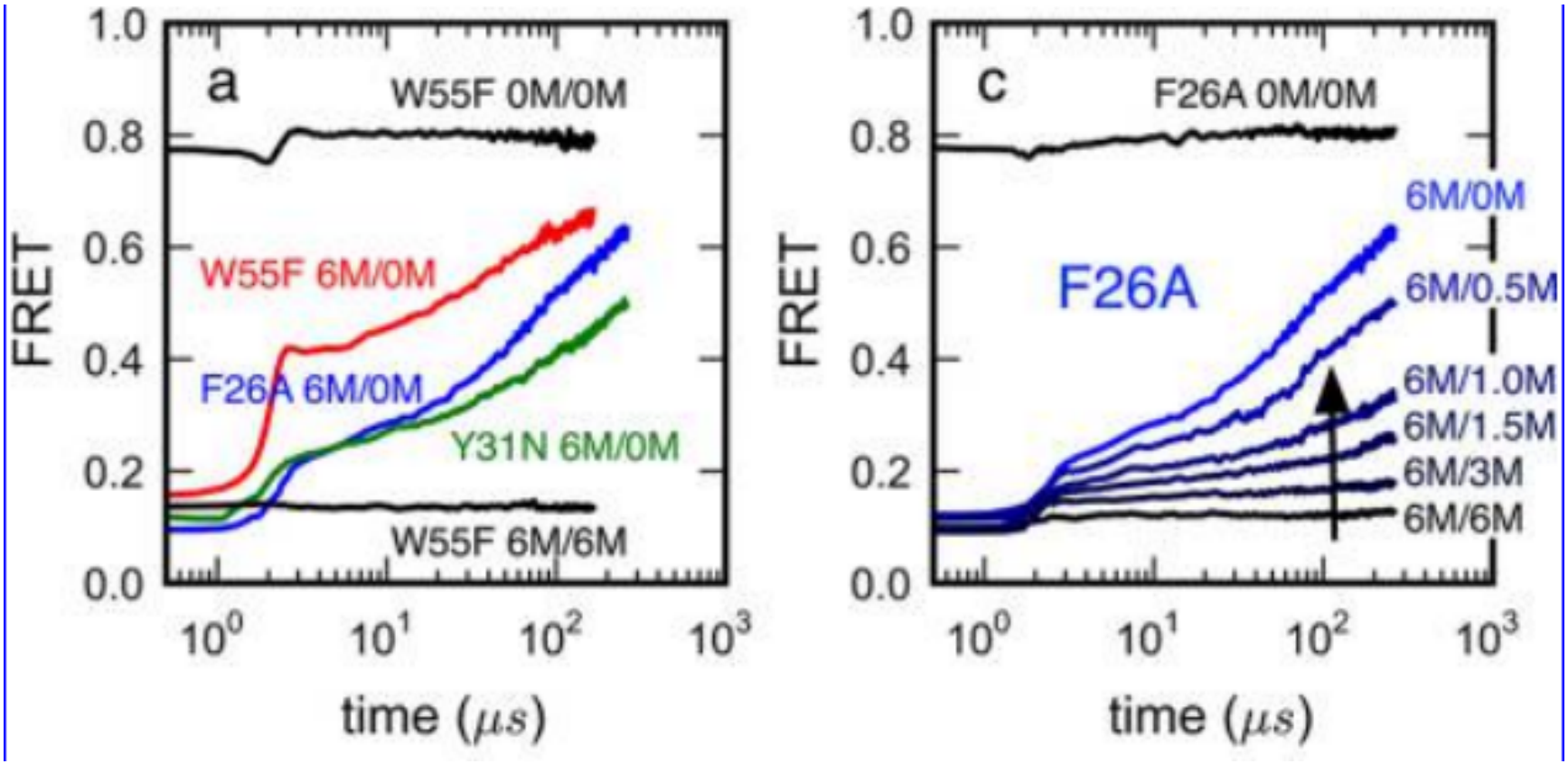
FRET sur ACBP



Vincent A. Voelz et al. (Aug. 2012). In: *J. Am. Chem. Soc.* 134.30

FRET sur ACBP - Collapse très lent

Solution dénaturant (6 M GuHCl) vers solution de repliement (0 M GuHCl):

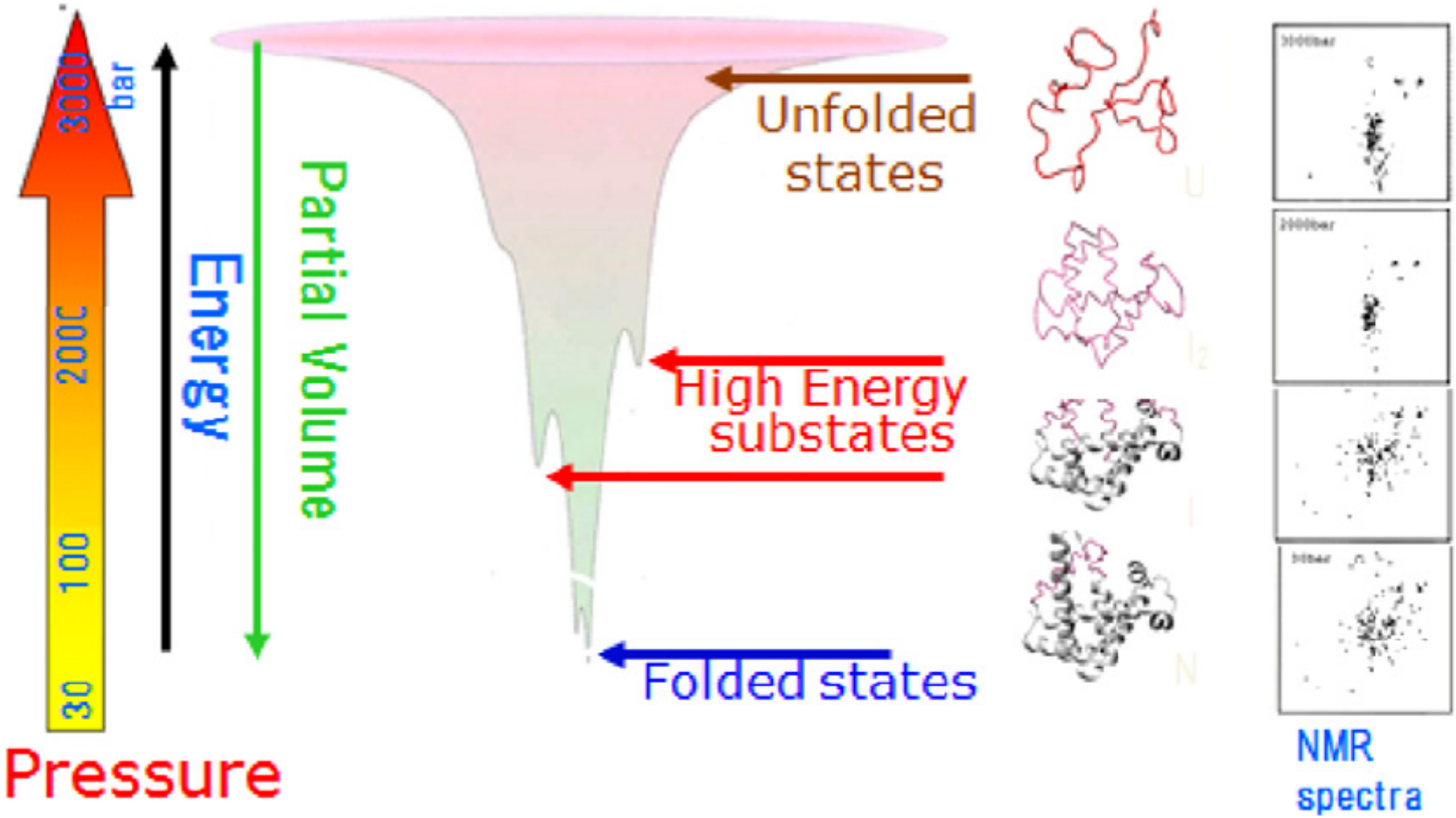


Indication pour: Formation d'une ensemble compact et hétérogènes de structures dépliées

Vincent A. Voelz et al. (Aug. 2012). In: *J. Am. Chem. Soc.* 134.30

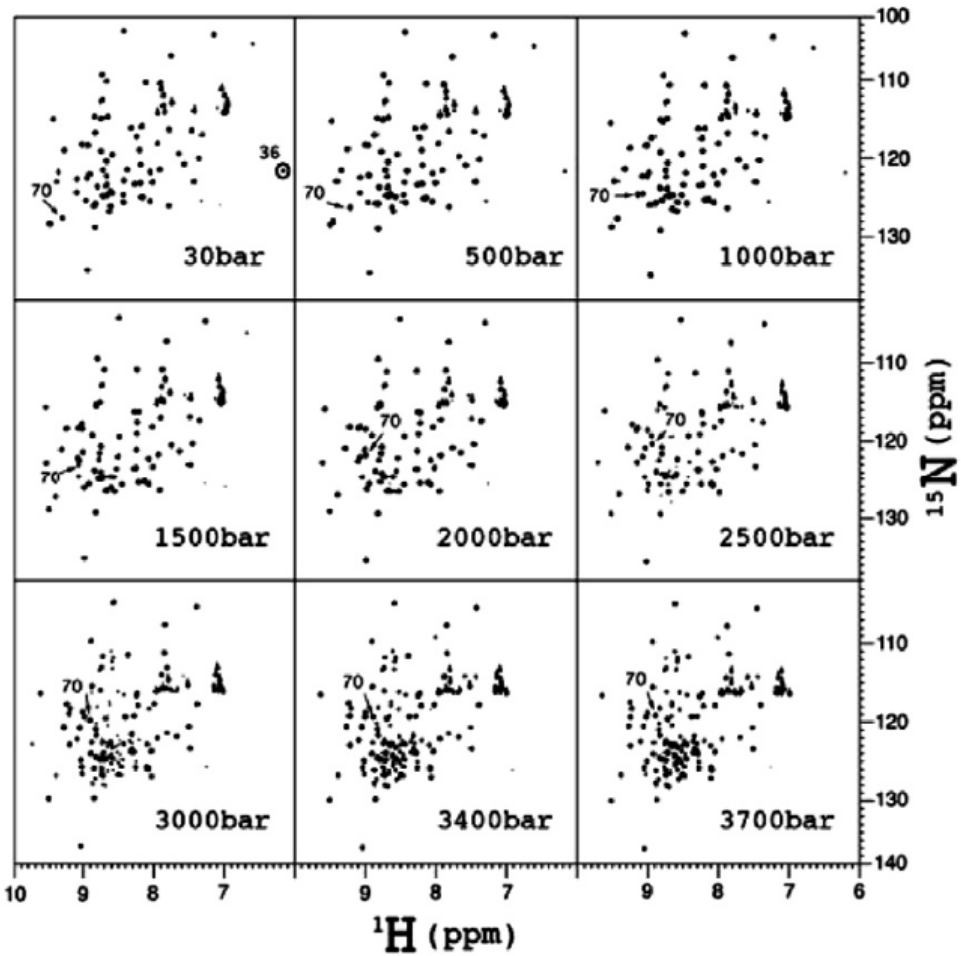
RMN

Pression et RMN



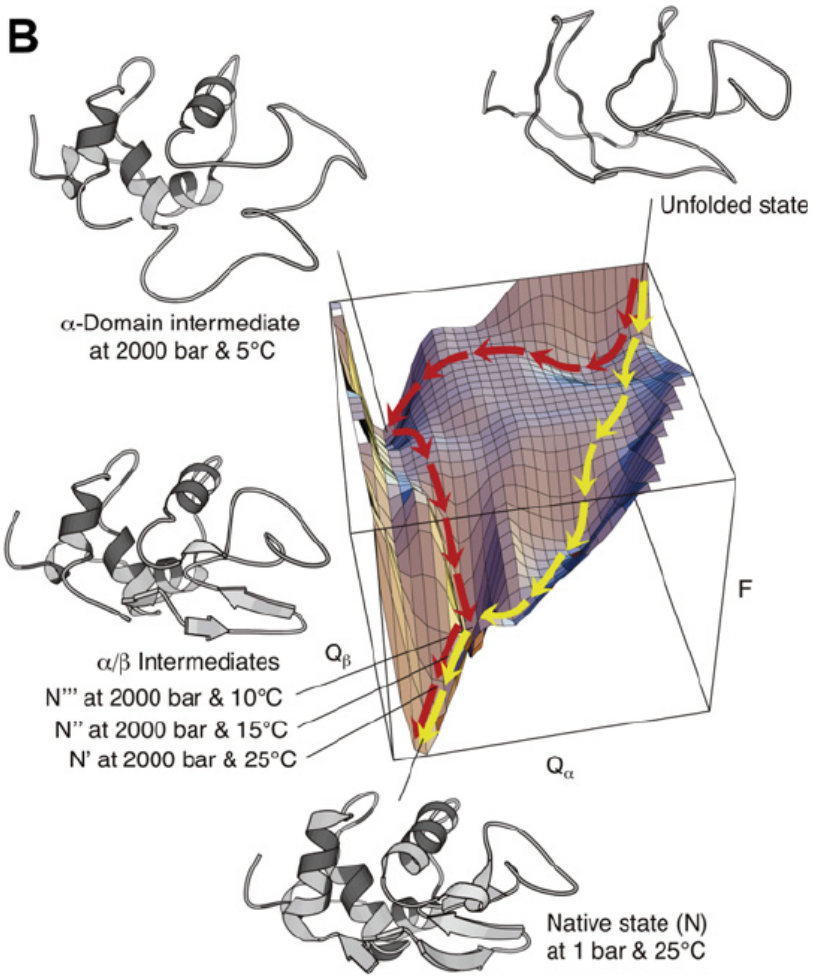
Kazuyuki Akasaka, Ryo Kitahara, and Yuji O. Kamatari (Mar. 2013). In: *Archives of Biochemistry and Biophysics*. Protein Folding and Stability 531.1–2

Pression et RMN



Kazuyuki Akasaka, Ryo Kitahara, and Yuji O. Kamatari (Mar. 2013). In: *Archives of Biochemistry and Biophysics*. Protein Folding and Stability 531.1–2

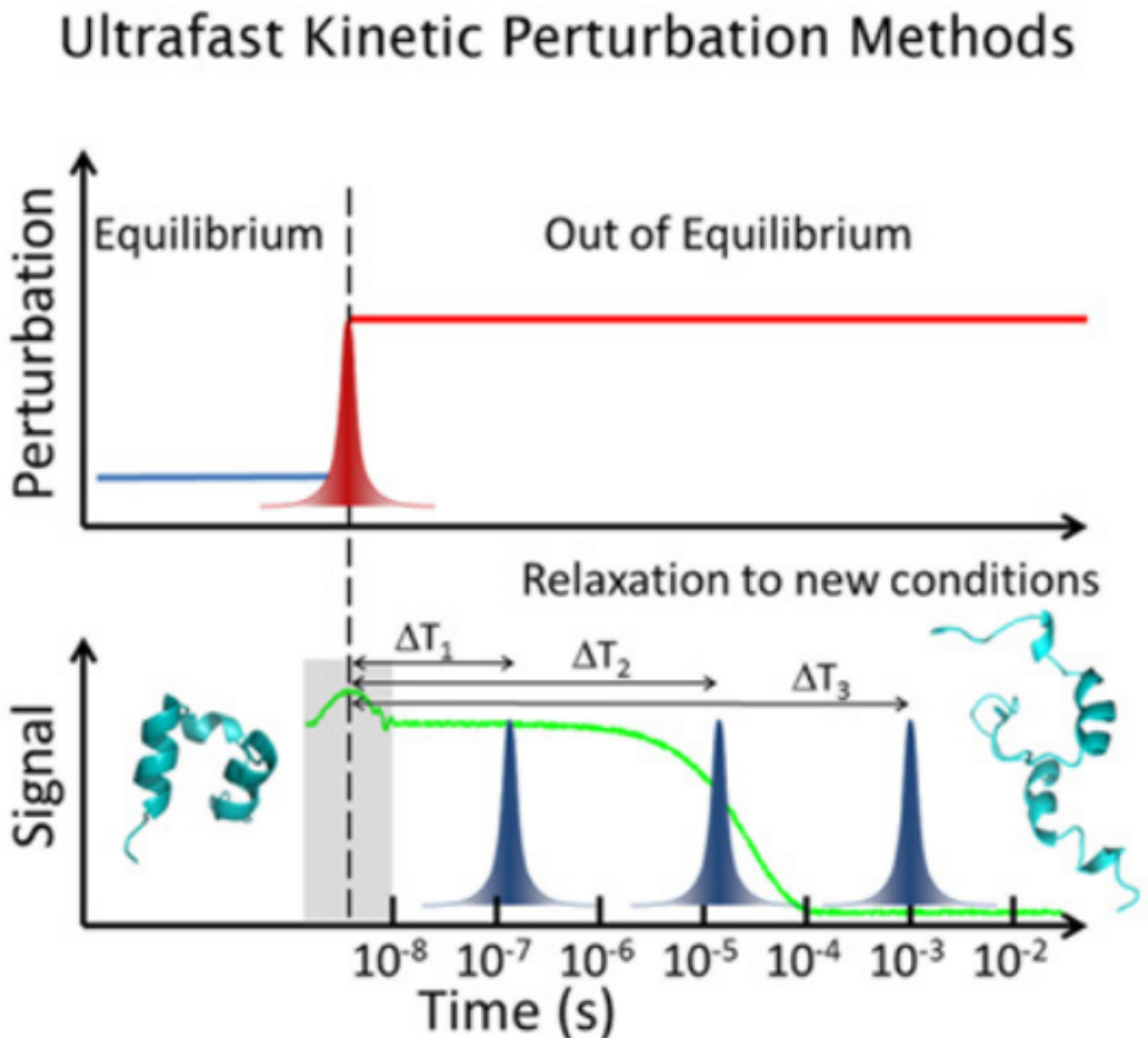
Pression et RMN



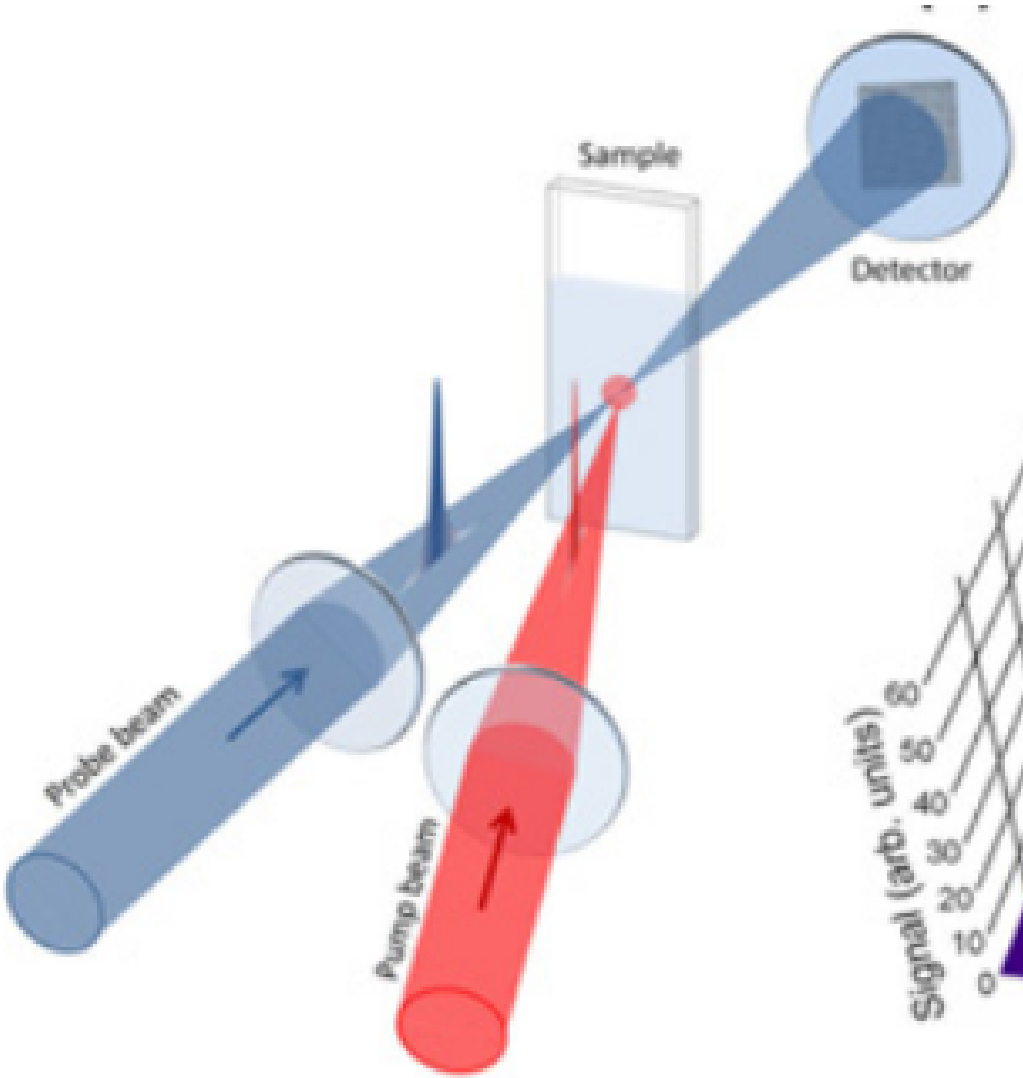
Kazuyuki Akasaka, Ryo Kitahara, and Yuji O. Kamatari (Mar. 2013). In: *Archives of Biochemistry and Biophysics*. Protein Folding and Stability 531.1–2

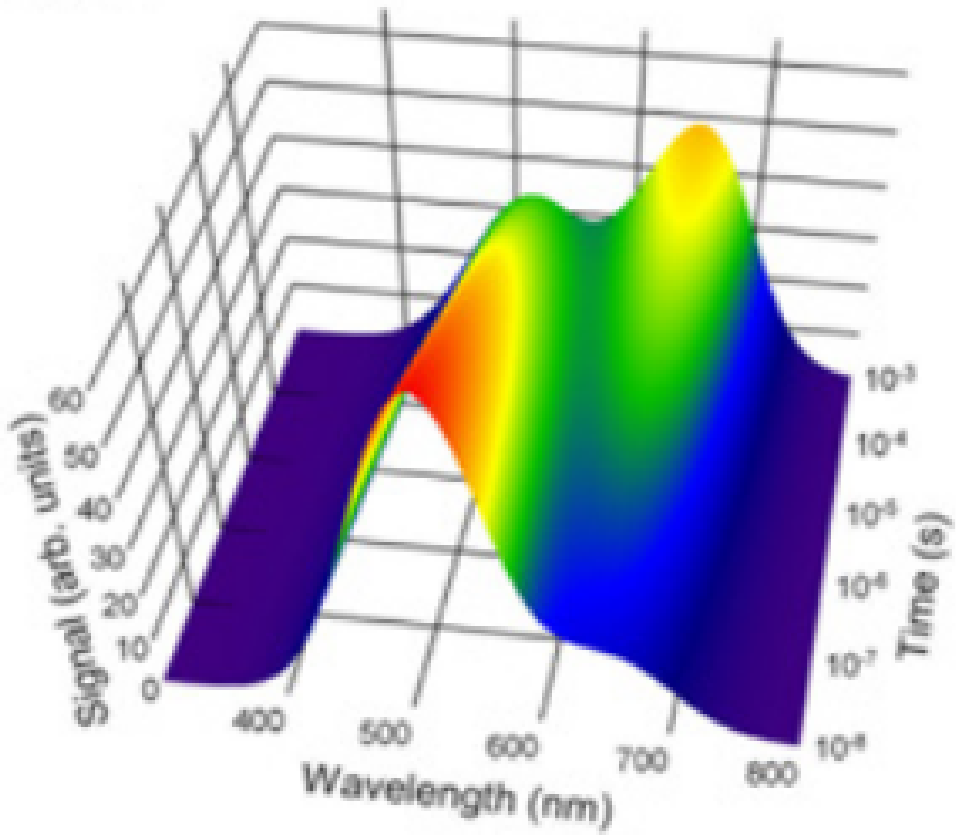
Méthodes pour cinétique "ultra rapide"

Laser T-jump


Ultrafast Kinetic Perturbation Methods
Perturbation
Equilibrium
Out of Equilibrium
Relaxation to new conditions
ΔT1
ΔT2
ΔT3
Signal
10-8 10-7 10-6 10-5 10-4 10-3 10-2
Time (s)

Laser T-jump


Sample
Detector
Probe beam
Pump beam


Signal (arb. units)
0
10
20
30
40
50
60
400
500
600
700
800
Wavelength (nm)
Time (s)
10⁻³
10⁻⁴
10⁻⁵
10⁻⁶
10⁻⁷
10⁻⁸

3 Repliement par dynamique moléculaire

- Introduction
- Temps de repliement
- Markov State Models (MSM)

Introduction

Anton

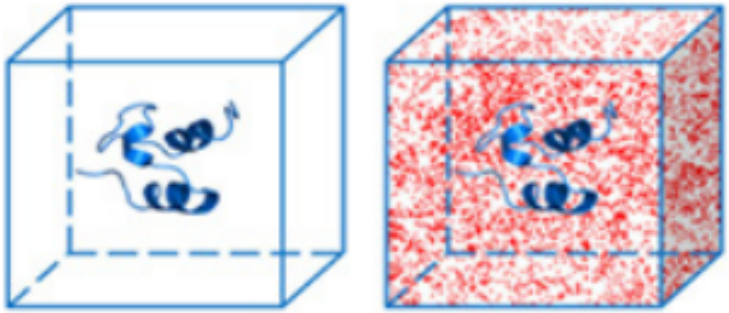
Molecular Dynamics Simulations

A. Force Field

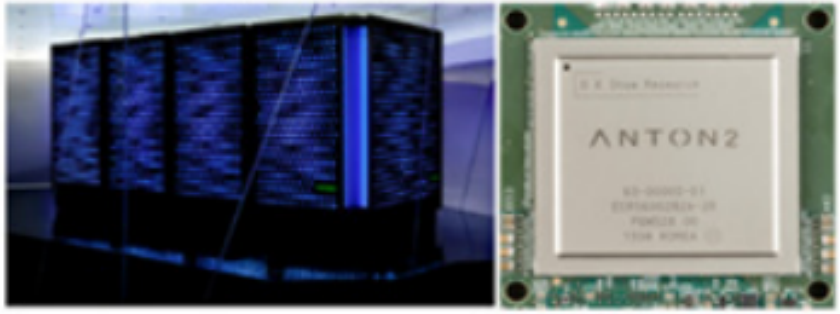
$$U = \sum k_b (r - r_0)^2 + \sum k_\theta (\theta - \theta_0)^2 + \sum A[1 + \cos(nT - \varphi)] + \sum\sum q_i q_j / r_{ij} + \sum\sum B\left[\left(\frac{\sigma_{ij}}{r_{ij}}\right)^{12} - \left(\frac{\sigma_{ij}}{r_{ij}}\right)^{6}\right]$$

bond stretching | bending | torsional | electrostatics | van der Waals

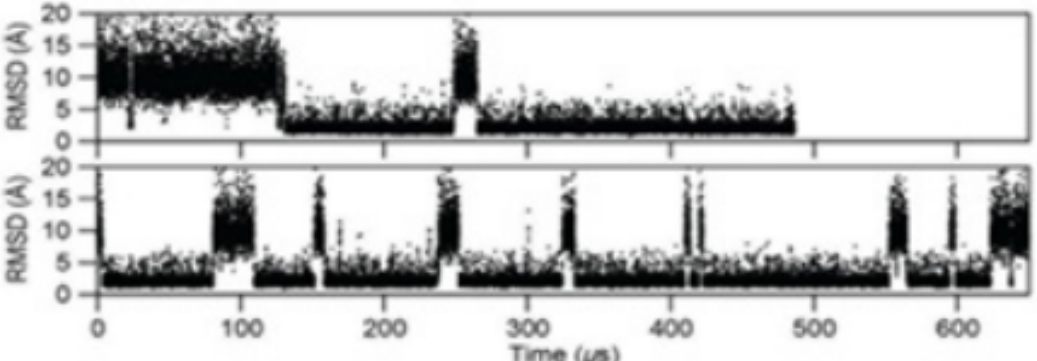
B. Simulation System



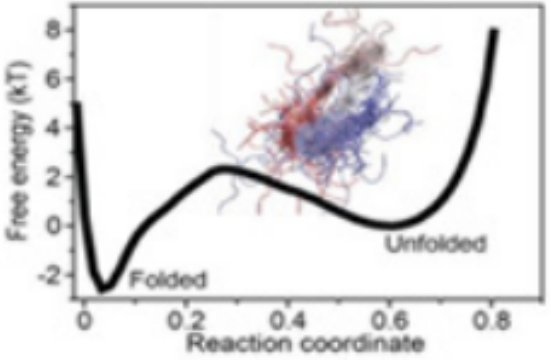
C. Specialized High Performance Computing



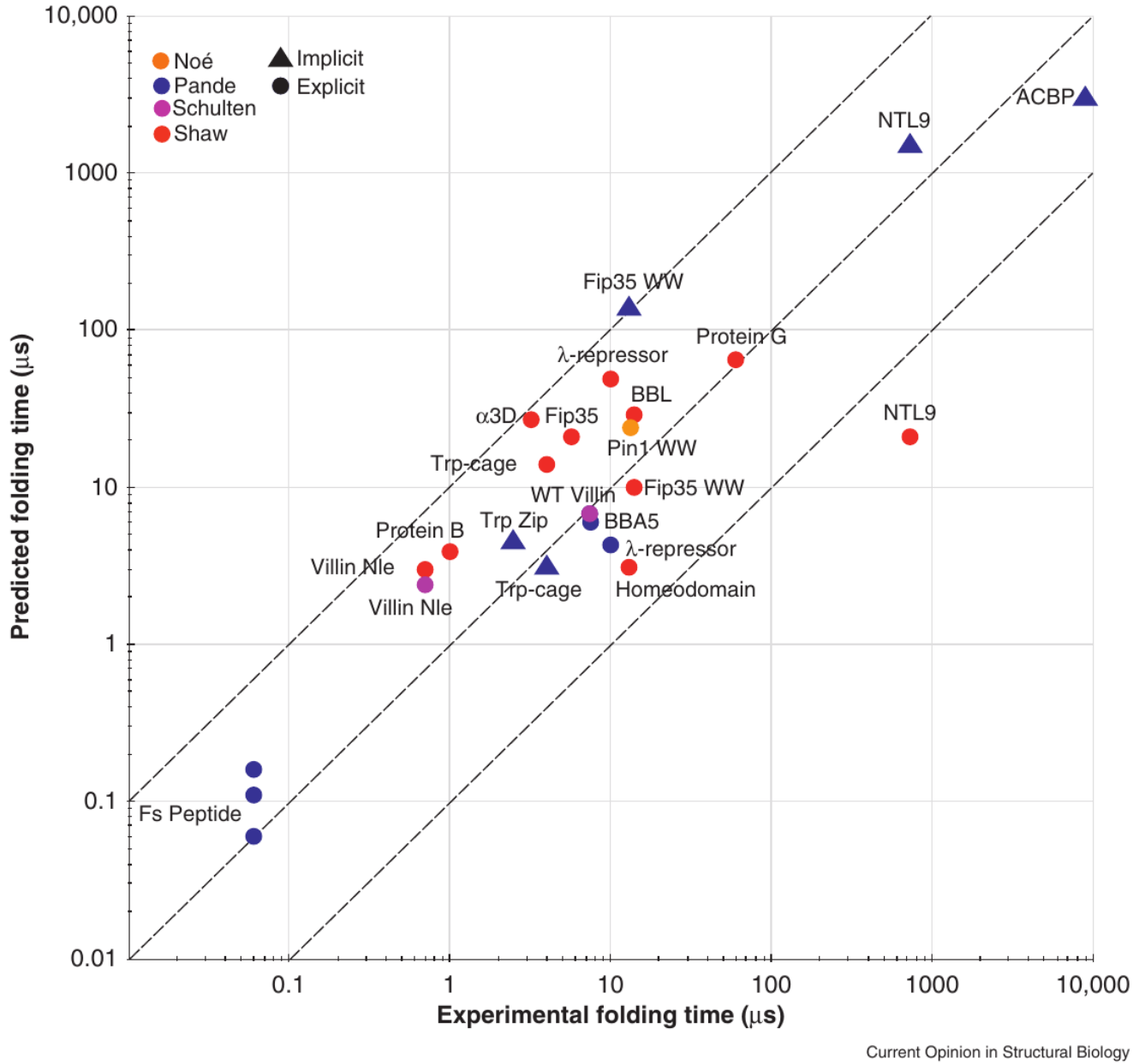
D. Molecular Trajectories



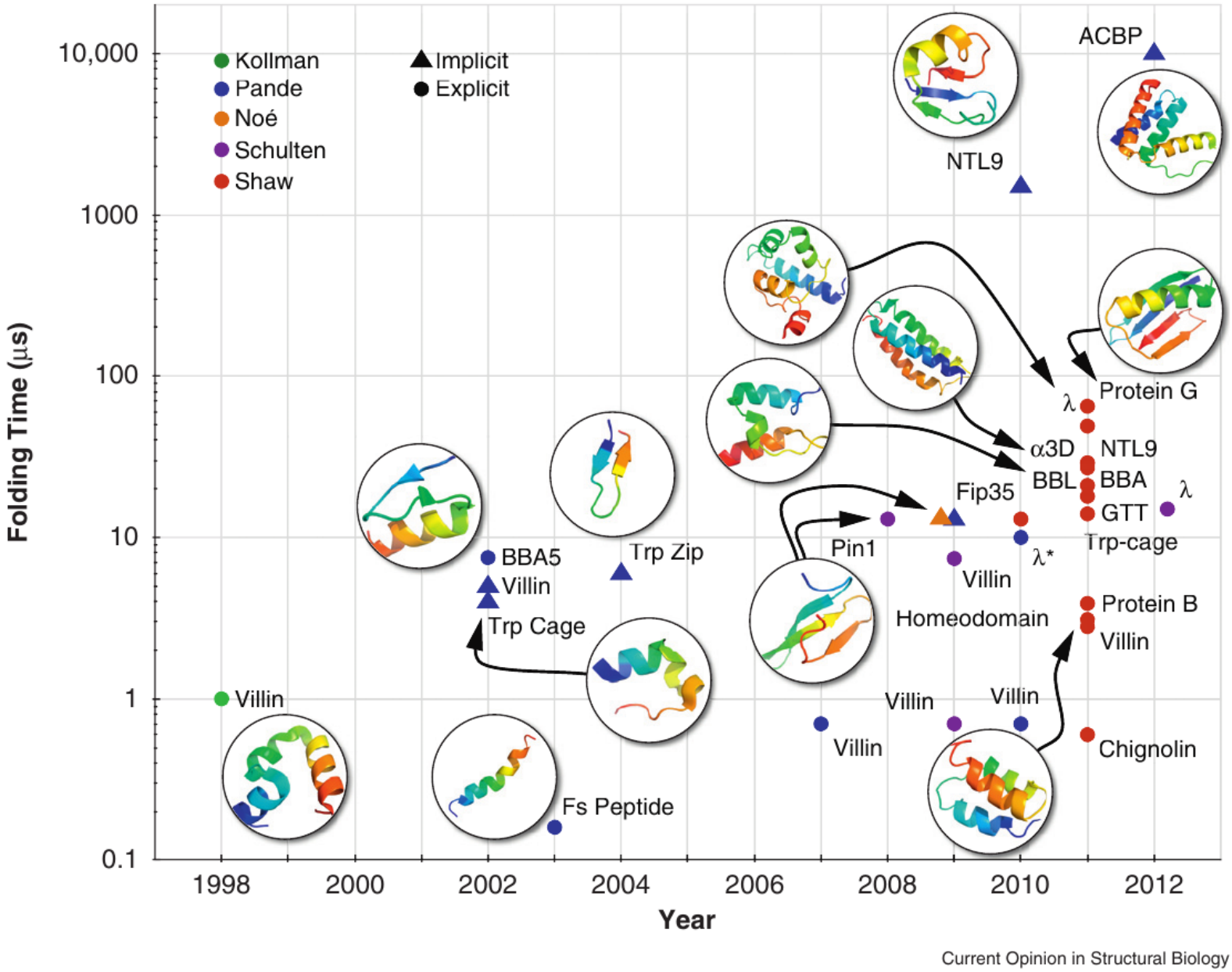
E. Free Energy Surface calculation



Temps de repliement

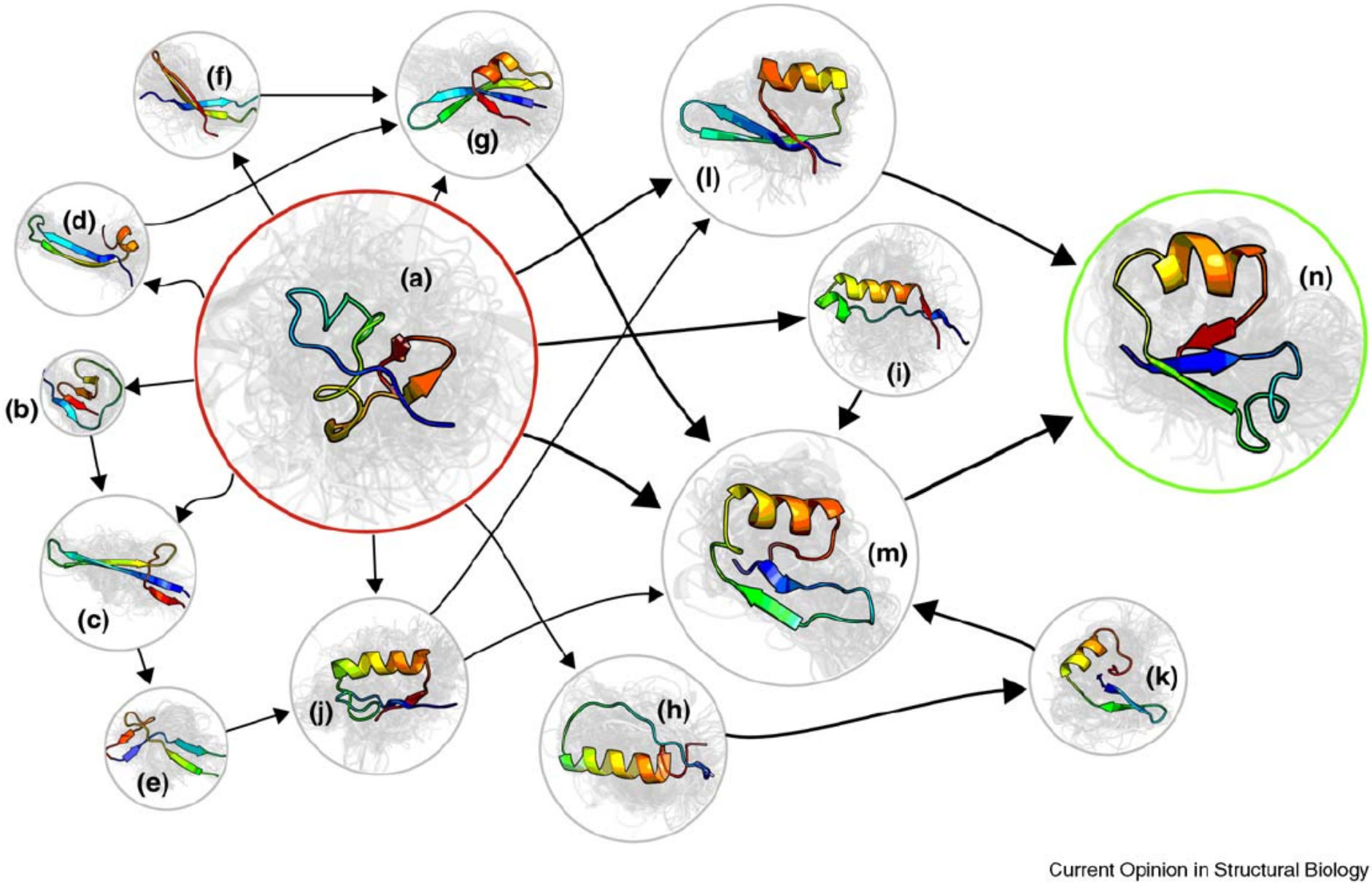


Thomas J Lane et al. (Feb. 2013). en. In: *Current Opinion in Structural Biology* 23.1

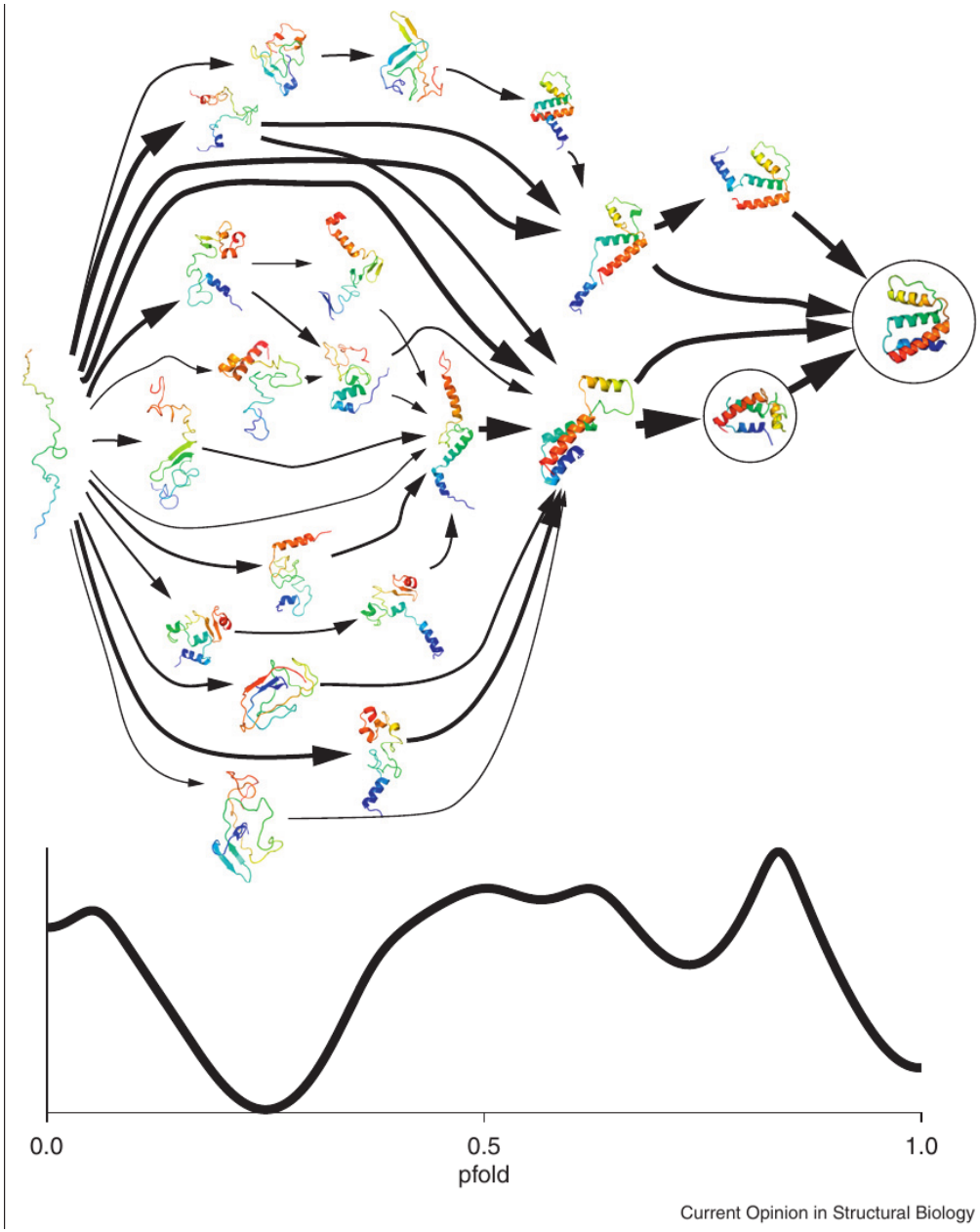


Thomas J Lane et al. (Feb. 2013). en. In: *Current Opinion in Structural Biology* 23.1

Markov State Models (MSM)



protéine NTL9, Gregory R Bowman, Vincent A Voelz, and Vijay S Pande (Feb. 2011). en. In: *Current Opinion in Structural Biology* 21.1



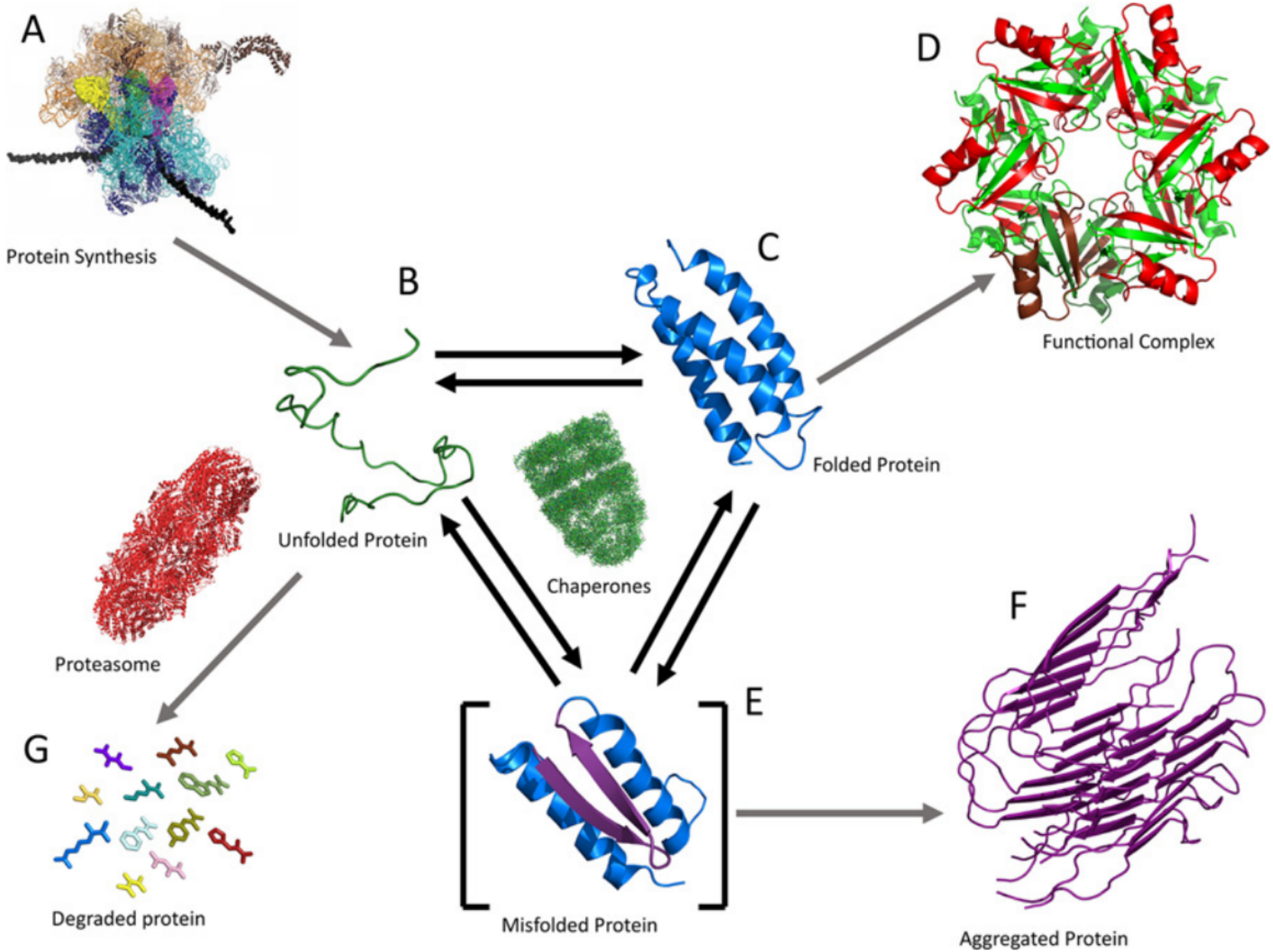
protéine ACBP,

Thomas J Lane et al. (Feb. 2013). en. In: *Current Opinion in Structural Biology* 23.1

4 Repliement in vivo

- Introduction
- Environnement cellulaire

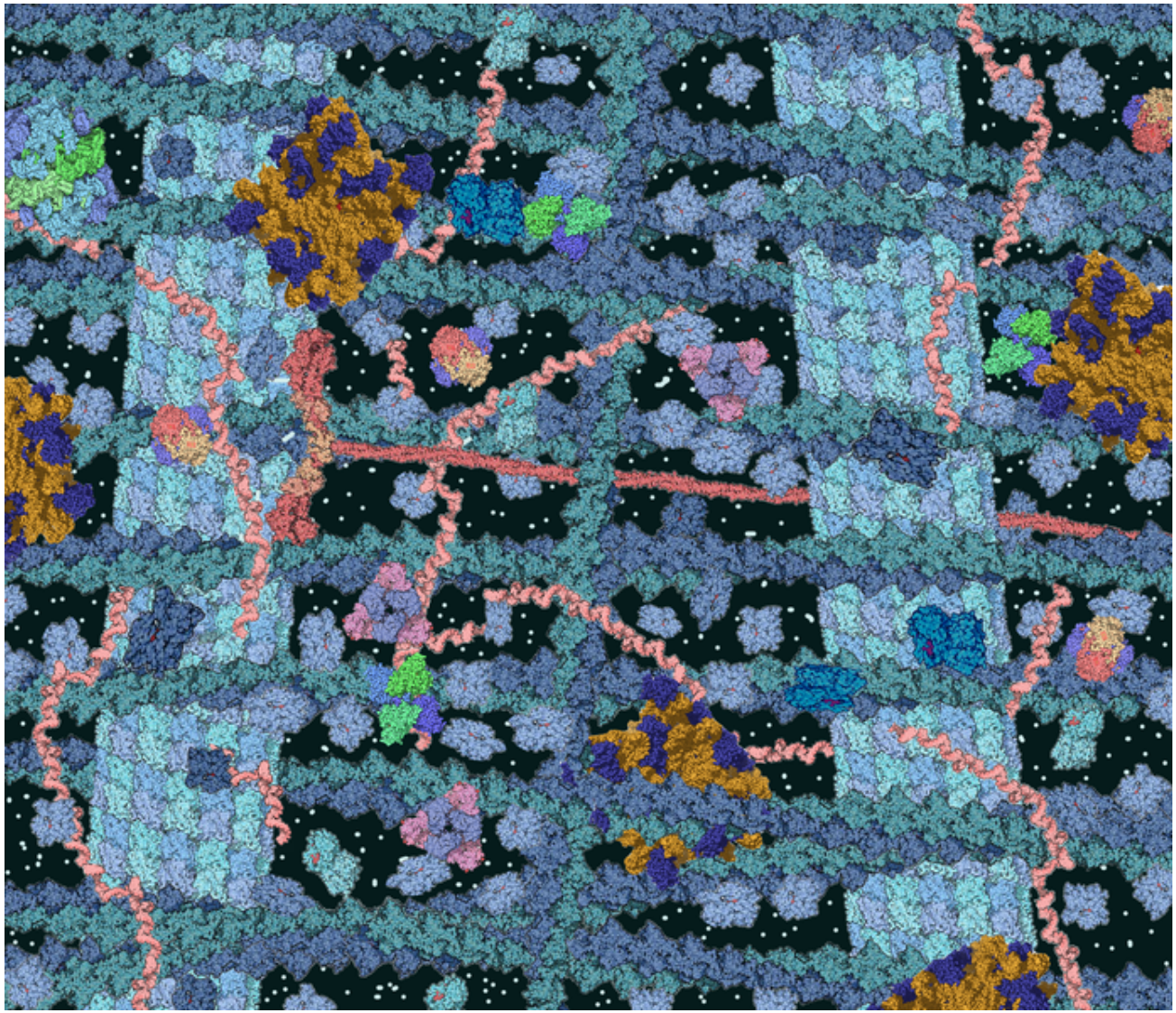
Introduction

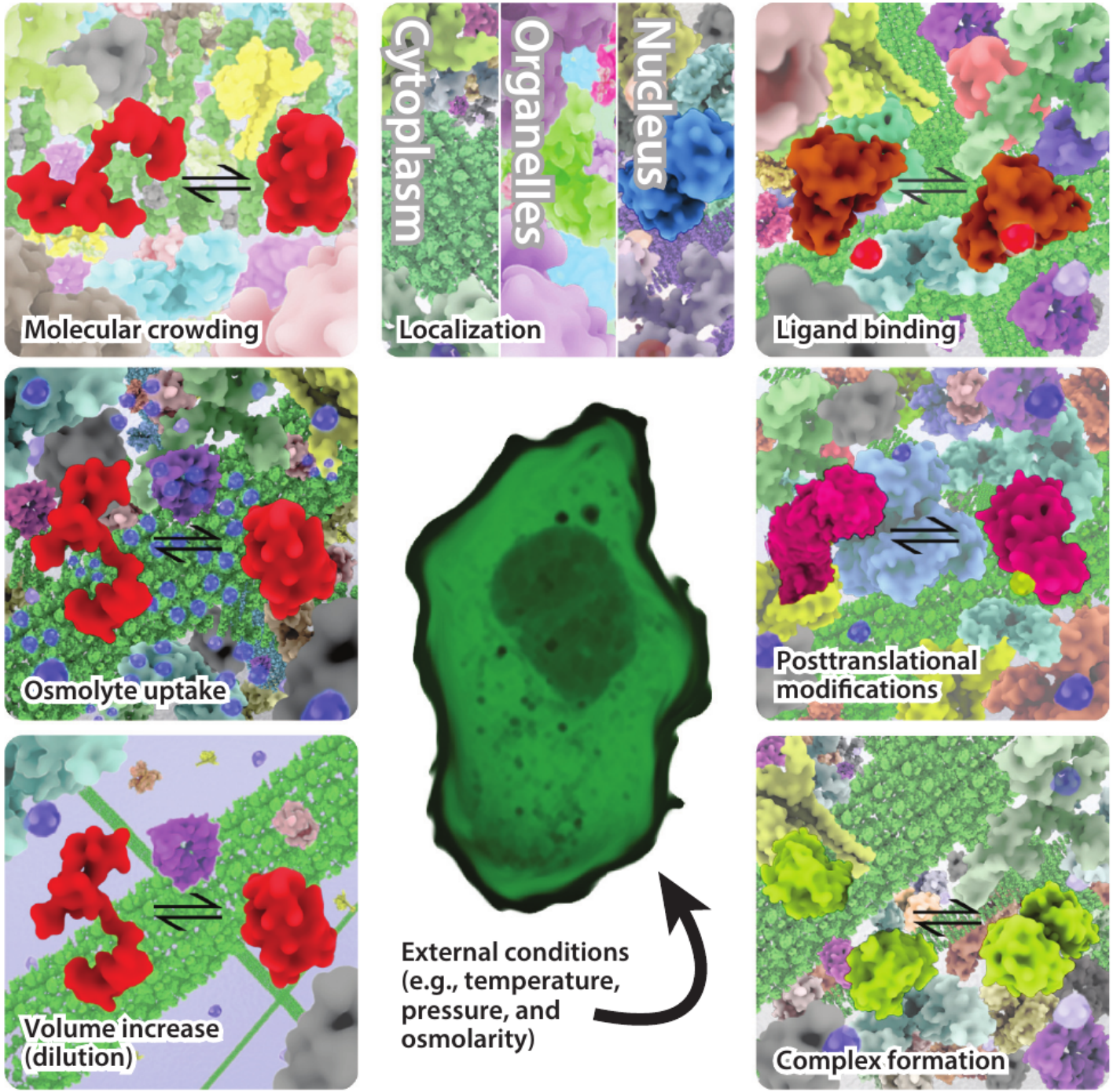


Victor Muñoz and Michele Cerminara (Sept. 2016). In: *Biochem J* 473.17

Environnement cellulaire

Molecular crowding




Molecular crowding
Cytoplasm
Organelles
Nucleus
Localization
Ligand binding
Osmolyte uptake
External conditions
(e.g., temperature,
pressure, and
osmolarity)
Posttranslational
modifications
Volume increase
(dilution)
Complex formation

5 Protein design

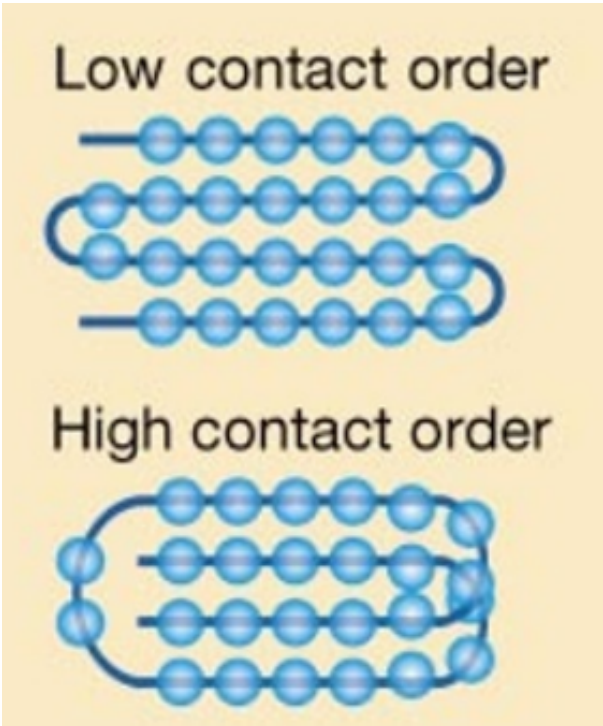
- David Baker - first work
- David Baker - 15 years of protein design
- Peptides cycliques ciblant des interactions protéine-protéine

David Baker - first work

Contact order and folding kinetics

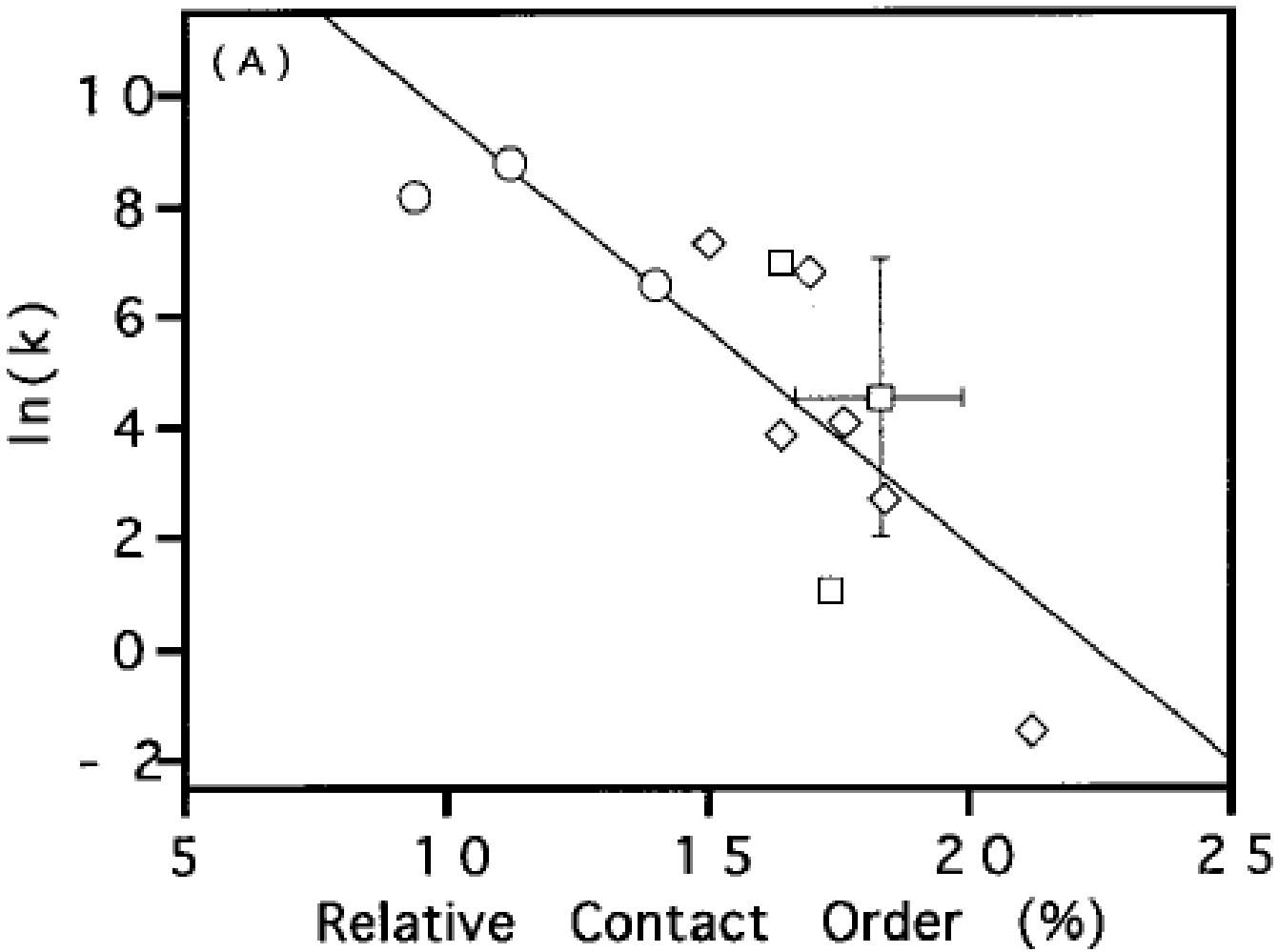
$$CO = \frac{1}{L \cdot N} \sum^{N} \Delta S_{i,j} \qquad (1)$$

where N is the total number of contacts, $\Delta S_{i,j}$ is the sequence separation, in residues, between contacting residues i and j, and L is the total number of residues in the protein.



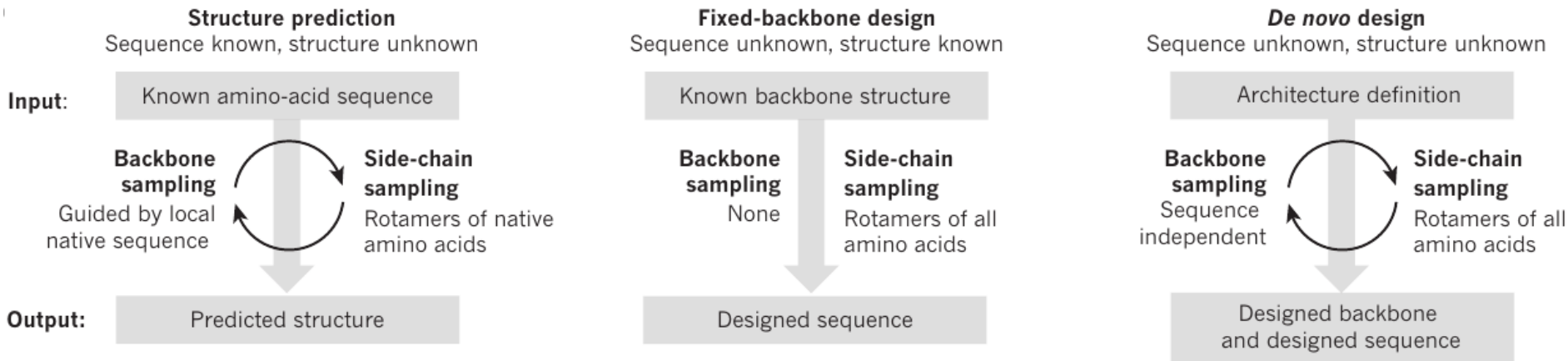
Kevin W Plaxco, Kim T Simons, and David Baker (Apr. 1998). In: *Journal of Molecular Biology* 277.4

Contact order and folding kinetics



Kevin W Plaxco, Kim T Simons, and David Baker (Apr. 1998). In: *Journal of Molecular Biology* 277.4

De novo protein design



Po-Ssu Huang, Scott E. Boyken, and David Baker (Sept. 2016). en. In: *Nature* 537.7620

Protein design - first sucess story

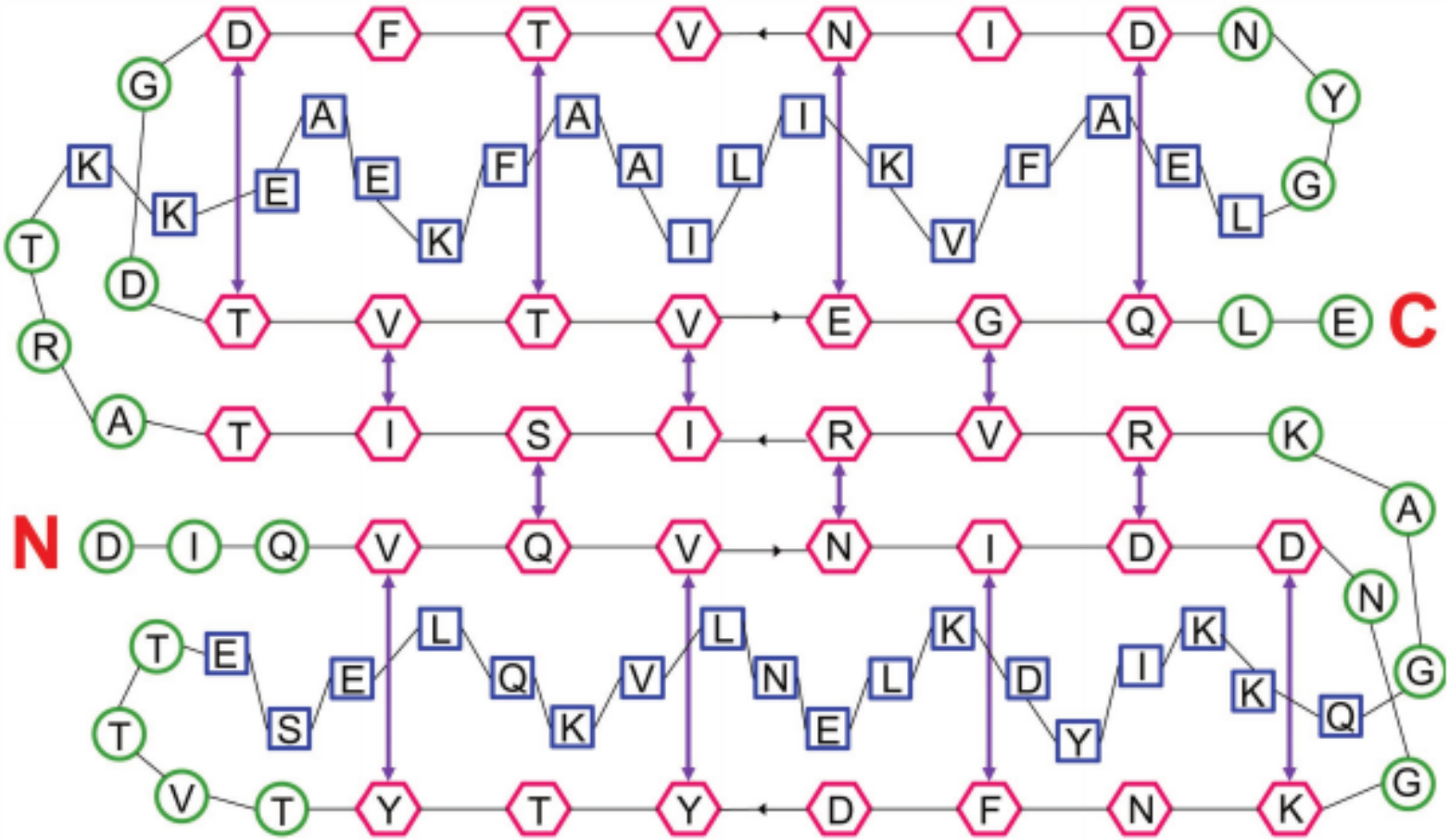
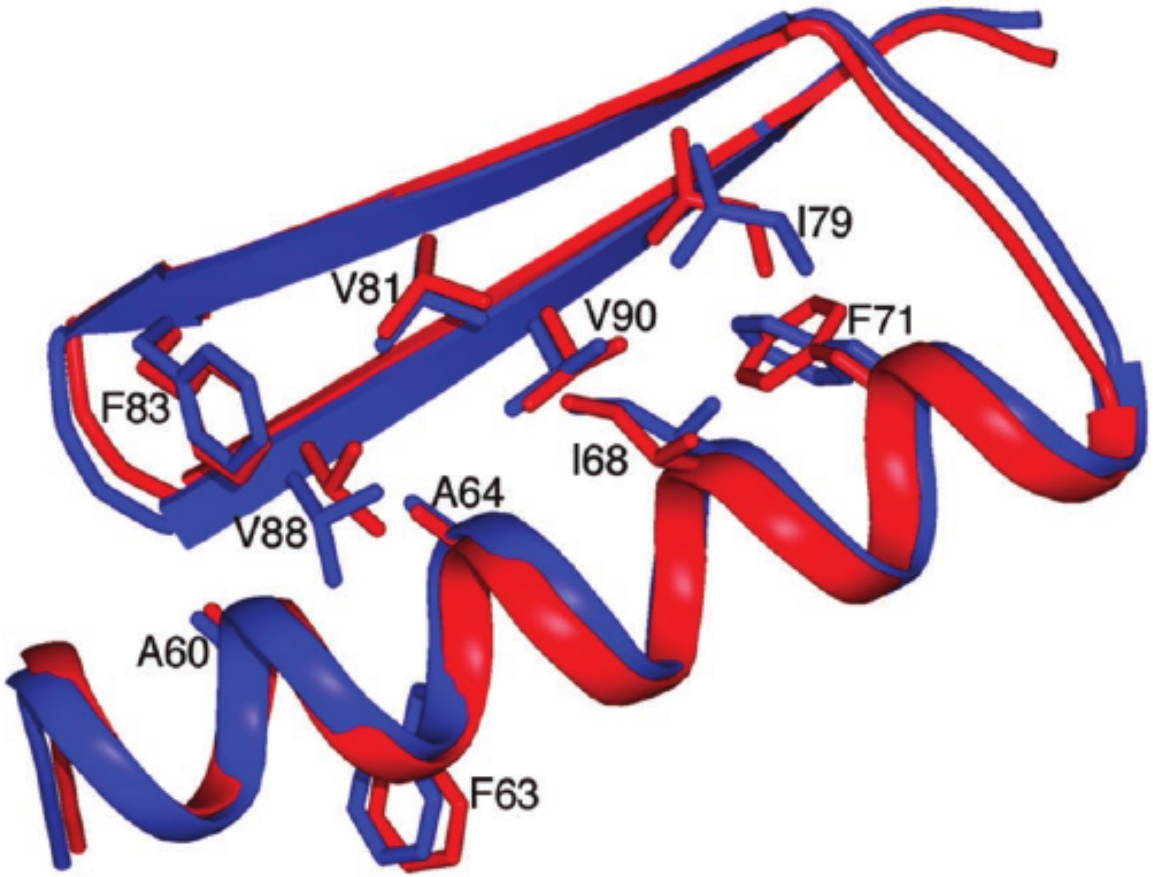


Fig. 1. A two-dimensional schematic of the target fold (hexagon, strand; square, helix; circle, other). Hydrogen bond partners are shown as purple arrows. The amino acids shown are those in the final designed (Top7) sequence.

Brian Kuhlman et al. (Nov. 2003). en. In: *Science* 302.5649

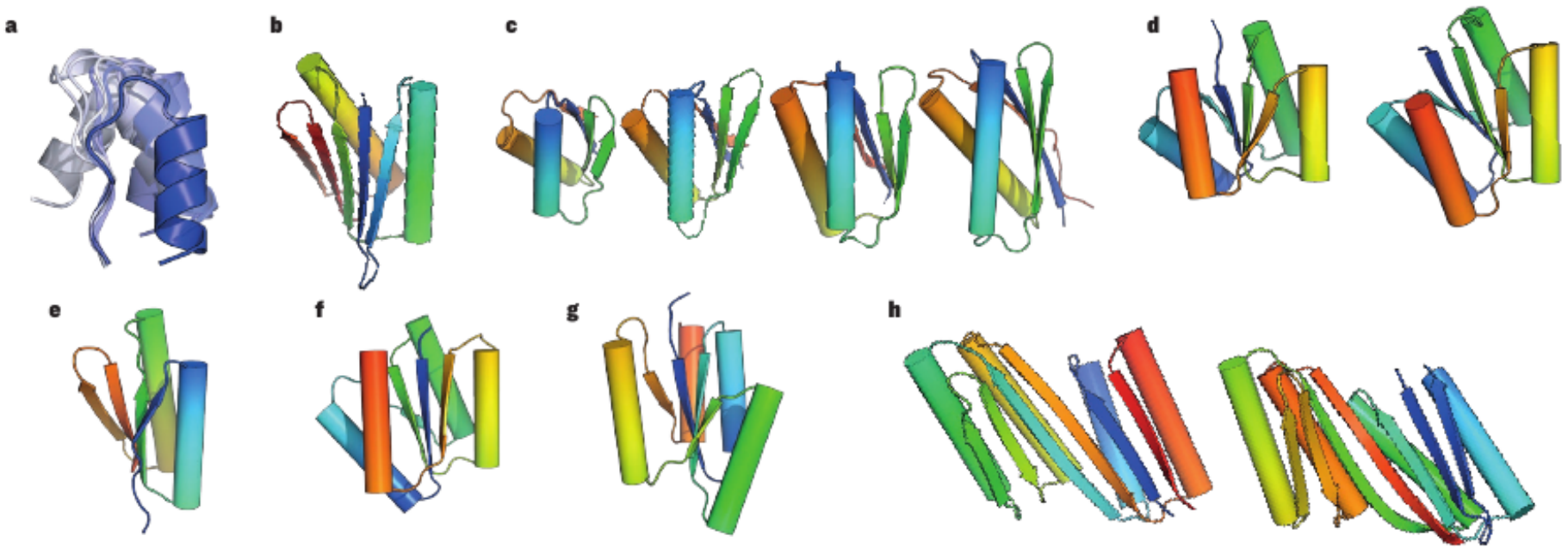
Protein design - first sucess story



Brian Kuhlman et al. (Nov. 2003). en. In: *Science* 302.5649

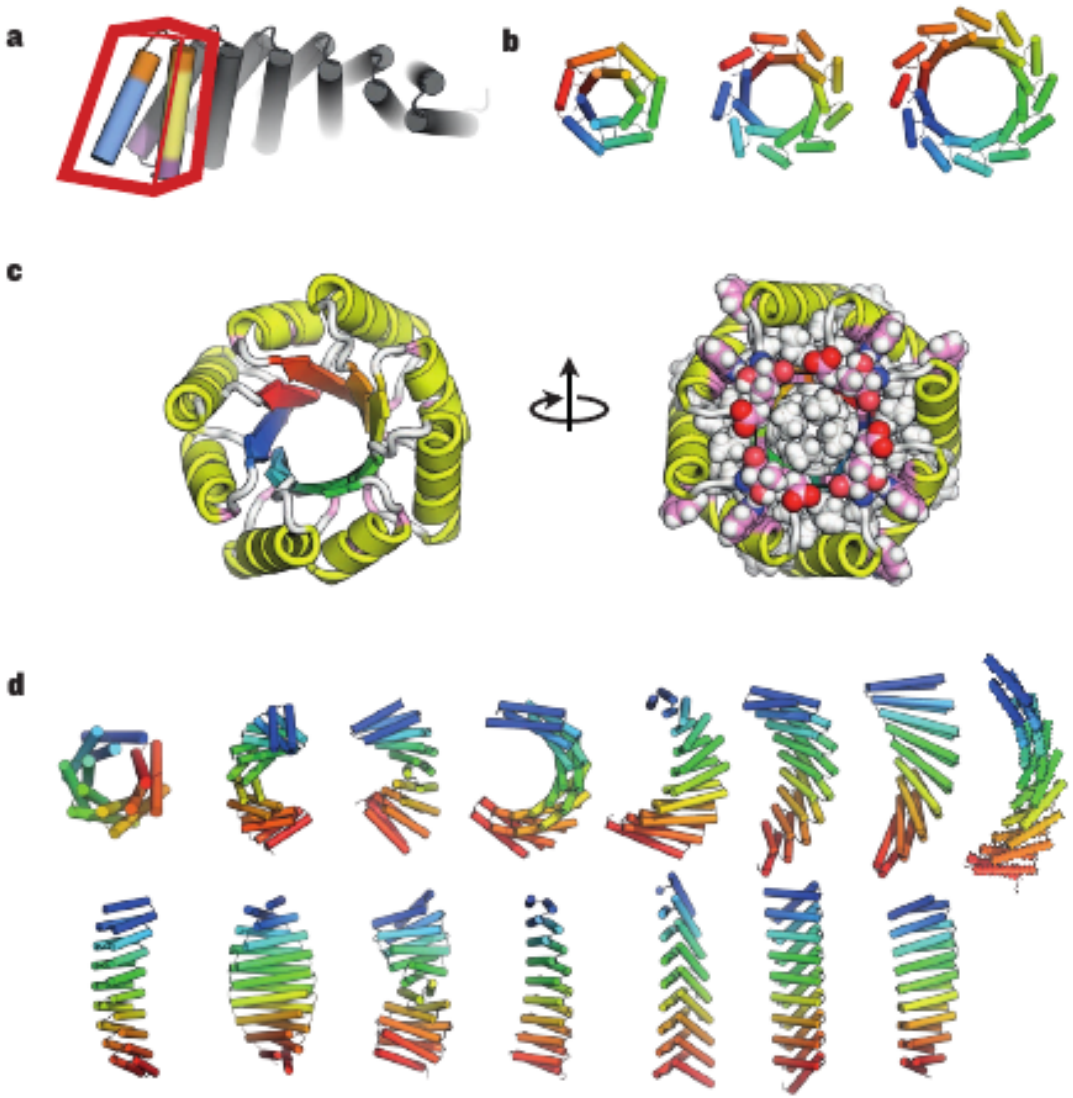
David Baker - 15 years of protein design

Protein design - examples



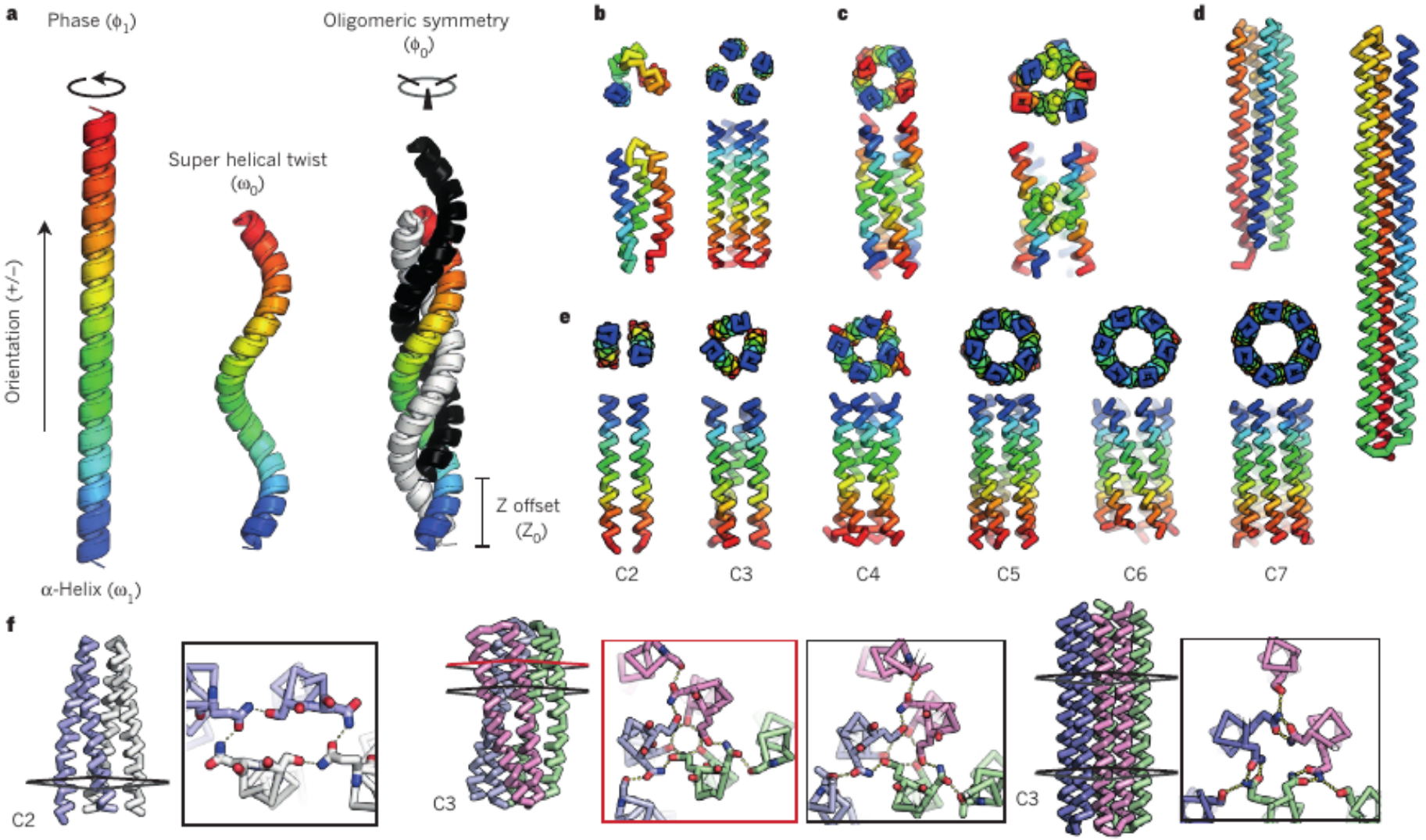
Po-Ssu Huang, Scott E. Boyken, and David Baker (Sept. 2016). en. In: *Nature* 537.7620

Protein design - examples



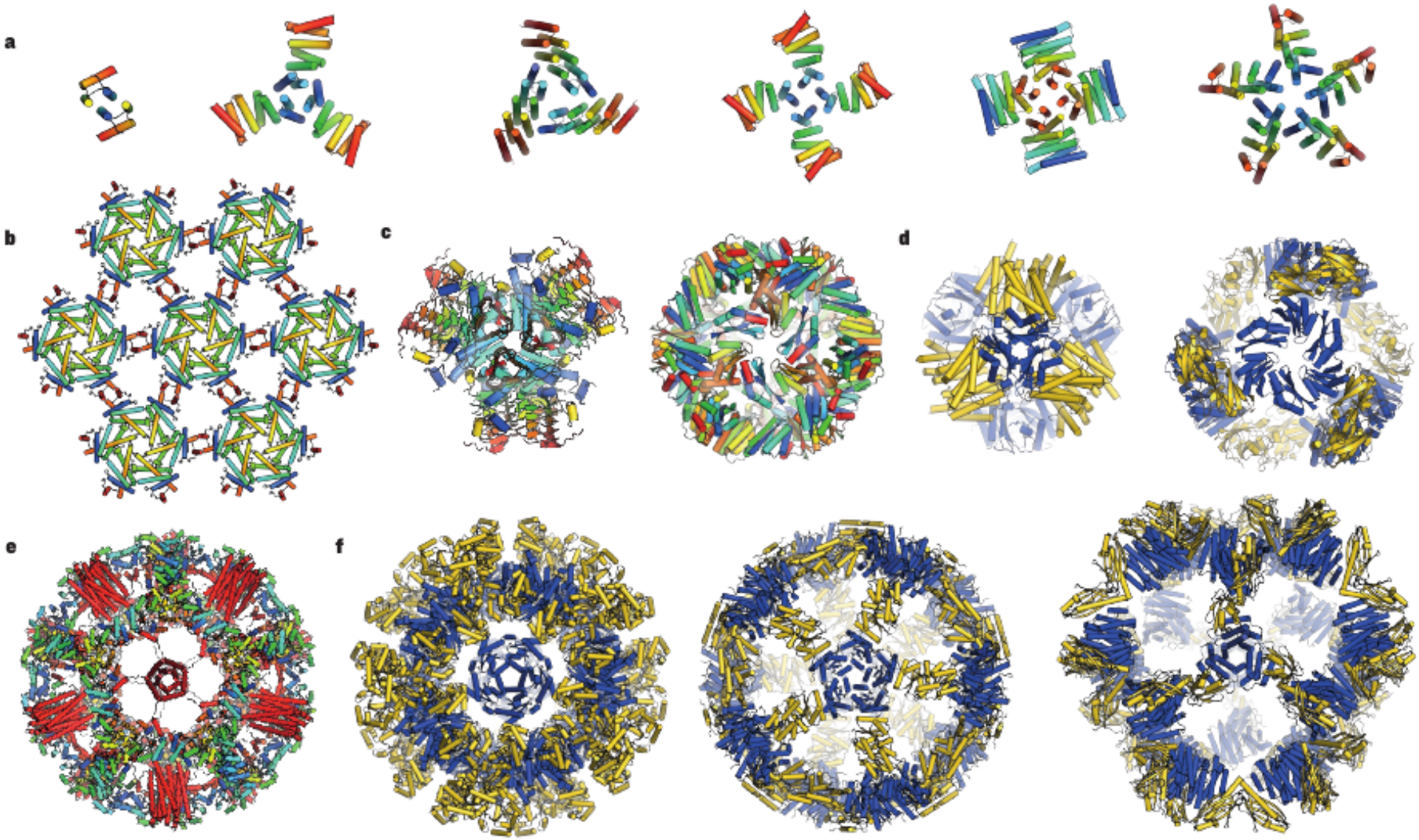
Po-Ssu Huang, Scott E. Boyken, and David Baker (Sept. 2016). en. In: *Nature* 537.7620

Protein design - examples



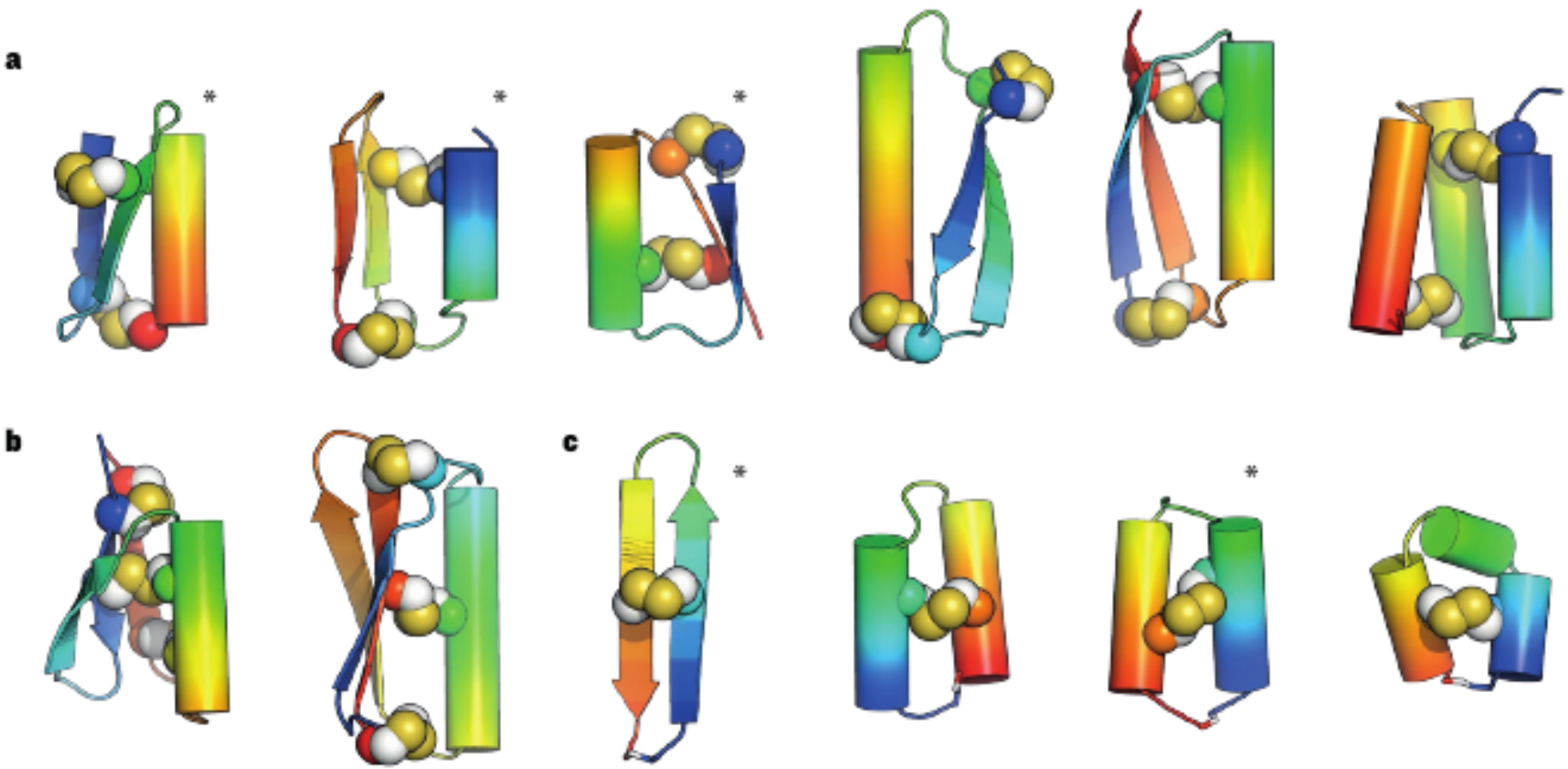
Po-Ssu Huang, Scott E. Boyken, and David Baker (Sept. 2016). en. In: *Nature* 537.7620

Protein design - examples



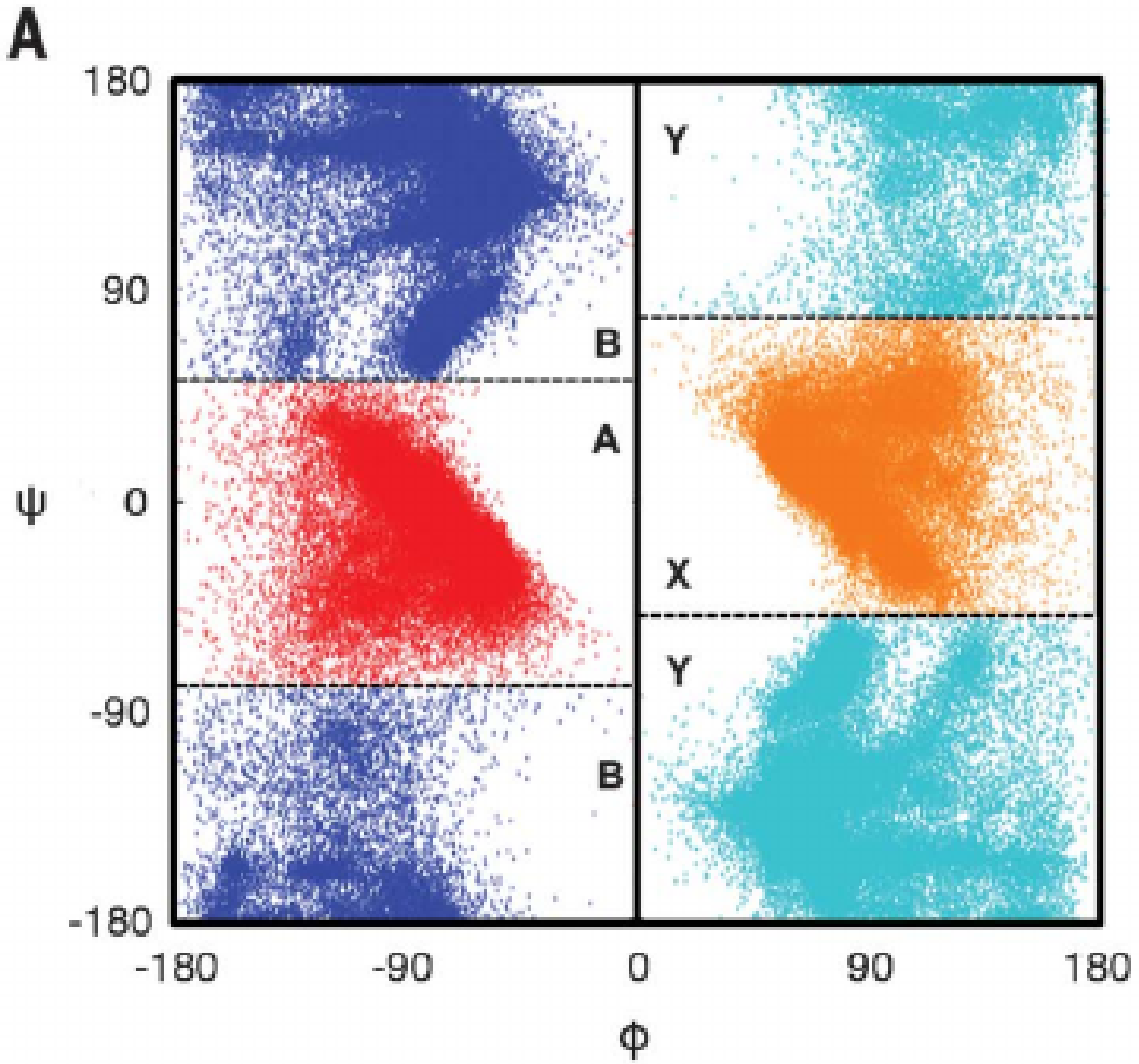
Po-Ssu Huang, Scott E. Boyken, and David Baker (Sept. 2016). en. In: *Nature* 537.7620

Protein design - examples



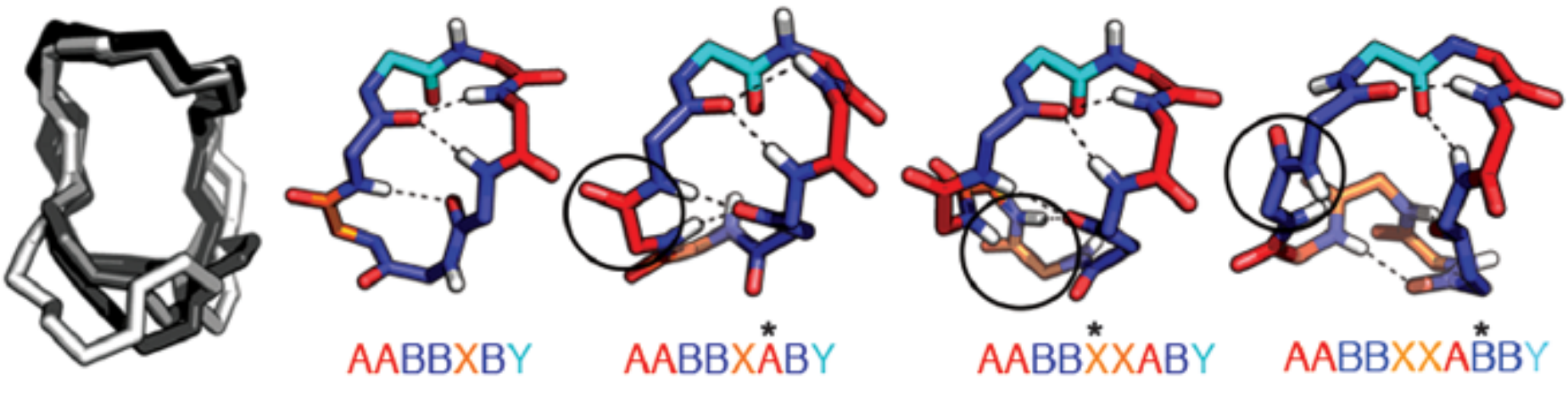
Po-Ssu Huang, Scott E. Boyken, and David Baker (Sept. 2016). en. In: *Nature* 537.7620

Protein design - macrocycles



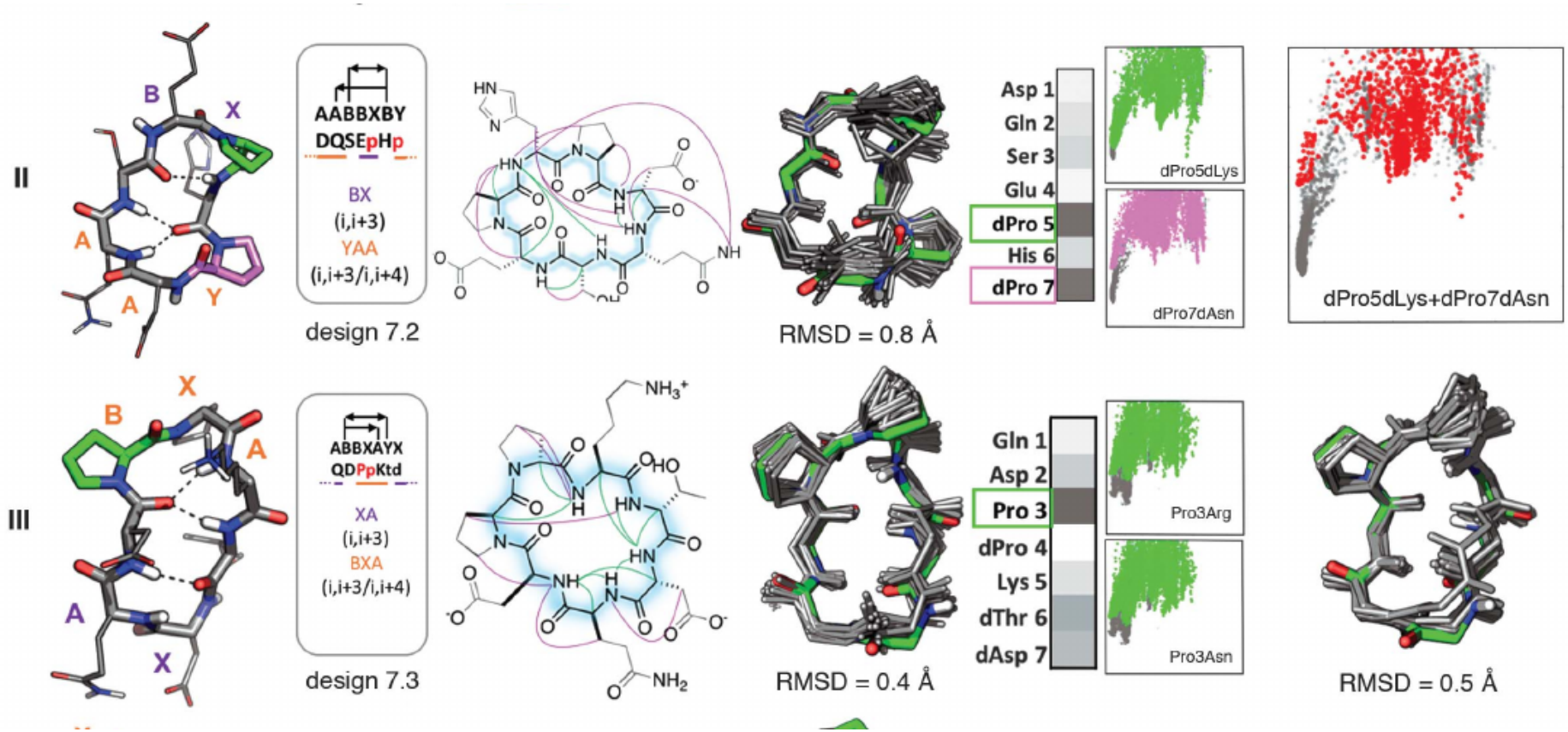
Parisa Hosseinzadeh et al. (Dec. 2017). en. In: *Science* 358.6369

Protein design - macrocycles



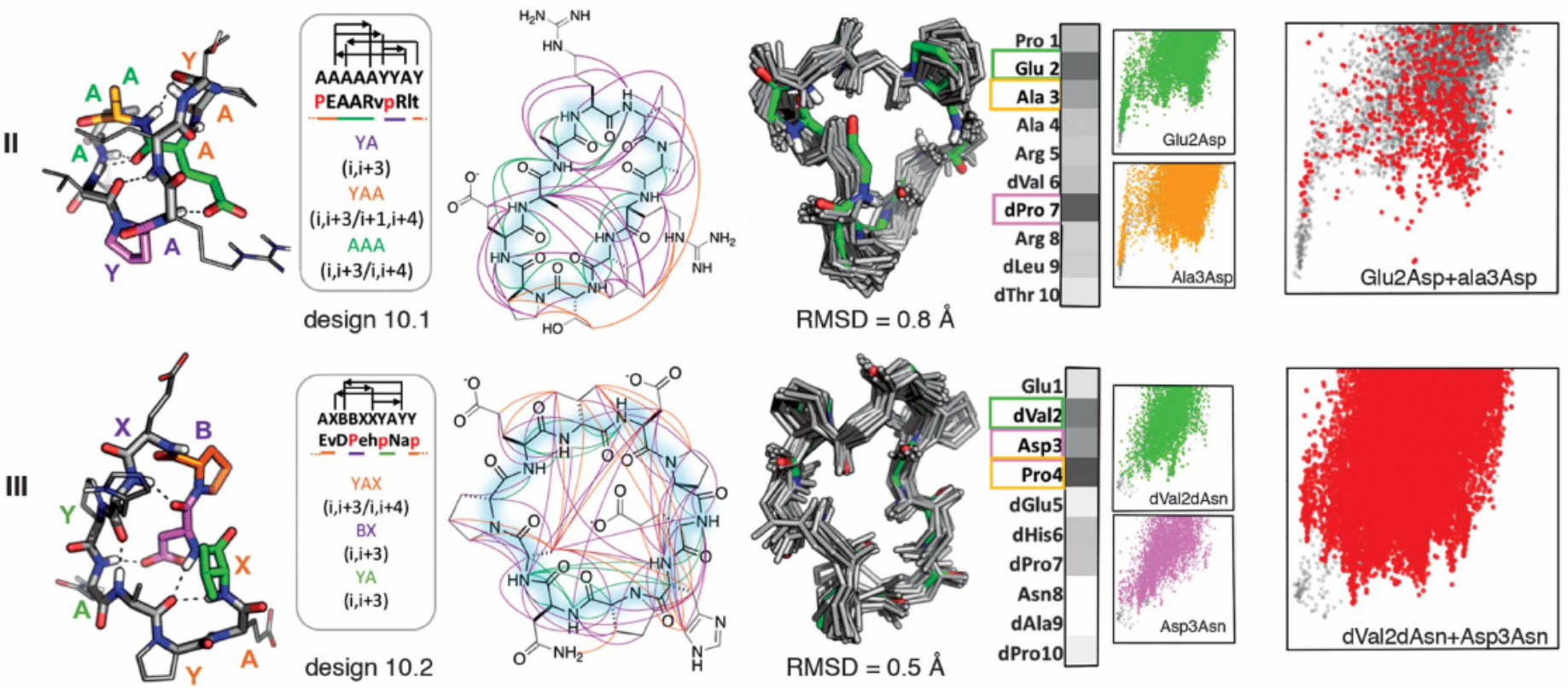
Parisa Hosseinzadeh et al. (Dec. 2017). en. In: *Science* 358.6369

Protein design - macrocycles



Parisa Hosseinzadeh et al. (Dec. 2017). en. In: *Science* 358.6369

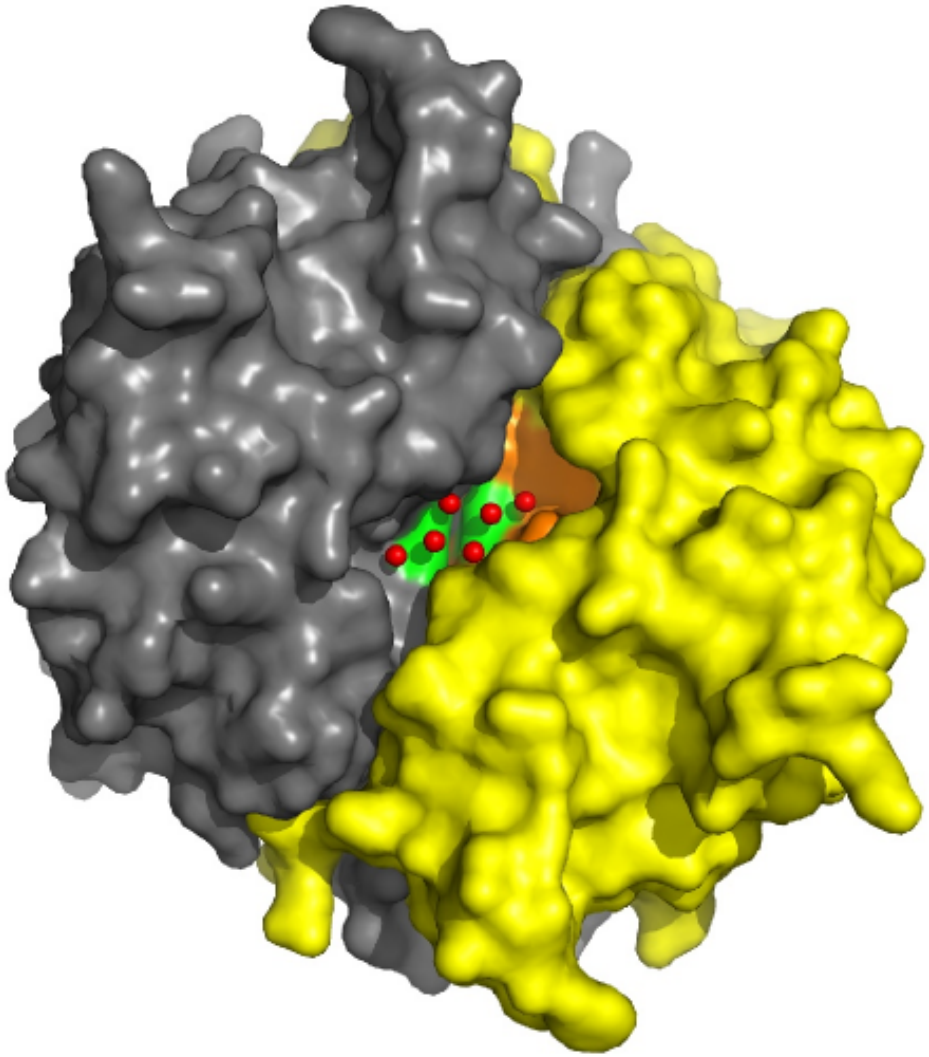
Protein design - macrocycles



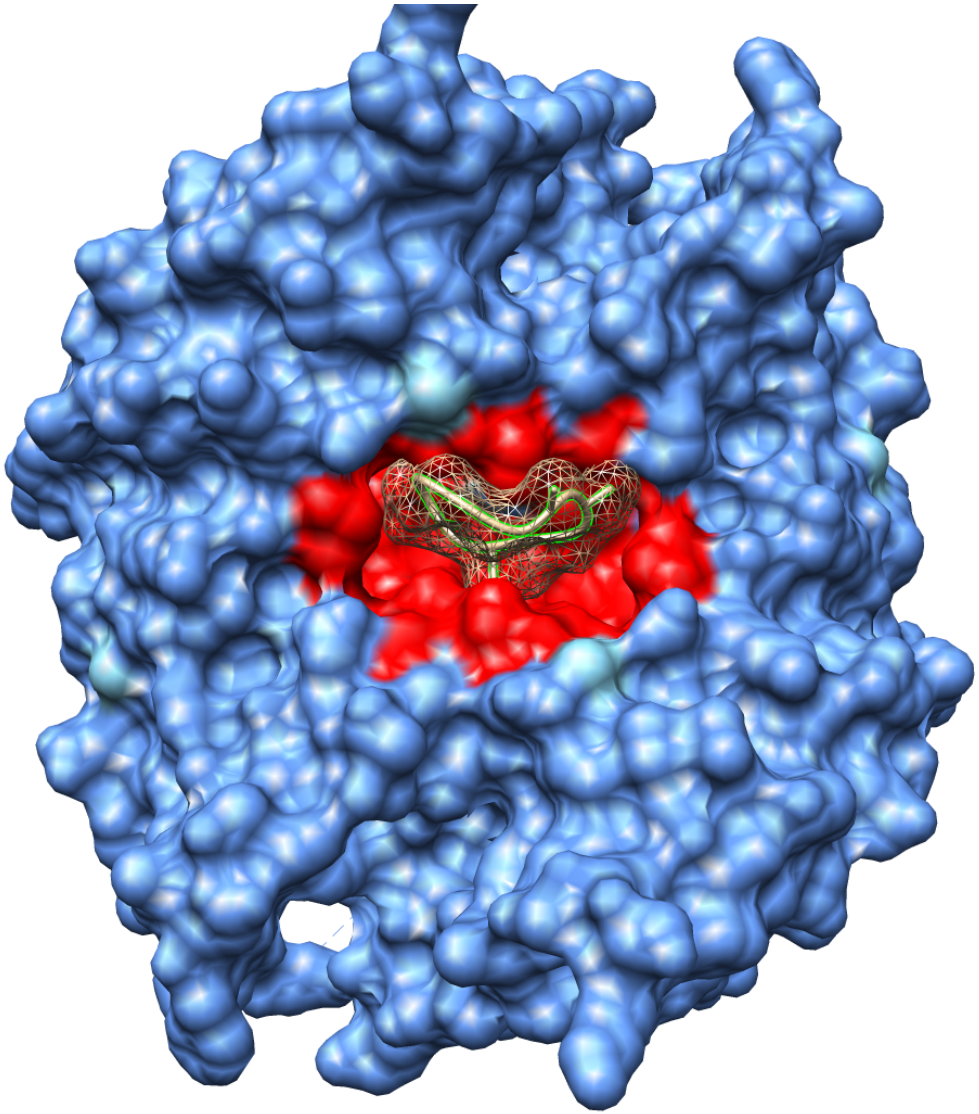
Parisa Hosseinzadeh et al. (Dec. 2017). en. In: *Science* 358.6369

Peptides cycliques ciblant des interactions protéine-protéine

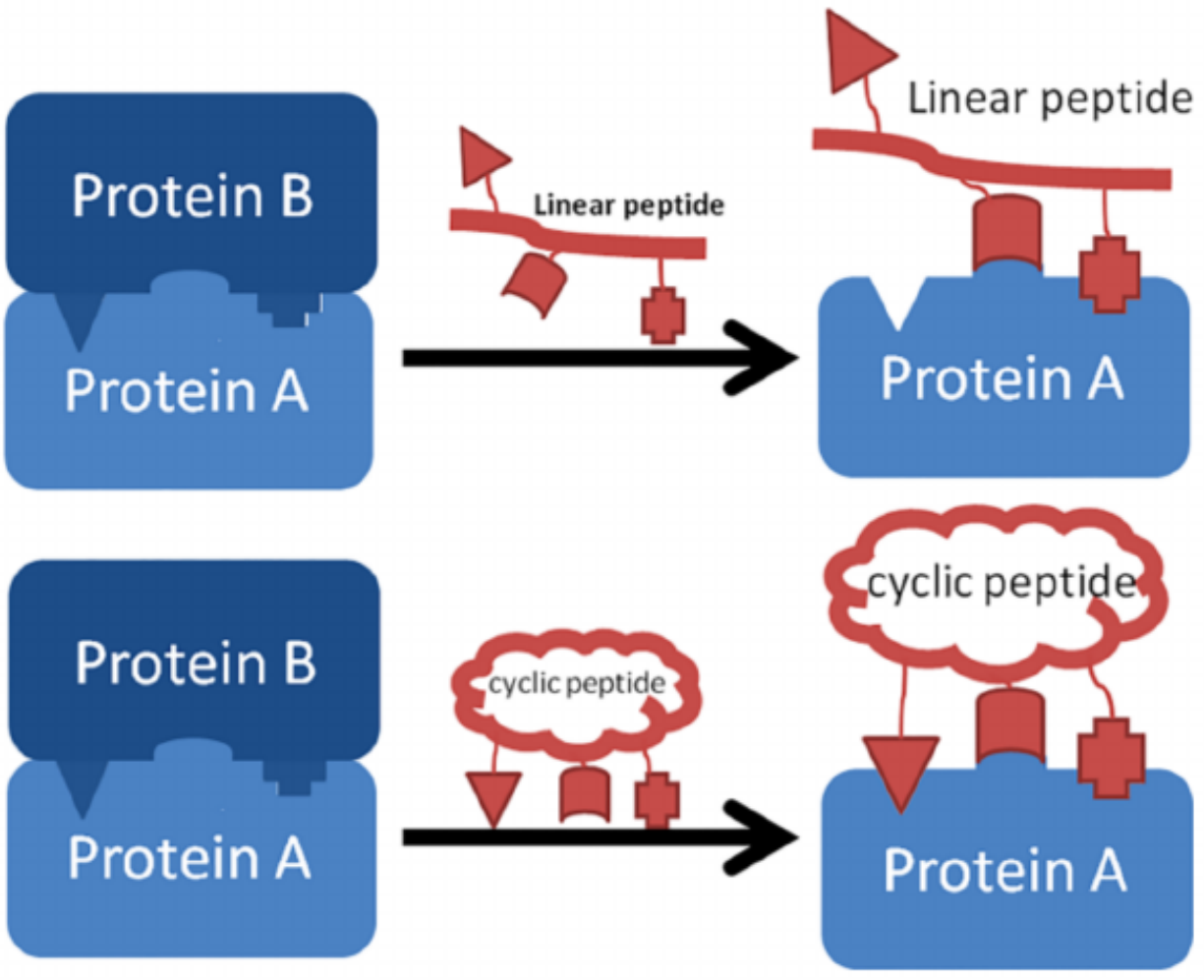
Drug design: cibler les interactions protéine-protéine



Drug = peptide cyclique



Drug = peptide cyclique



6 Alpha Fold

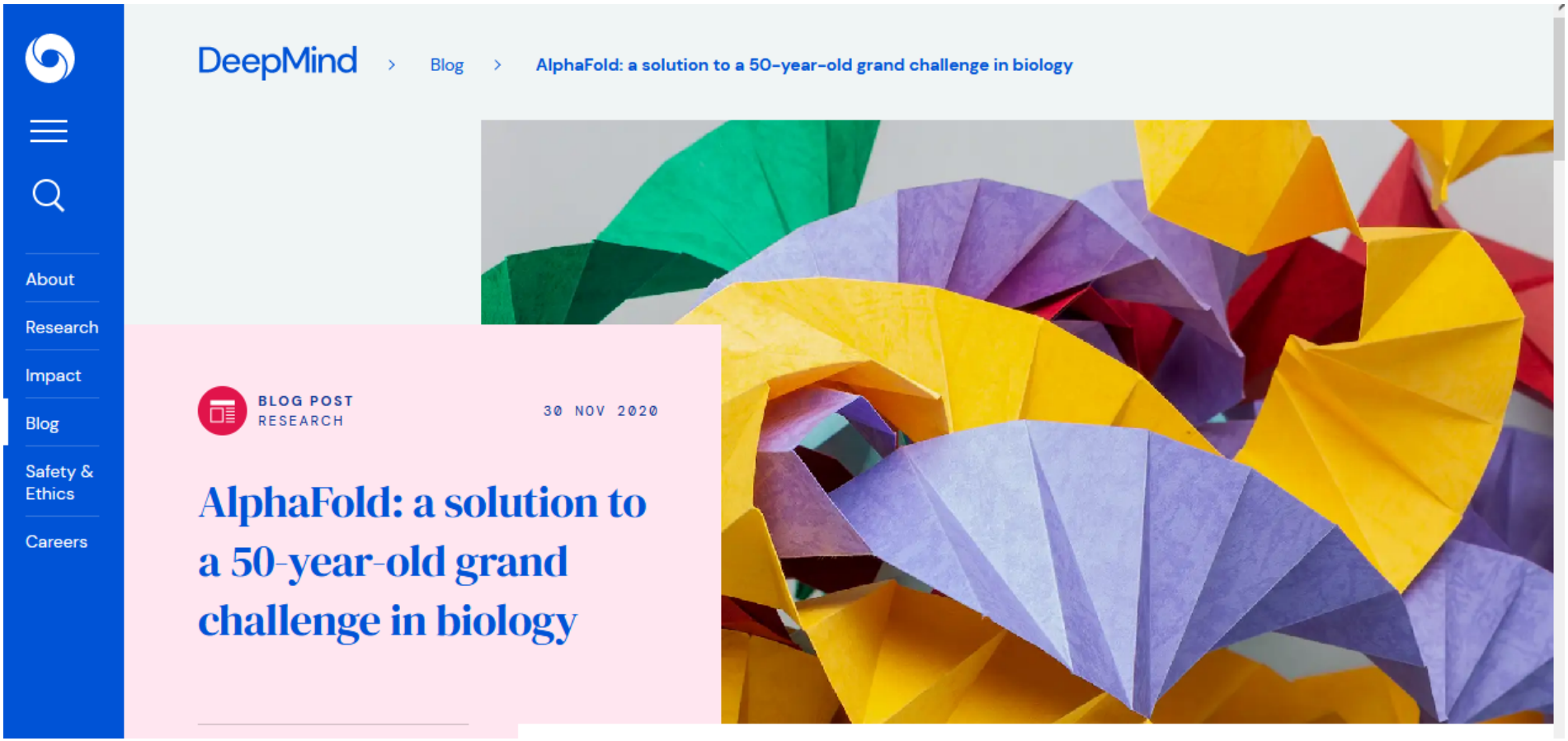
- Alpha Fold - La révolution
- Applications de Alpha Fold
- Autres méthodes utilisant le deep learning
- Comprendre Alpha Fold
- Mutations corrélées, MSA
- Utiliser Alpha Fold
- Pour aller plus loin: EMBL webinar
- Validation et impact d'AlphaFold en 2024
- AlphaFold3 et co.

Alpha Fold - La révolution

Le press release de CASP du 30 nov 2020

"Artificial intelligence solution to a 50-year-old science challenge could ‘revolutionise’ medical research"
"Today (Monday) researchers at the 14th Community Wide Experiment on the Critical Assessment of Techniques for Protein Structure Prediction (CASP14) will announce that an artificial intelligence (AI) solution to the challenge has been found."
"Nearly 50 years ago, Christian Anfinsen was awarded a Nobel Prize for showing that it should be possible to determine the shape of proteins based on their sequence of amino acids – the individual building blocks that make up proteins. That’s why our community of scientists have been working on the biennial CASP challenge."
`https://predictioncenter.org/casp14/doc/CASP14_press_release.html`

DeepMind de google basé à Londres



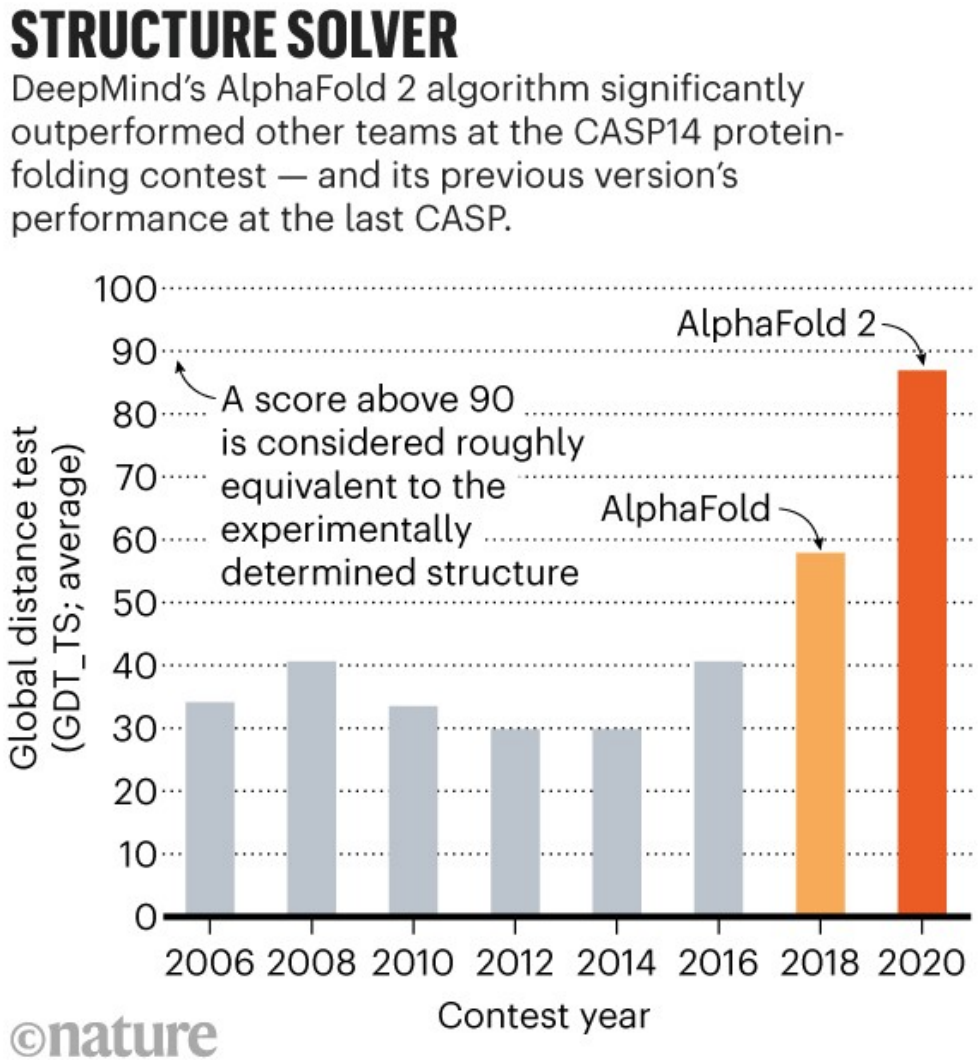
Le relais immédiat dans Nature de l'annonce de CASP

30 nov 2020, Nature, "'It will change everything': DeepMind's AI makes gigantic leap in solving protein structures"
https://www.nature.com/articles/d41586-020-03348-4

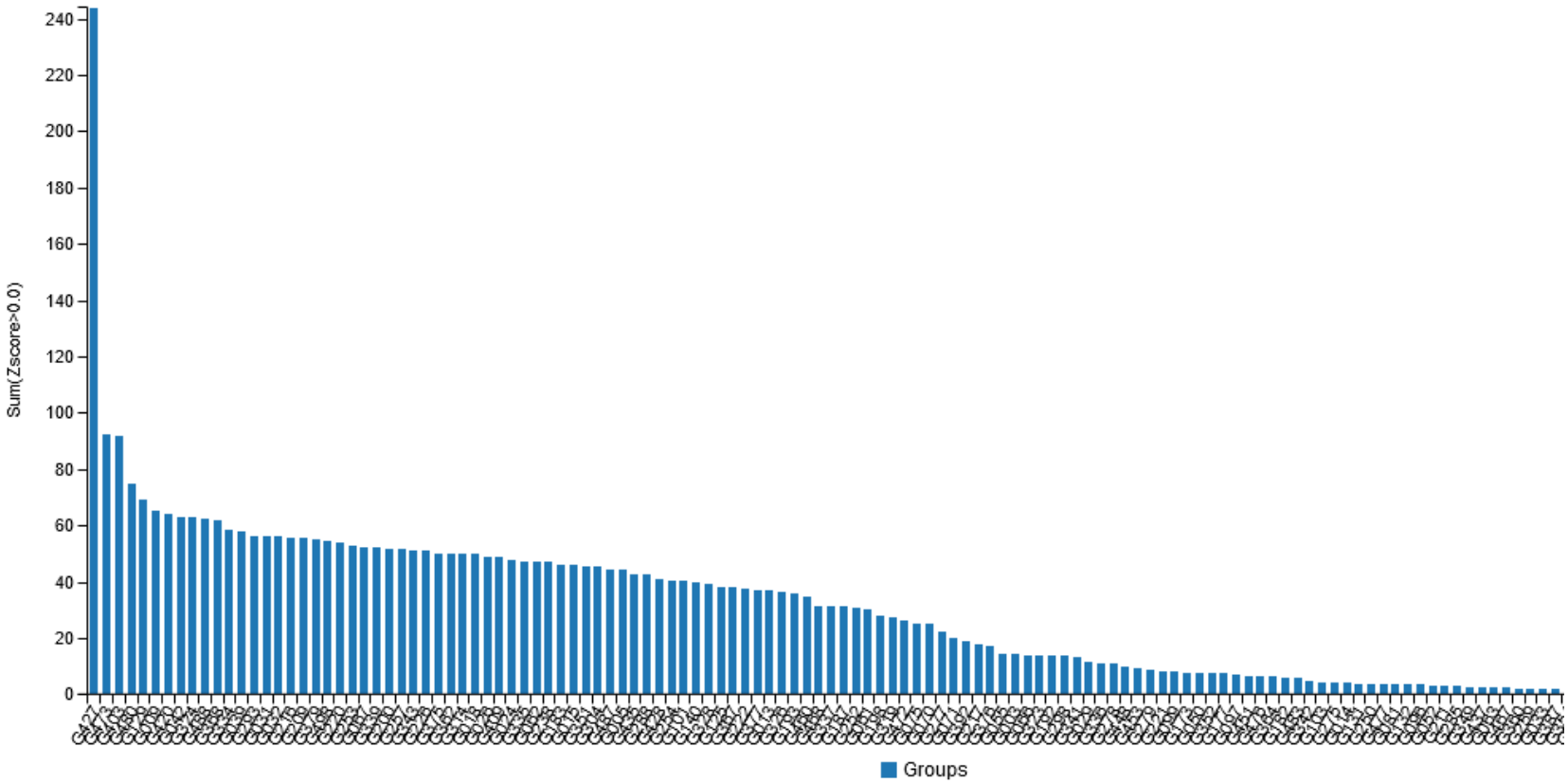
La presse

30 nov 2020, The New York Times, London AI claims breakthrough that could accelerate drug discovery
1 déc 2020, France Culture, Le "problème de repliement des protéines" résolu par une intelligence artificielle
11 déc 2020, Les Échos, DeepMind met les chercheurs du monde entier au tapis

CASP 14 et AlphaFold 2

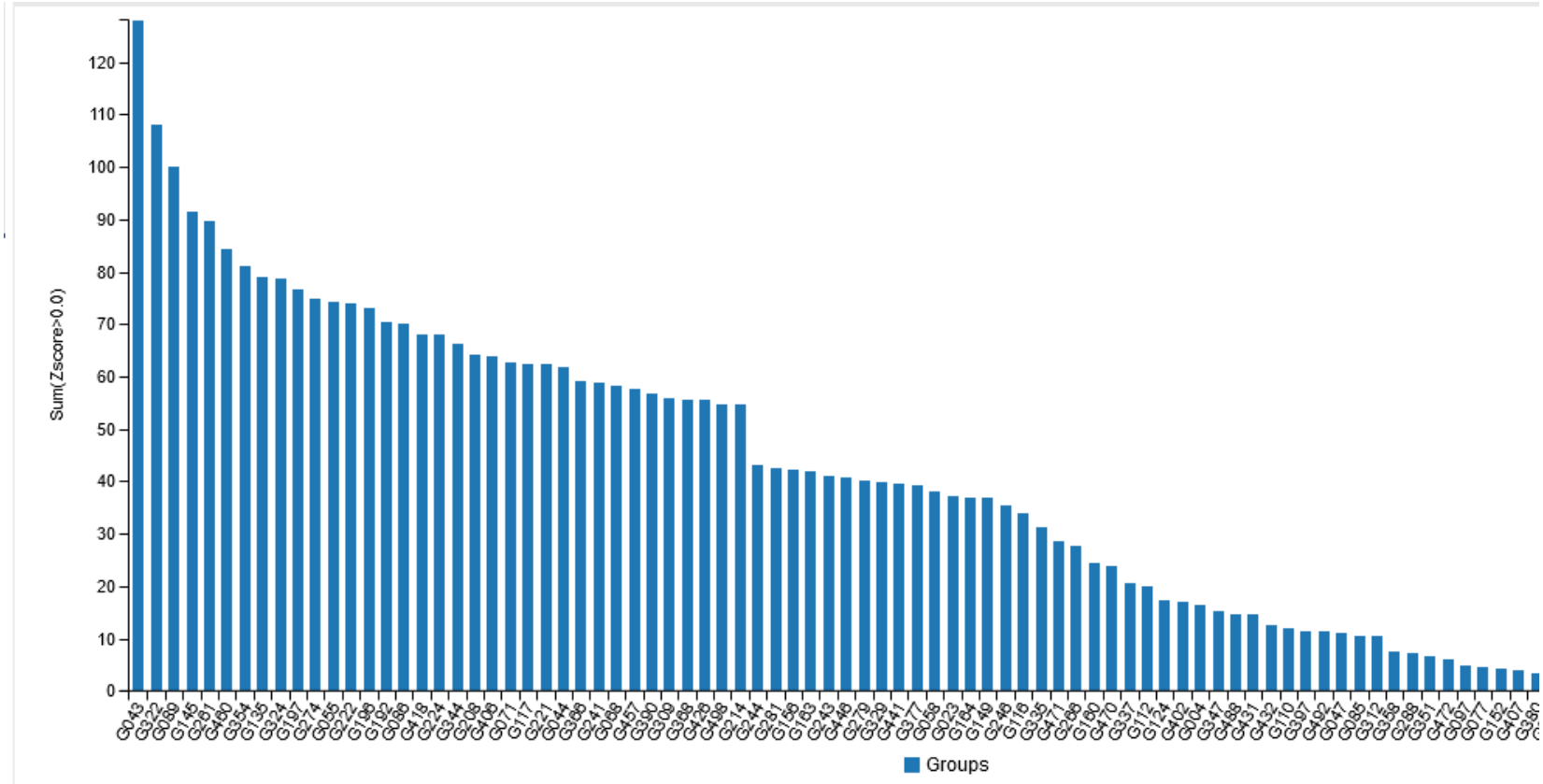


CASP 14 et AlphaFold 2



#	GR code	GR name	Domains Count	SUM Zscore (>-2.0)	Rank SUM Zscore (>-2.0)	AVG Zscore (>-2.0)	Rank AVG Zscore (>-2.0)	SUM Zscore (>0.0)	Rank SUM Zscore (>0.0)
1	427	AlphaFold2	92	244.0217	1	2.6524	1	244.0217	1
2	473	BAKER	92	90.8241	2	0.9872	2	92.1241	2
3	403	BAKER-experimental	92	88.9672	3	0.9670	3	91.4731	3
4	480	FEIG-R2	92	72.5351	4	0.7884	4	74.5627	4
5	129	Zhang	92	67.9065	5	0.7381	5	68.8922	5

CASP 13 et AlphaFold 1

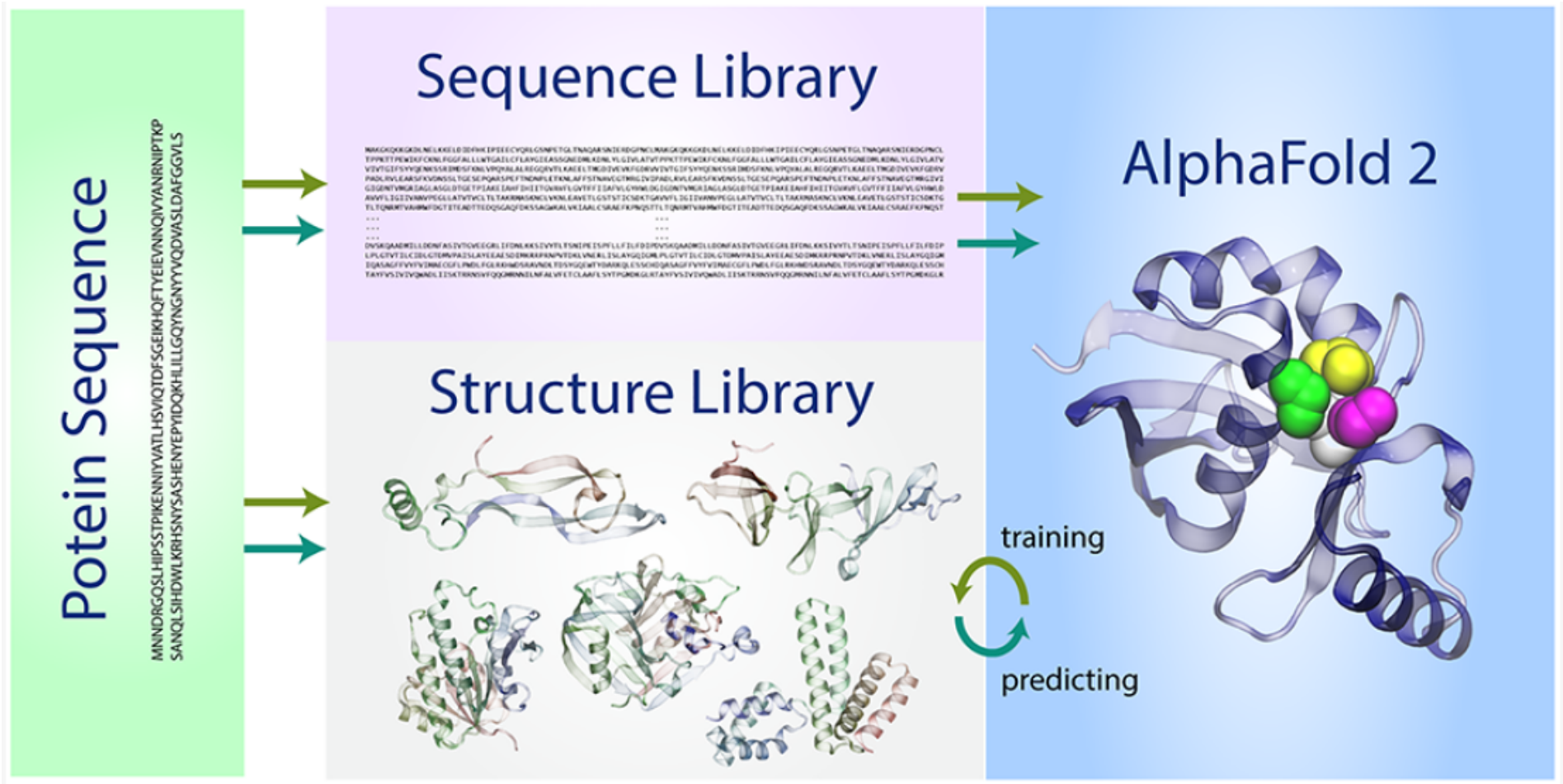


#	GR code	GR name	Domains Count	SUM Zscore (>-2.0)	Rank SUM Zscore (>-2.0)	AVG Zscore (>-2.0)	Rank AVG Zscore (>-2.0)	SUM Zscore (>0.0)	Rank SUM Zscore (>0.0)
1	043	A7D	104	120.4307	1	1.1580	1	128.0693	1
2	322	Zhang	104	107.5948	2	1.0346	2	108.1948	2
3	089	MULTICOM	104	99.4661	3	0.9564	3	99.9886	3
4	145	QUARK	104	90.9915	4	0.8749	4	91.5625	4
5	261	Zhang-Server	104	88.9540	5	0.8553	5	89.7597	5

La presse en 2021

3 oct 2021, Forbes: "AlphaFold is the most important achievement in AI - Ever"
https://www.forbes.com/sites/robtoews/2021/10/03/alphafold-is-the-most-important-achievement-in-ai-ever/?sh=2857ff656e0a
22 juillet 2021, Fortune: "In giant leap for biology, DeepMind's A.I. reveals secret building blocks of human life"
https://fortune.com/2021/07/22/deepmind-alphafold-human-proteome-database-proteins/

Principe de Alpha Fold 2



15 juillet 2021: Détails de la méthode et code source

Article

Highly accurate protein structure prediction with AlphaFold

https://doi.org/10.1038/s41586-021-03819-2

Received: 11 May 2021

Accepted: 12 July 2021

Published online: 15 July 2021

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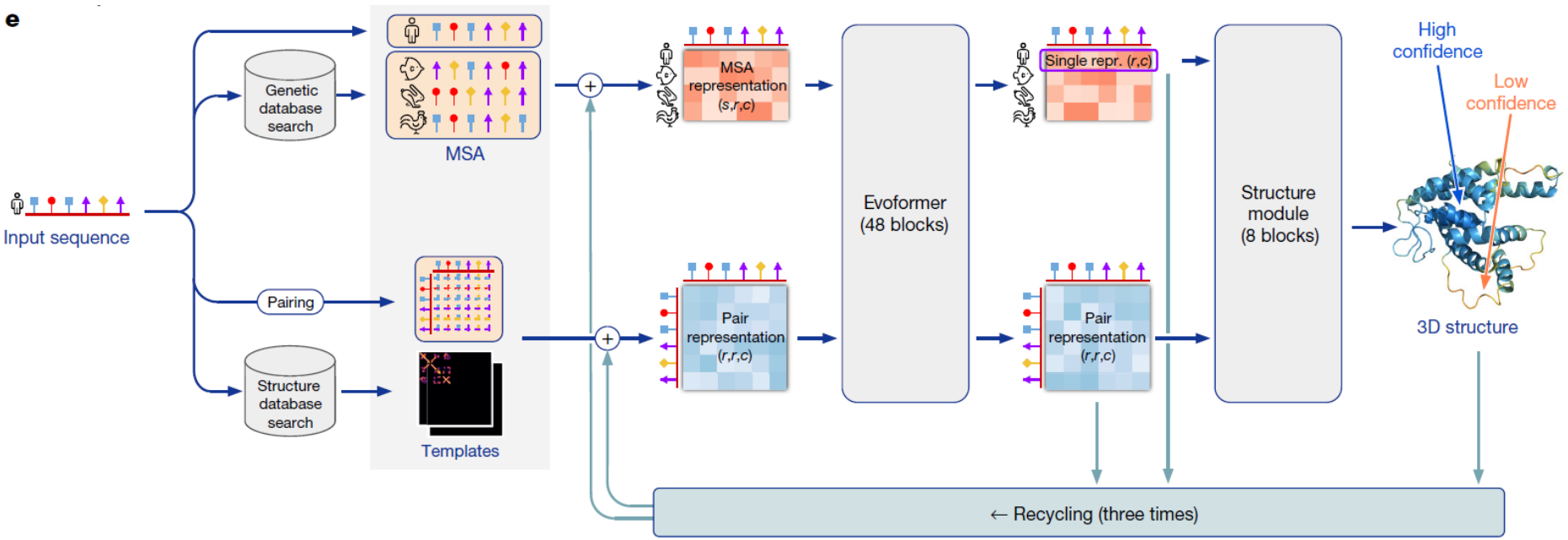
Check for updates

John Jumper[1,4]✉, Richard Evans[1,4], Alexander Pritzel[1,4], Tim Green[1,4], Michael Figurnov[1,4], Olaf Ronneberger[1,4], Kathryn Tunyasuvunakool[1,4], Russ Bates[1,4], Augustin Žídek[1,4], Anna Potapenko[1,4], Alex Bridgland[1,4], Clemens Meyer[1,4], Simon A. A. Kohl[1,4], Andrew J. Ballard[1,4], Andrew Cowie[1,4], Bernardino Romera-Paredes[1,4], Stanislav Nikolov[1,4], Rishub Jain[1,4], Jonas Adler[1], Trevor Back[1], Stig Petersen[1], David Reiman[1], Ellen Clancy[1], Michal Zielinski[1], Martin Steinegger[2,3], Michalina Pacholska[1], Tamas Berghammer[1], Sebastian Bodenstein[1], David Silver[1], Oriol Vinyals[1], Andrew W. Senior[1], Koray Kavukcuoglu[1], Pushmeet Kohli[1] & Demis Hassabis[1,4]✉

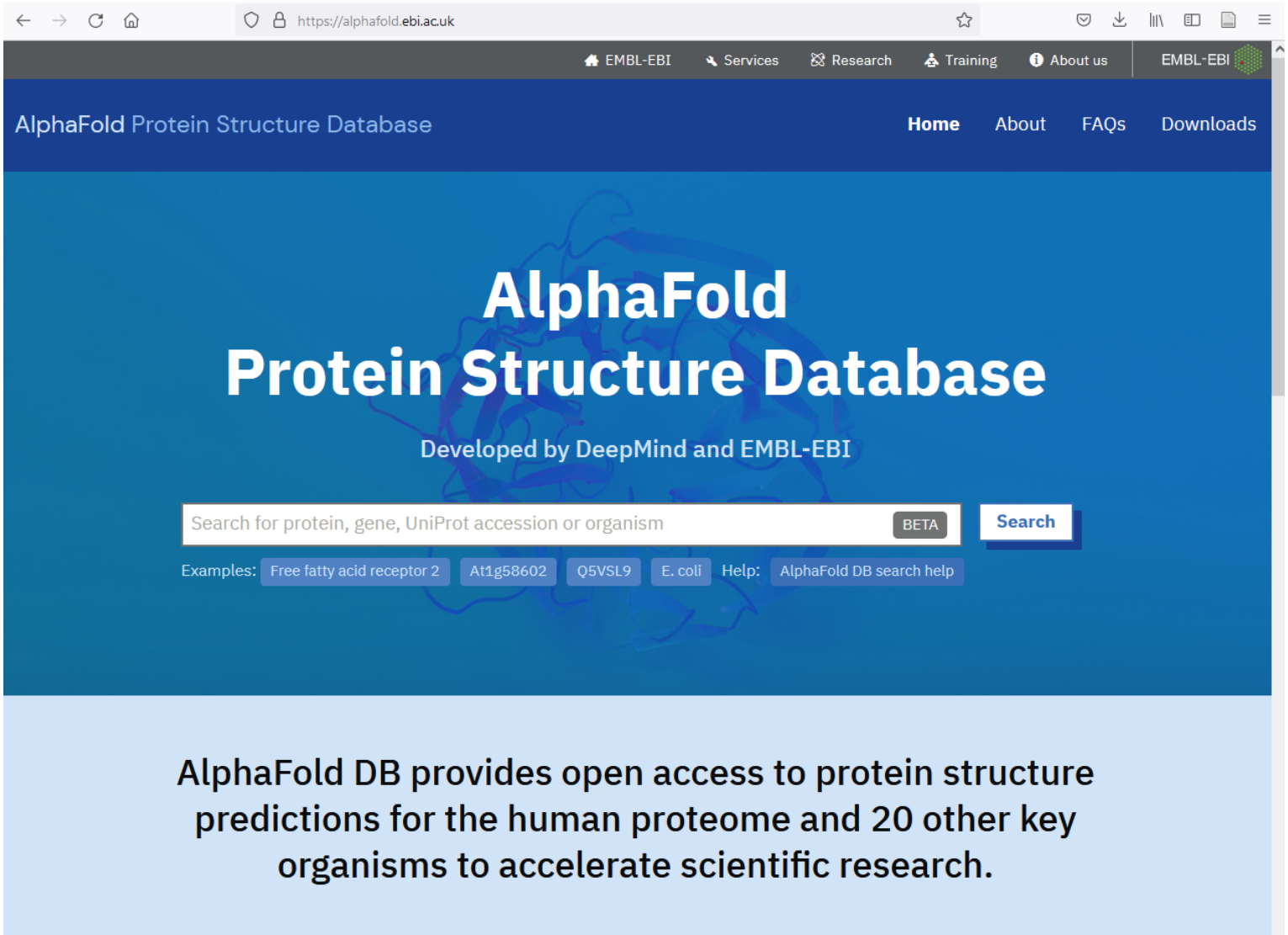
+27000 citations sur google scholar à ce jour (8 oct 2024) !

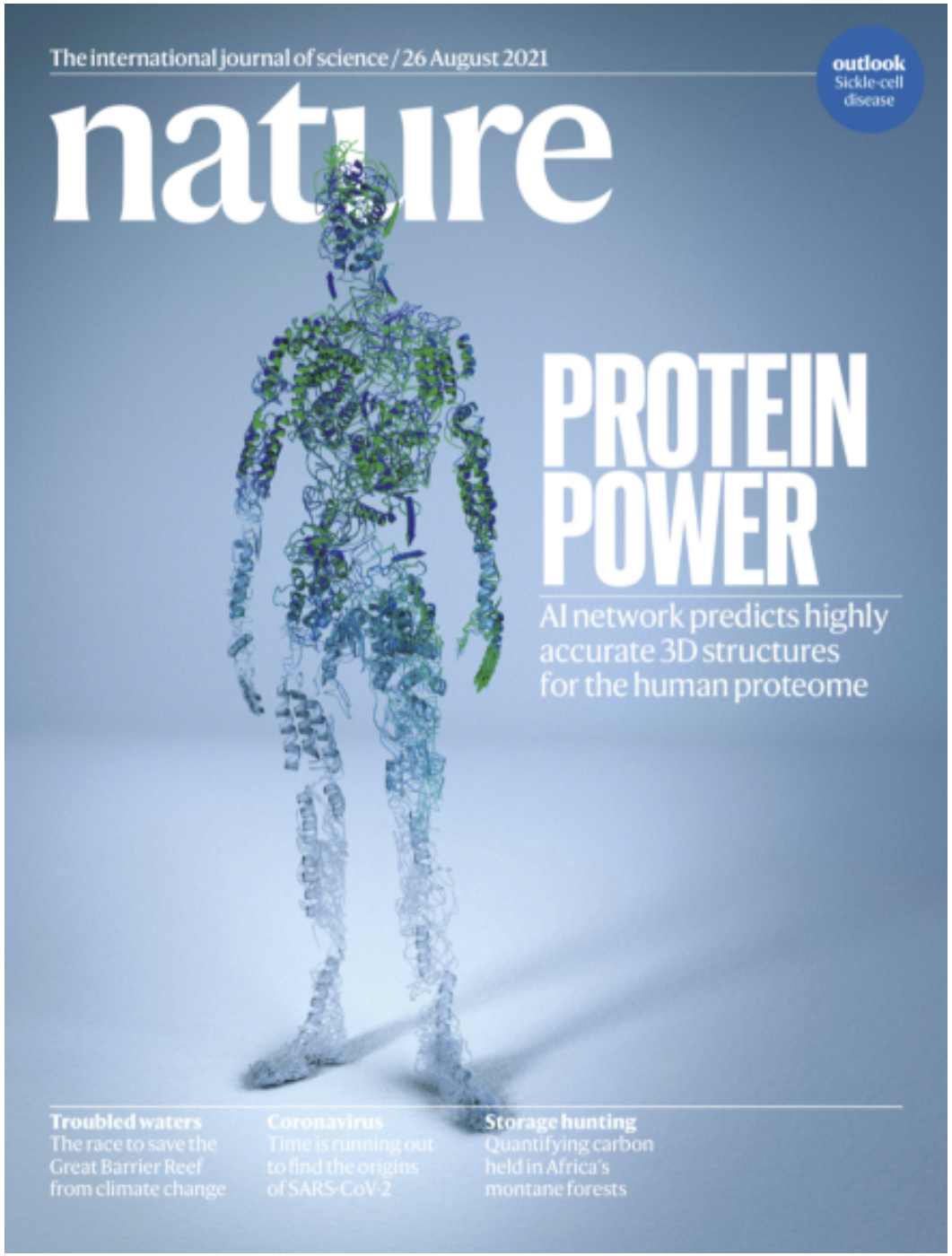
Principe de Alpha Fold 2



Applications de Alpha Fold

Alpha Fold DB




The international journal of science / 26 August 2021
nature
outlook
Sickle-cell disease
PROTEIN POWER
AI network predicts highly accurate 3D structures for the human proteome
Troubled waters
The race to save the Great Barrier Reef from climate change
Coronavirus
Time is running out to find the origins of SARS-CoV-2
Storage hunting
Quantifying carbon held in Africa's montane forests

Autres méthodes utilisant le deep learning

Le pionnier: RaptorX de Jinbo Xu et al.

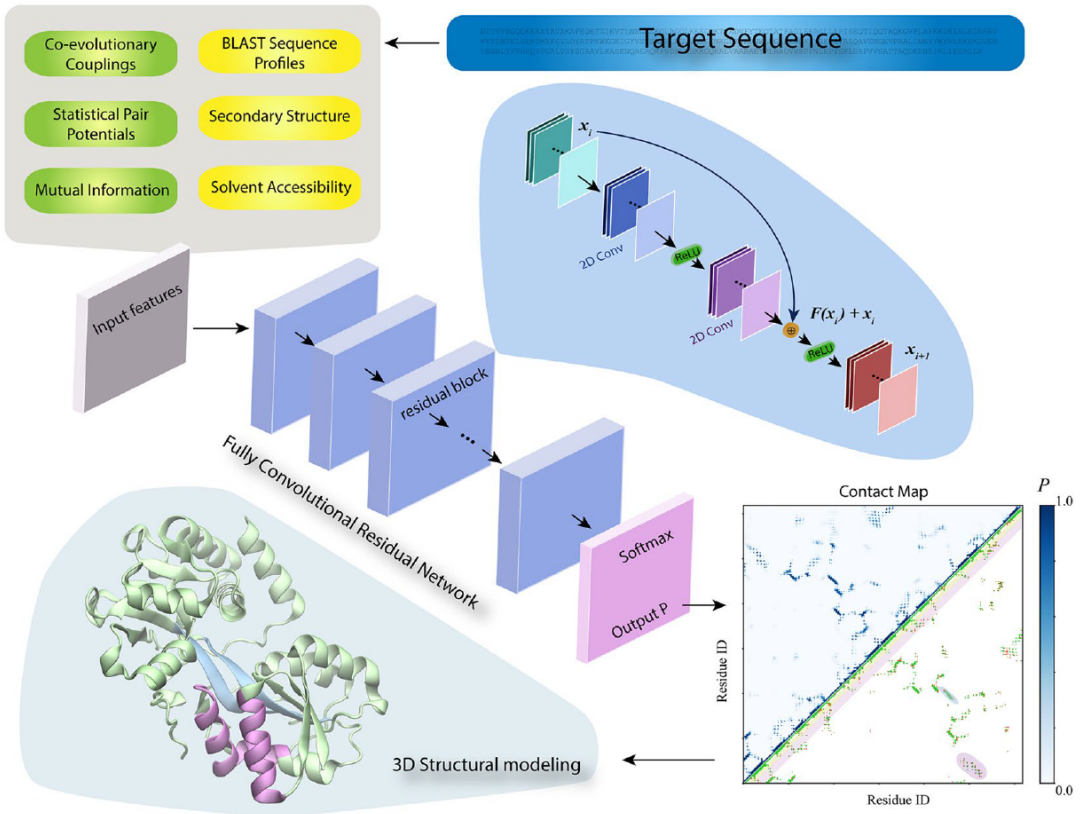
Serveur RaptorX:
`http://raptorx.uchicago.edu/`
CASP 14: score 38 vs 244 pour AlphaFold 2
"This server was officially ranked 1st in contact prediction in both CASP12 and CASP13 and initiated the revolution of protein structure prediction by deep learning."

DESTINI de Jeffrey Skolnick et al.

Serveur DESTINI:

`https://sites.gatech.edu/cssb/destini/`

CASP 14: score 29 vs 244 pour AlphaFold 2



Le rattrapage de David Baker

INSTITUTE FOR Protein Design
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JULY 15, 2021

RoseTTAFold: Accurate protein structure prediction accessible to all

Le rattrapage de David Baker

Science

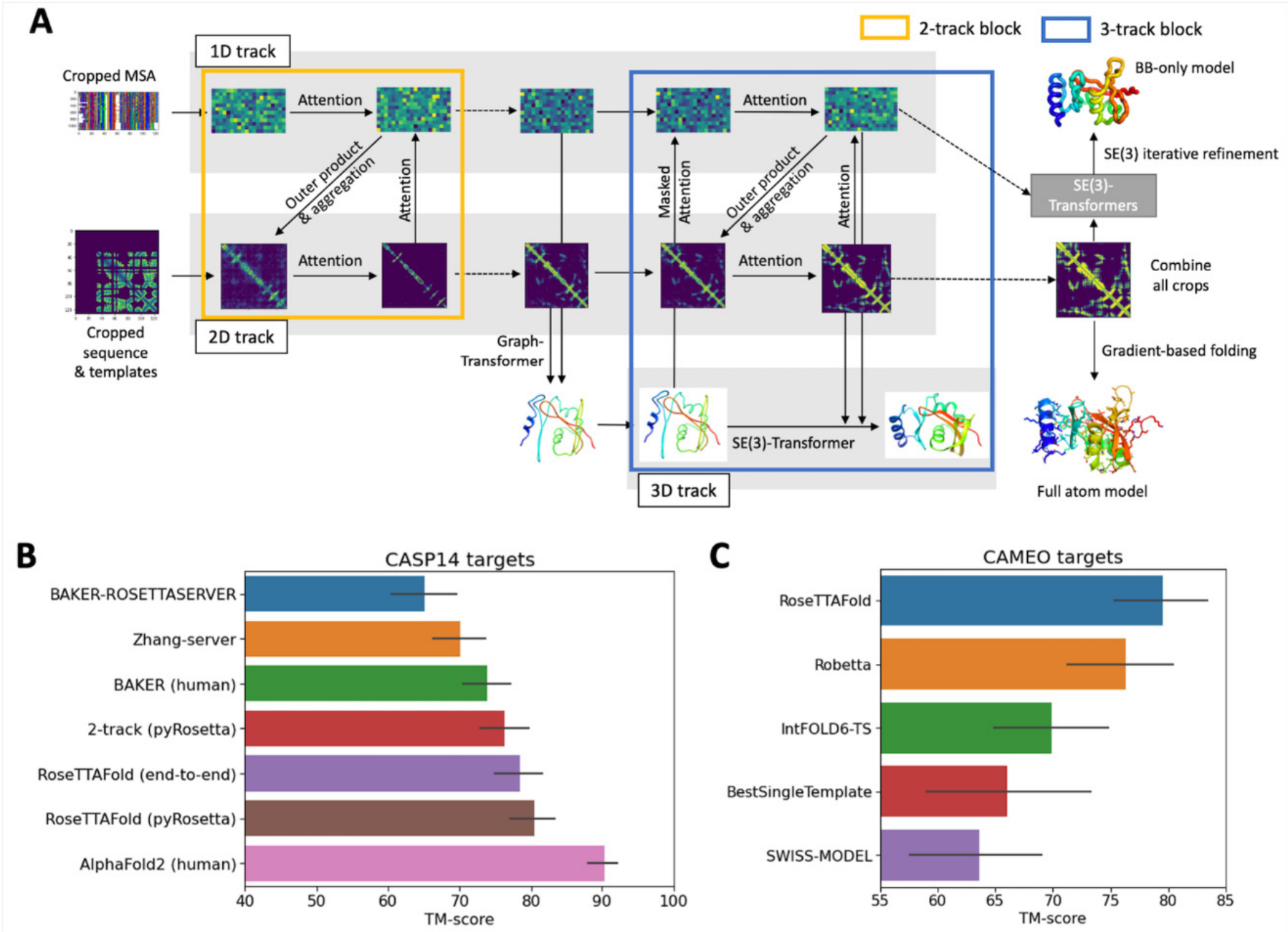
RESEARCH ARTICLES

Cite as: M. Baek *et al.*, *Science*
10.1126/science.abj8754 (2021).

Accurate prediction of protein structures and interactions using a three-track neural network

Minkyung Baek[1,2], Frank DiMaio[1,2], Ivan Anishchenko[1,2], Justas Dauparas[1,2], Sergey Ovchinnikov[3,4], Gyu Rie Lee[1,2], Jue Wang[1,2], Qian Cong[5,6], Lisa N. Kinch[7], R. Dustin Schaeffer[6], Claudia Millán[8], Hahnbeom Park[1,2], Carson Adams[1,2], Caleb R. Glassman[9,10], Andy DeGiovanni[12], Jose H. Pereira[12], Andria V. Rodrigues[12], Alberdina A. van Dijk[13], Ana C. Ebrecht[13], Diederik J. Opperman[14], Theo Sagmeister[15], Christoph Buhlheller[15,16], Tea Pavkov-Keller[15,17], Manoj K. Rathinaswamy[18], Udit Dalwadi[19], Calvin K. Yip[19], John E. Burke[18], K. Christopher Garcia[9,10,11,20], Nick V. Grishin[6,21,7], Paul D. Adams[12,22], Randy J. Read[8], David Baker[1,2,23]*

publié le même jour que le Nature sur AlphaFold2 (15 juillet 2021) !
mais "que" 4000 citations sur google scholar à ce jour (8 oct 2024)


A
2-track block
3-track block
1D track
Cropped MSA
Attention
Outer product & aggregation
Attention
Cropped sequence & templates
Attention
2D track
Graph-Transformer
Masked Attention
Attention
Outer product & aggregation
Attention
Attention
SE(3)-Transformer
3D track
BB-only model
SE(3) iterative refinement
SE(3)-Transformers
Combine all crops
Gradient-based folding
Full atom model
B
CASP14 targets
BAKER-ROSETTASERVER
Zhang-server
BAKER (human)
2-track (pyRosetta)
RoseTTAFold (end-to-end)
RoseTTAFold (pyRosetta)
AlphaFold2 (human)
40
50
60
70
80
90
100
TM-score
C
CAMEO targets
RoseTTAFold
Robetta
IntFOLD6-TS
BestSingleTemplate
SWISS-MODEL
55
60
65
70
75
80
85
TM-score

Next frontier: Oligomères

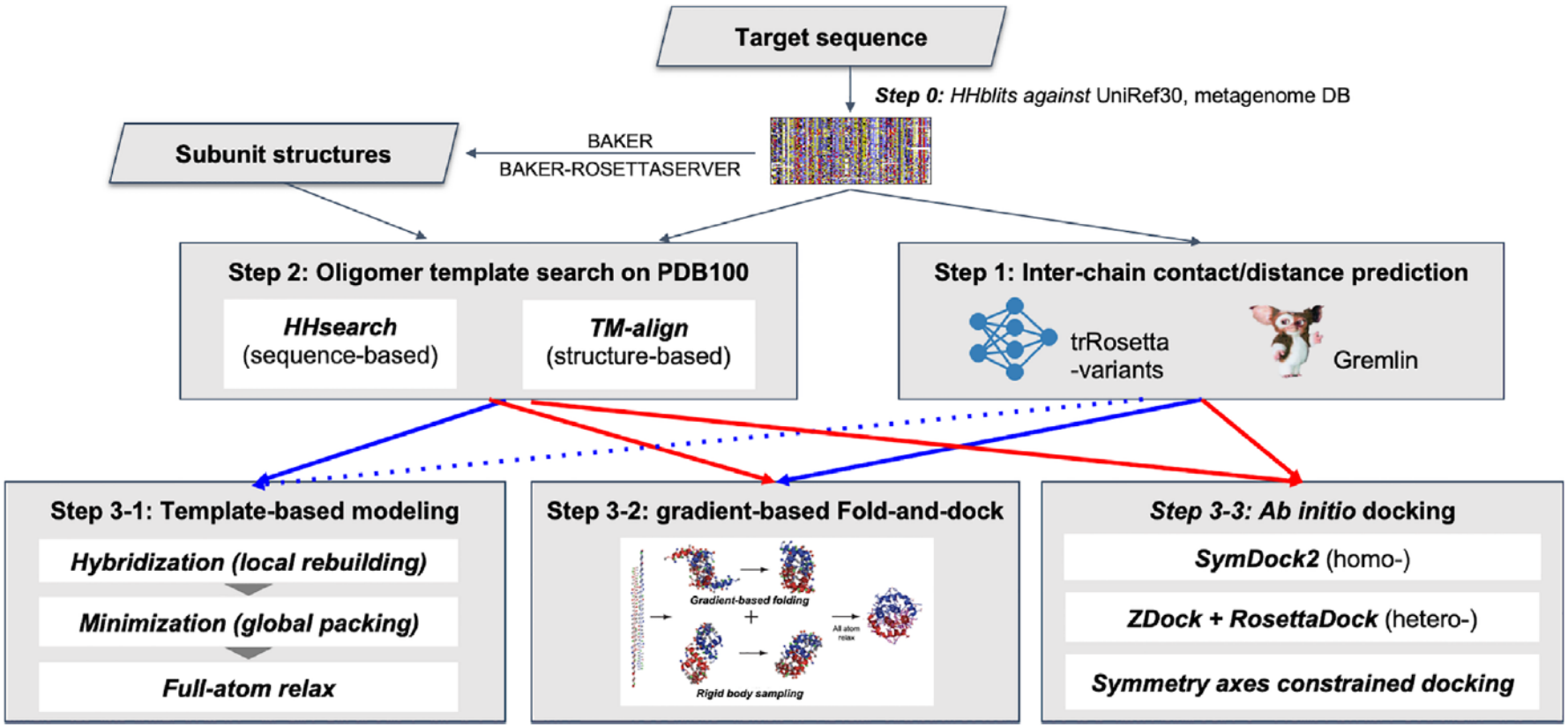


FIGURE 1 The oligomer structure modeling procedure used by the BAKER-experimental group

RosettaFold: github et serveur

https://github.com/RosettaCommons/RoseTTAFold

https://robetta.bakerlab.org/

voir aussi ici:

https://www.rosettacommons.org/

Comprendre Alpha Fold

Le blog de "Oxford Protein Informatics Group"

https://www.blopig.com/blog/2021/07/
alphafold-2-is-here-whats-behind-the-structure-predict

"... we have many new questions. What is the secret sauce before the news splash, and why is it so effective? Is it a piece of code that the average user can actually run? What are AlphaFold 2's shortcomings? And, most important of all, what will it mean for computational biology? And for all of us?"

Le blog de Mohammed AlQuraishi

https://moalquraishi.wordpress.com/2020/12/08/alphafold2-casp14-it-feels-like-ones-child-has-left-h
amp/

Mutations corrélées, MSA

Mutations corrélées, MSA

OPEN ACCESS Freely available online

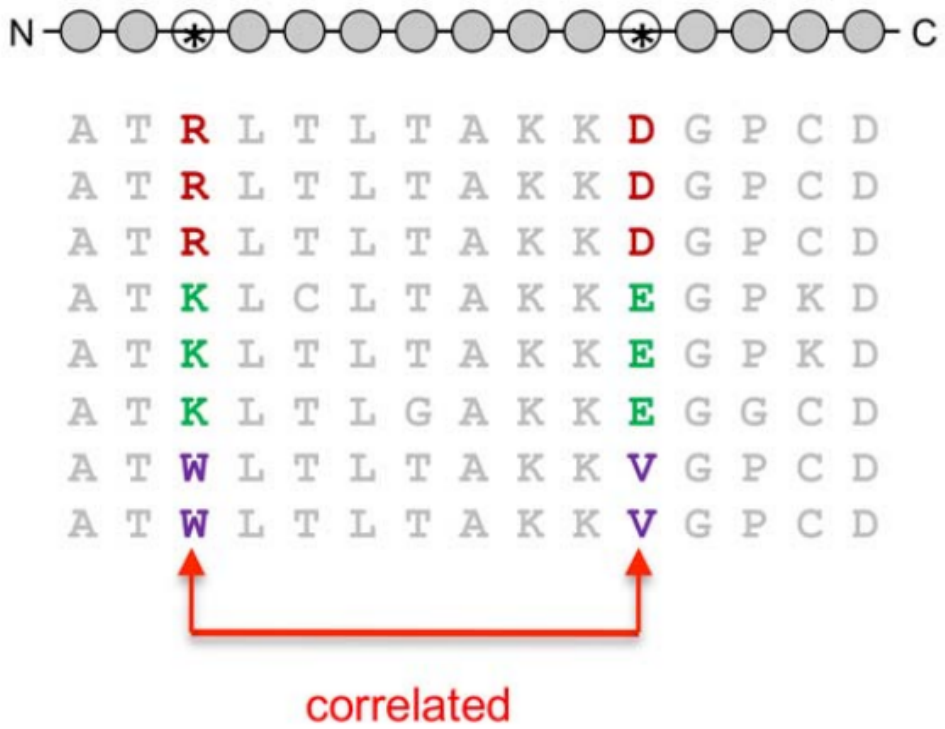
PLoS one

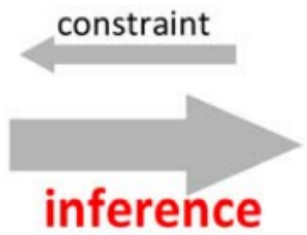
Protein 3D Structure Computed from Evolutionary Sequence Variation

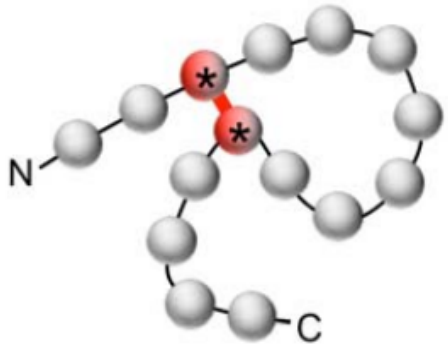
Debora S. Marks[1*☯], Lucy J. Colwell[2☯], Robert Sheridan[3], Thomas A. Hopf[1], Andrea Pagnani[4], Riccardo Zecchina[4,5], Chris Sander[3]

1 Department of Systems Biology, Harvard Medical School, Boston, Massachusetts, United States of America, **2** MRC Laboratory of Molecular Biology, Hills Road, Cambridge, United Kingdom, **3** Computational Biology Center, Memorial Sloan-Kettering Cancer Center, New York, New York, United States of America, **4** Human Genetics Foundation, Torino, Italy, **5** Politecnico di Torino, Torino, Italy

Principe

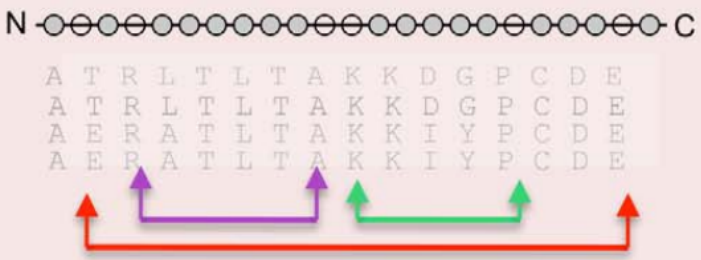

N
C
A T R L T L T A K K D G P C D
A T R L T L T A K K D G P C D
A T R L T L T A K K D G P C D
A T K L C L T A K K E G P K D
A T K L T L T A K K E G P K D
A T K L T L G A K K E G G C D
A T W L T L T A K K V G P C D
A T W L T L T A K K V G P C D
correlated


constraint
inference


N
C
contact in 3D

Protocole de 2011 sans deep-learning

Align evolutionary diverged sequences



Calculate covariance matrix for each pair of sequence positions for all pairs of amino acids (A,B)

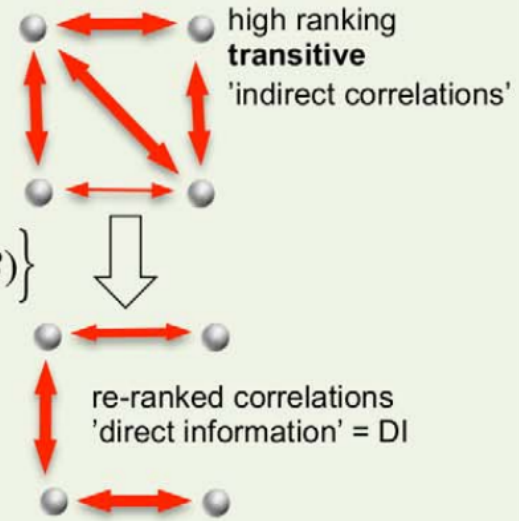
$$C_{ij}(A,B) = f_{ij}(A,B) - f_i(A)P_j(B)$$

$$C_{ij}^{-1}(A,B) = -e_{ij}(A,B)_{i \neq j}$$

Identify maximally informative pair couplings using **statistical model** of entire protein to infer residue-residue co-evolution

$$P_{ij}^{Dir}(A,B) = \frac{1}{Z}\exp\left\{e_{ij}(A,B) + \tilde{h}_i(A) + \tilde{h}_j(B)\right\}$$

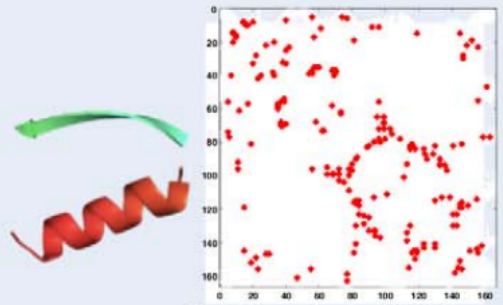
$$DI_{ij} = \sum_{A,B=1}^{q} P_{ij}^{Dir}(A,B)\ln\frac{P_{ij}^{Dir}(A,B)}{f_i(A)f_j(B)}$$



Protocole de 2011 sans deep-learning

Analyze the highest scoring pairs to produce ranked list of residue pairs which we predict to be close in 3D space. Use these pairs as predicted close “evolutionary inferred contacts” , EICs, in folding calculations

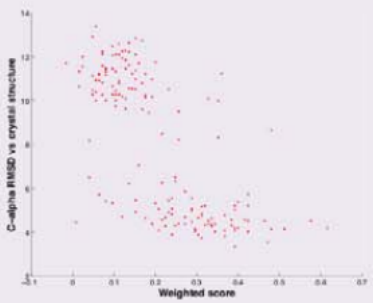
```
assign (resid 143 and name CA) (resid 123 and name CA)  4 4 3
assign (resid 16 and name CA) (resid 10 and name CA)  4 4 3
assign (resid 141 and name CA) (resid 82 and name CA)  4 4 3
assign (resid 129 and name CA) (resid 87 and name CA)  4 4 3
assign (resid 92 and name CA) (resid 11 and name CA)  4 4 3
assign (resid 116 and name CA) (resid 81 and name CA)  4 4 3
```



predicted contacts (EICs)

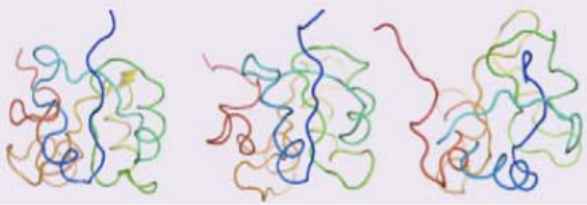
Start with extended structure
use **distance geometry** and **simulated annealing** with predicted constraints, EICs, to fold the chain

Rank predicted structures using quality measure of backbone alpha torsion and beta sheet twist



good scores

bad scores



Utiliser Alpha Fold

google colab

https://colab.research.google.com/notebooks/intro.ipynb?utm_source=scs-index

https://www.youtube.com/watch?v=inN8seMm7UI

Exemple pour faire de la MD ou aussi AlphaFold2 + MD:

https://github.com/pablo-arantes/Making-it-rain

Alpha Fold avec google colab

une version amélioré d'alphaFold2:
`https://colab.research.google.com/github/sokrypton/ColabFold/blob/main/AlphaFold2.ipynb`
ou l'original:
`https://colab.research.google.com/github/deepmind/alphafold/blob/main/notebooks/AlphaFold.ipynb`

Alpha Fold en local

On peux l'installer aussi sur une machine en local, mais il faut qu'elle soit très puissante:

`https://github.com/deepmind/alphafold`

" The simplest way to run AlphaFold is using the provided Docker script. This was tested on Google Cloud with a machine using the nvidia-gpu-cloud-image with 12 vCPUs, 85 GB of RAM, a 100 GB boot disk, the databases on an additional 3 TB disk, and an A100 GPU."

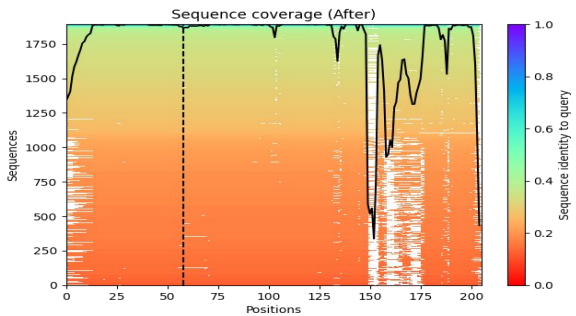
Rien que la carte graphique A100 coûte le prix d'une petite voiture:

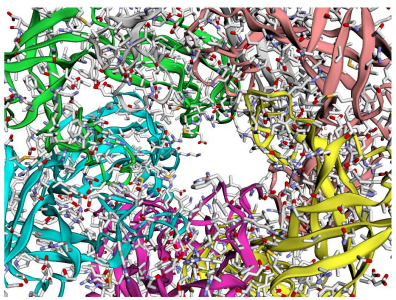
`https://fr.aliexpress.com/item/1005002408111365.html`

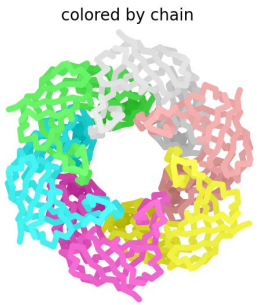
ColabFold

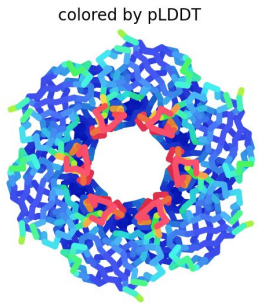
Making Protein folding accessible to all via Google Colab

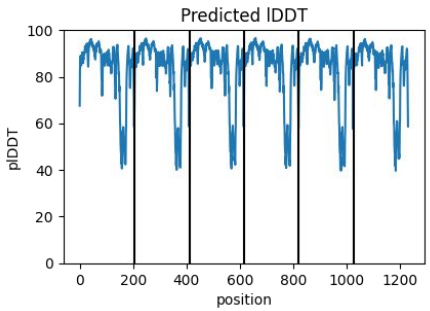
(and the unintended uses of AlphaFold)

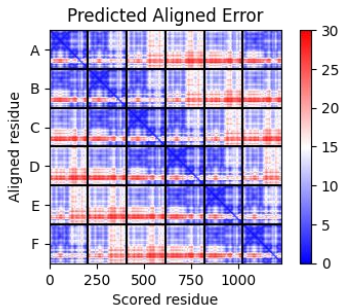








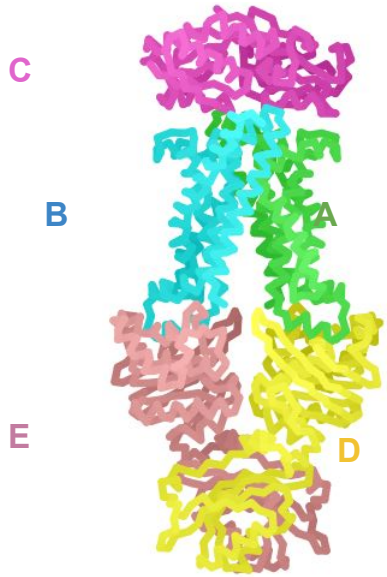


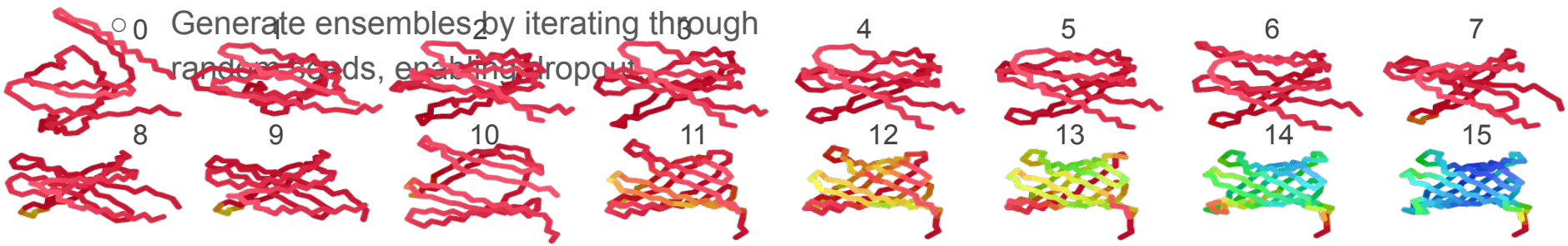


github.com/sokrypton/ColabFold

ColabFold - Advanced options

- Modify MSA input
  - Custom or MMseqs2 (much faster)
  - Trim
- **Complexes**
  - **Homo-oligomers**
  - **Hetero-oligomers**
- Fine control
  - Number of recycles
- Sample (Output more than 5 models)
  - Generate ensembles by iterating through random seeds, enabling dropout



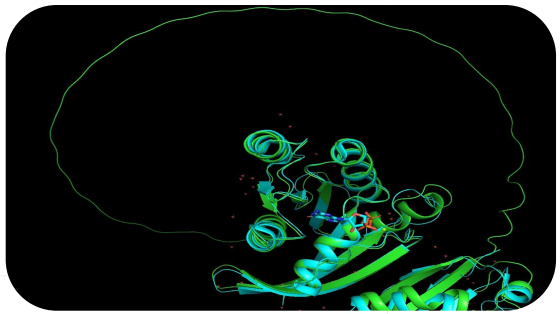


Can predict protein-protein/peptide interactions

Yoshitaka Moriwaki @Ag_smith · Jul 19

AlphaFold2 can also predict heterocomplexes. All you have to do is input the two sequences you want to predict and connect them with a long linker.

G-linker!



Hiroki Onoda
@onoda_hiroki

Unknown linker may be useful for multimer prediction on the local Alphafold2!!

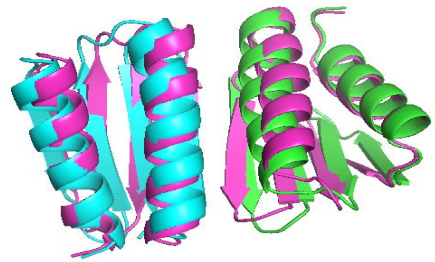
UNK-linker!

Minkyung Baek
@minkbaek

Don't actually need a G-linker!

Adding a big enough number for "residue_index" feature is enough to model hetero-complex using AlphaFold (green&cyan: crystal structure / magenta: predicted model w/ residue_index modification).
#AlphaFold #alphafold2

```
# add big enough number to residue index to indicate chain breaks
idx_res = feature_dict['residue_index']
L_prev = 0
# Ls: number of residues in each chain
for L_i in Ls[:-1]:
    idx_res[L_prev+L_i:] += 200
    L_prev += L_i
feature_dict['residue_index'] = idx_res
```

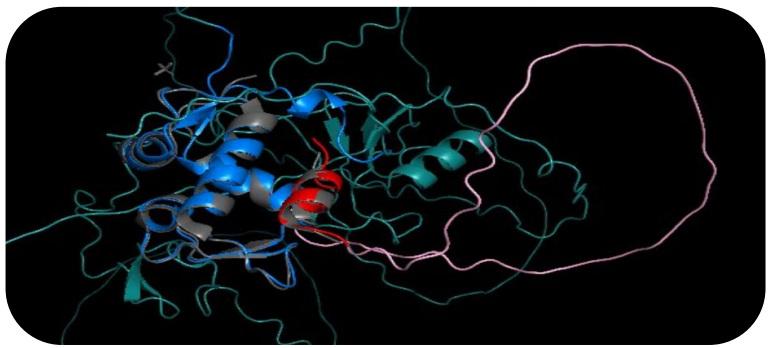


大上雅史 | Ohue M 2.5G
@tonets

あ、AlphaFold2でペプチドドッキングできる

Translated from Japanese by Google

Oh, I was able to dock the peptide with AlphaFold2

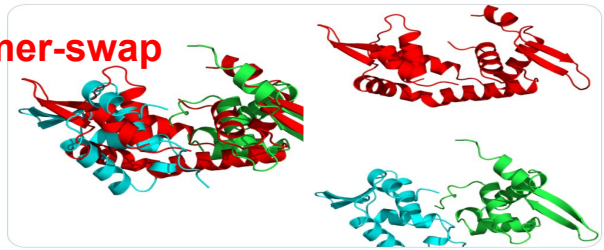


Protein-peptide interaction

Can predict protein-protein/peptide interactions

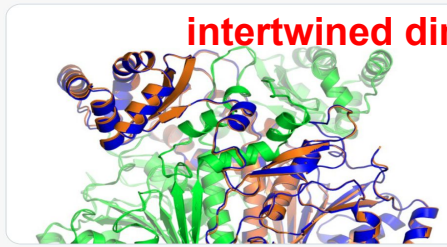
Cesar Ramirez-Sarmiento @cxarramirez · Jul 22
Replying to @cxarramirez @sokrypton and 3 others
OMG😱 the monomers in the predicted "homodimer" of FoxP1 (cyan, green) show very similar orientations when compared to the monomers in the domain-swapped structure (red)🤯

dimer-swap



James Murray @jwm_imperial · Jul 24
Replying to @drpetermoody
There are already notebooks to predict heterodimers and homooligomers. github.com/sokrypton/Cola... For my unpublished intertwined dimer, the monomer and dimer predictions were essentially identical, also matching the crystal structure exactly except for a few rotamers.

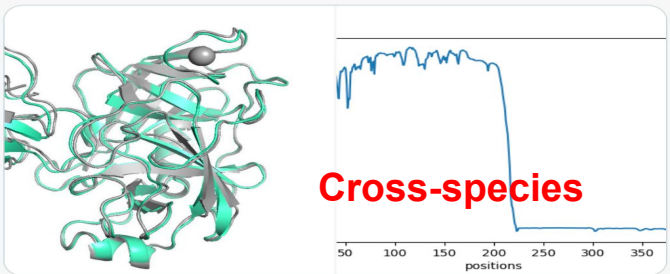
intertwined dimer



Eugene Valkov @eugenevalkov · Jul 26
After days of running #alphafold2, I am still astounded by insights it provides. Here, it predicted mode of peptide binding *completely consistent* with biochemical data and mutagenesis and gave additional clues which we will explore!

consistent w/ biochem data

padhorny @padhorny · Jul 20
Replying to @sokrypton and @minkbaek
Amazing stuff. Seemingly can even do cross-species complexes (at least the strong binders). Here is what it gave me for rcsb.org/structure/1AVX (not the top model though):



Cross-species

Preprints rolling in...

Can AlphaFold2 predict protein-peptide complex structures accurately?

Junsu Ko, Juyong Lee

bioRxiv 2021.07.27.453972; doi: https://doi.org/10.1101/2021.07.27.453972

Harnessing protein folding neural networks for peptide-protein docking

Tomer Tsaban, Julia Varga, Orly Avraham, Ziv Ben-Aharon, Alisa Khramushin, Ora Schueler-Furman

bioRxiv 2021.08.01.454656; doi: https://doi.org/10.1101/2021.08.01.454656

Improved Docking of Protein Models by a Combination of Alphafold2 and ClusPro

Usman Ghani, Israel Desta, Akhil Jindal, Omeir Khan, George Jones, Sergey Kotelnikov, Dzmitry Padhorny, Sandor Vajda, Dima Kozakov

bioRxiv 2021.09.07.459290; doi: https://doi.org/10.1101/2021.09.07.459290

ColabFold se diversifie, services en 2022

Making Protein folding accessible to all via Google Colab!

Notebooks	monomers	complexes	mmseqs2	jackhmmer	templates
AlphaFold2_mmseqs2	Yes	Yes	Yes	No	Yes
AlphaFold2_batch	Yes	Yes	Yes	No	Yes
RoseTTAFold	Yes	No	Yes	No	No
AlphaFold2 (from Deepmind)	Yes	Yes	No	Yes	No
BETA (in development) notebooks					
OmegaFold	Yes	No	No	No	No
AlphaFold2_advanced	Yes	Yes	Yes	Yes	No
OLD retired notebooks					
AlphaFold2_complexes	No	Yes	No	No	No
AlphaFold2_jackhmmer	Yes	No	Yes	Yes	No
AlphaFold2_noTemplates_noMD					
AlphaFold2_noTemplates_yesMD					

ColabFold se diversifie, services en 2024

https://github.com/sokrypton/ColabFold

Making Protein folding accessible to all via Google Colab!

Notebooks	monomers	complexes	mmseqs2	jackhmmer	templates
AlphaFold2_mmseqs2	Yes	Yes	Yes	No	Yes
AlphaFold2_batch	Yes	Yes	Yes	No	Yes
AlphaFold2 (from Deepmind)	Yes	Yes	No	Yes	No
relax_amber (relax input structure)					
ESMFold	Yes	Maybe	No	No	No
BETA (in development) notebooks					
RoseTTAFold2	Yes	Yes	Yes	No	WIP
OmegaFold	Yes	Maybe	No	No	No
AlphaFold2_advanced_v2 (new experimental notebook)	Yes	Yes	Yes	No	Yes

ColabDesign

https://github.com/sokrypton/ColabDesign

ColabDesign

Making Protein Design accessible to all via Google Colab!

- P(structure | sequence)
  - TrDesign - using TrRosetta for design
  - AfDesign - using AlphaFold for design
  - [WIP] RfDesign - using RoseTTAFold for design
- P(sequence | structure)
  - ProteinMPNN
  - [WIP] TrMRF
- P(sequence)
  - [WIP] MSA_transformer
  - [WIP] SEQ - (GREMLIN, mfDCA, arDCA, plmDCA, bmDCA, etc)
- P(structure)
  - Rfdiffusion

AlphaFold-Multimer de DeepMind

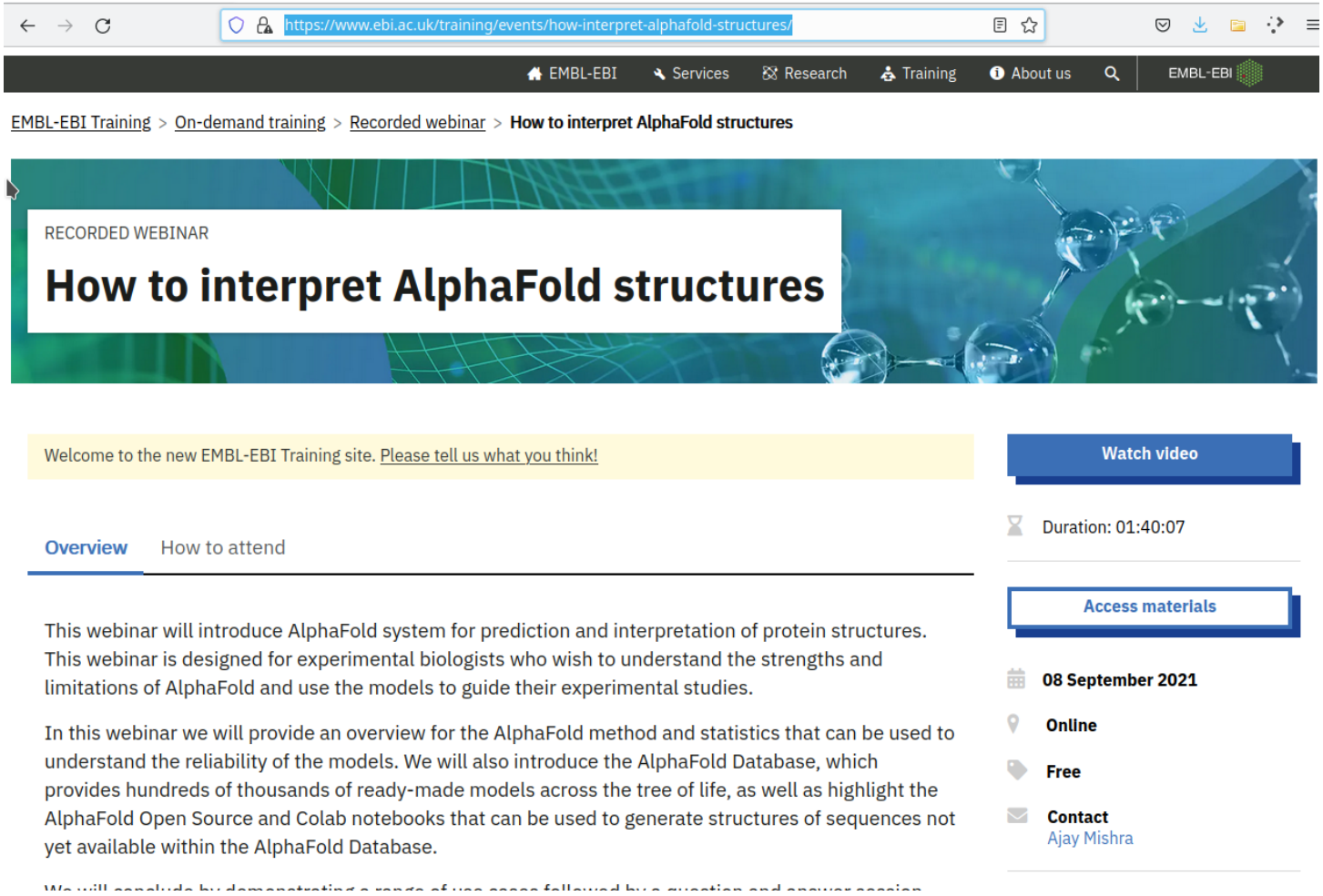
Richard Evans et al. en preprint sur bioRxiv depuis 2021, jamais publié dans un journal avec processus de review !
`https://www.biorxiv.org/content/10.1101/2021.10.04.463034v2`

Pour aller plus loin: EMBL webinar

EMBL webinar "How to interpret AlphaFold structures"

8 sept 2021

https://www.ebi.ac.uk/training/events/how-interpret-alphafold-structures/



DeepMind

Introduction to AlphaFold

Presenter: Kathryn Tunyasuvunakool
Research Scientist at DeepMind

How does it work? (the short version)

Article

Highly accurate protein structure prediction with AlphaFold

https://doi.org/10.1038/s41586-021-03819-2
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Open access
Check for updates

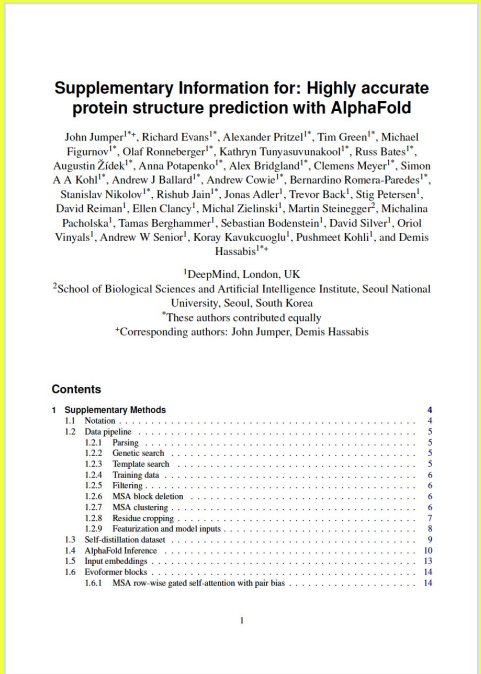
John Jumper[1,4,5✉], Richard Evans[1,4], Alexander Pritzel[1,4], Tim Green[1,4], Michael Figurnov[1,4], Olaf Ronneberger[1,4], Kathryn Tunyasuvunakool[1,4], Russ Bates[1,4], Augustin Žídek[1,4], Anna Potapenko[1,4], Alex Bridgland[1,4], Clemens Meyer[1,4], Simon A. A. Kohl[1,4], Andrew J. Ballard[1,4], Andrew Cowie[1,4], Bernardino Romera-Paredes[1,4], Stanislav Nikolov[1,4], Rishub Jain[1,4], Jonas Adler[1], Trevor Back[1], Stig Petersen[1], David Reiman[1], Ellen Clancy[1], Michal Zielinski[2], Martin Steinegger[2,3], Michalina Pacholska[1], Tamas Berghammer[1], Sebastian Bodenstein[1], David Silver[1], Oriol Vinyals[1], Andrew W. Senior[1], Koray Kavukcuoglu[1], Pushmeet Kohli[1] & Demis Hassabis[1,4✉]

Proteins are essential to life, and understanding their structure can facilitate a mechanistic understanding of their function. Through an enormous experimental effort[1–4], the structures of around 100,000 unique proteins have been determined[5], but this represents a small fraction of the billions of known protein sequences[6,7]. Structural coverage is bottlenecked by the months to years of painstaking effort required to determine a single protein structure. Accurate computational approaches are needed to address this gap and to enable large-scale structural bioinformatics. Predicting the three-dimensional structure that a protein will adopt based solely on its amino acid sequence—the structure prediction component of the 'protein folding problem'[8]—has been an important open research problem for more than 50 years[9]. Despite recent progress[10–14], existing methods fall far short of atomic accuracy, especially when no homologous structure is available. Here we provide the first computational method that can regularly predict protein structures with atomic accuracy even in cases in which no similar structure is known. We validated an entirely redesigned version of our neural network-based model, AlphaFold, in the challenging 14th Critical Assessment of protein Structure Prediction (CASP14)[15], demonstrating accuracy competitive with experimental structures in a majority of cases and greatly outperforming other methods. Underpinning the latest version of AlphaFold is a novel machine learning approach that incorporates physical and biological knowledge about protein structure, leveraging multi-sequence alignments, into the design of the deep learning algorithm.

The development of computational methods to predict three-dimensional (3D) protein structures from the protein sequence has proceeded along two complementary paths that focus on either the physical interactions or the evolutionary history. The physical interaction programme heavily integrates our understanding of molecular driving forces into either thermodynamic or kinetic simulation of protein physics[16] or statistical approximations thereof[17]. Although theoretically very appealing, this approach has proved highly challenging for even moderate-sized proteins due to the computational intractability of molecular simulation, the context dependence of protein stability and the difficulty of producing sufficiently accurate models of protein physics. The evolutionary programme has provided an alternative in recent years, in which the constraints on protein structure are derived from bioinformatics analysis of the evolutionary history of proteins, homology to solved structures[18–20] and pairwise evolutionary correlations[21,22]. This bioinformatics approach has benefited greatly from the steady growth of experimental protein structures deposited in the Protein Data Bank (PDB)[5], the explosion of genomic sequencing and the rapid development of deep learning techniques to interpret these correlations. Despite these advances, contemporary physical and evolutionary-history-based approaches produce predictions that are far short of experimental accuracy in the majority of cases in which a close homologue has not been solved experimentally and this has limited their utility for many biological applications.

In this study, we develop the first, to our knowledge, computational approach capable of predicting protein structures to near experimental accuracy in a majority of cases. The neural network AlphaFold that we developed was entered into the CASP14 assessment (May–July 2020; entered under the team name 'AlphaFold2' and a completely different model from our CASP13 AlphaFold system[23]). The CASP assessment is carried out biennially using recently solved structures that have not been deposited in the PDB or publicly disclosed so that it is a blind test

[1]DeepMind, London, UK. [2]School of Biological Sciences, Seoul National University, Seoul, South Korea. [3]Artificial Intelligence Institute, Seoul National University, Seoul, South Korea. [4]These authors contributed equally: John Jumper, Richard Evans, Alexander Pritzel, Tim Green, Michael Figurnov, Olaf Ronneberger, Kathryn Tunyasuvunakool, Russ Bates, Augustin Žídek, Anna Potapenko, Alex Bridgland, Clemens Meyer, Simon A. A. Kohl, Andrew J. Ballard, Andrew Cowie, Bernardino Romera-Paredes, Stanislav Nikolov, Rishub Jain, Demis Hassabis. [5]✉e-mail: jumper@deepmind.com; dhcontact@deepmind.com



Supplementary Information for: Highly accurate protein structure prediction with AlphaFold

John Jumper[1*+], Richard Evans[1*], Alexander Pritzel[1*], Tim Green[1*], Michael Figurnov[1*], Olaf Ronneberger[1*], Kathryn Tunyasuvunakool[1*], Russ Bates[1*], Augustin Žídek[1*], Anna Potapenko[1*], Alex Bridgland[1*], Clemens Meyer[1*], Simon A A Kohl[1*], Andrew J Ballard[1*], Andrew Cowie[1*], Bernardino Romera-Paredes[1*], Stanislav Nikolov[1*], Rishub Jain[1*], Jonas Adler[1], Trevor Back[1], Stig Petersen[1], David Reiman[1], Ellen Clancy[1], Michal Zielinski[1], Martin Steinegger[2], Michalina Pacholska[1], Tamas Berghammer[1], Sebastian Bodenstein[1], David Silver[1], Oriol Vinyals[1], Andrew W Senior[1], Koray Kavukcuoglu[1], Pushmeet Kohli[1], and Demis Hassabis[1*+]

[1]DeepMind, London, UK
[2]School of Biological Sciences and Artificial Intelligence Institute, Seoul National University, Seoul, South Korea
[*]These authors contributed equally
[+]Corresponding authors: John Jumper, Demis Hassabis

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1.2.2 Genetic search 5
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1.2.4 Training data 6
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1.2.6 MSA block deletion 6
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1.2.8 Residue cropping 7
1.2.9 Featurization and model inputs 8
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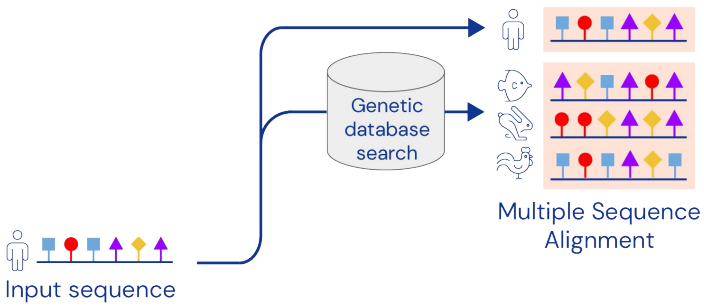
1.6 Evoformer blocks 14
1.6.1 MSA row-wise gated self-attention with pair bias 14

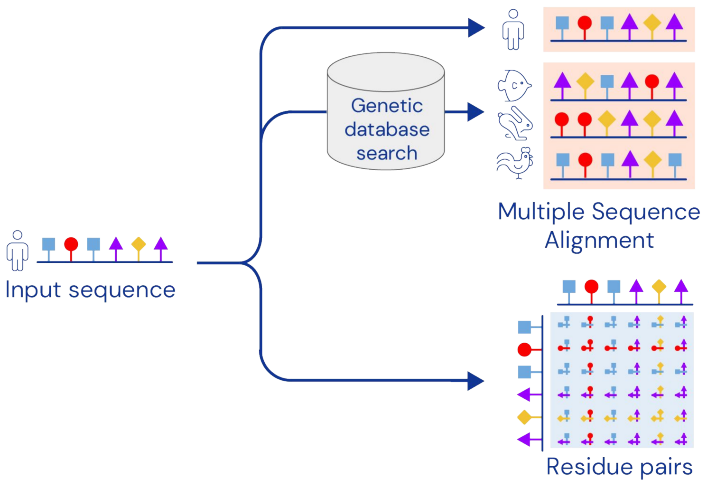
See Jumper et al. 2021 (especially the SI) for details

Inputs



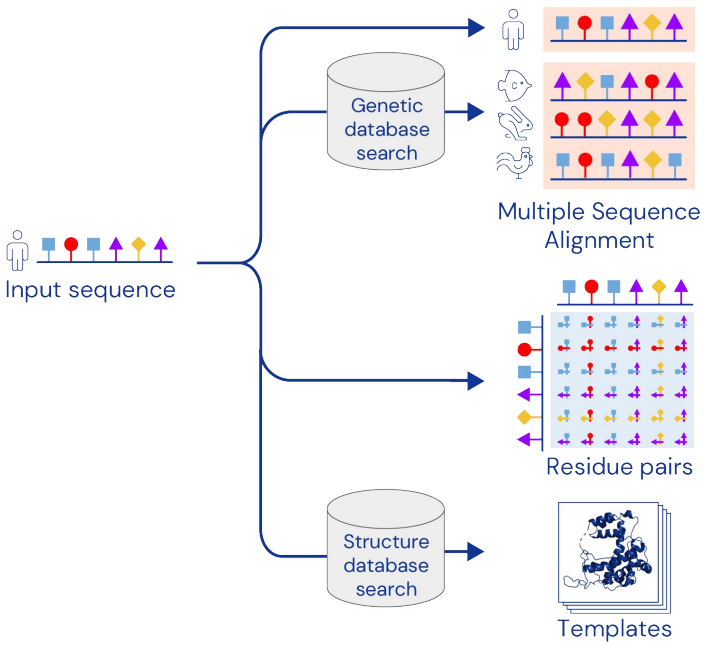
A key AlphaFold input is the MSA, containing sequences evolutionarily related to the target. Related sequences are found using standard tools and public databases.

Inputs



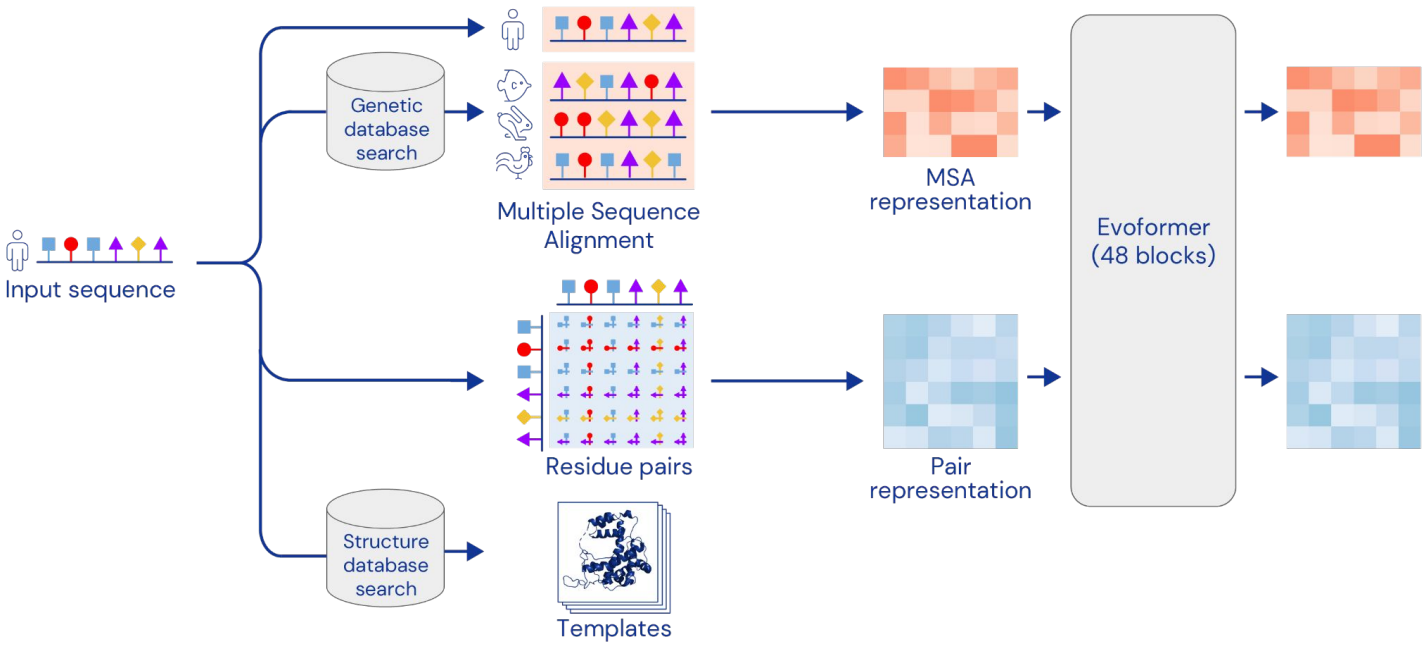
The input sequence is used to create an array of numbers representing all residue pairs.

Inputs



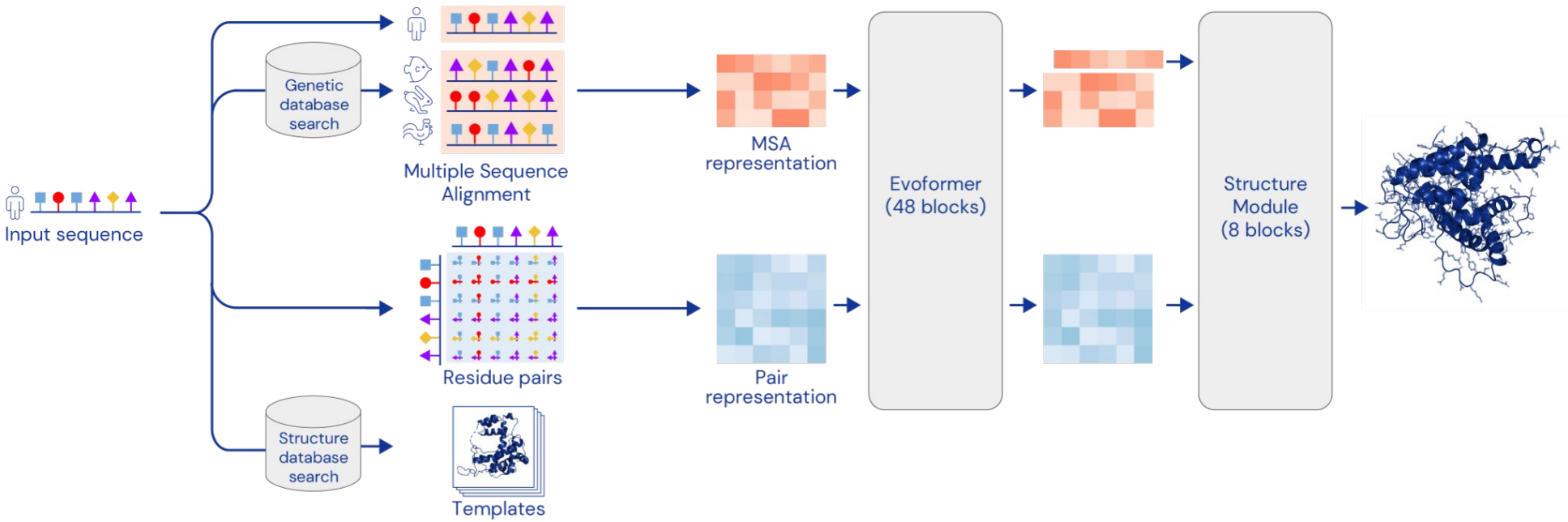
AlphaFold can also use template structures from the PDB, found using standard tools. However, it often produces accurate predictions without a template.

Network



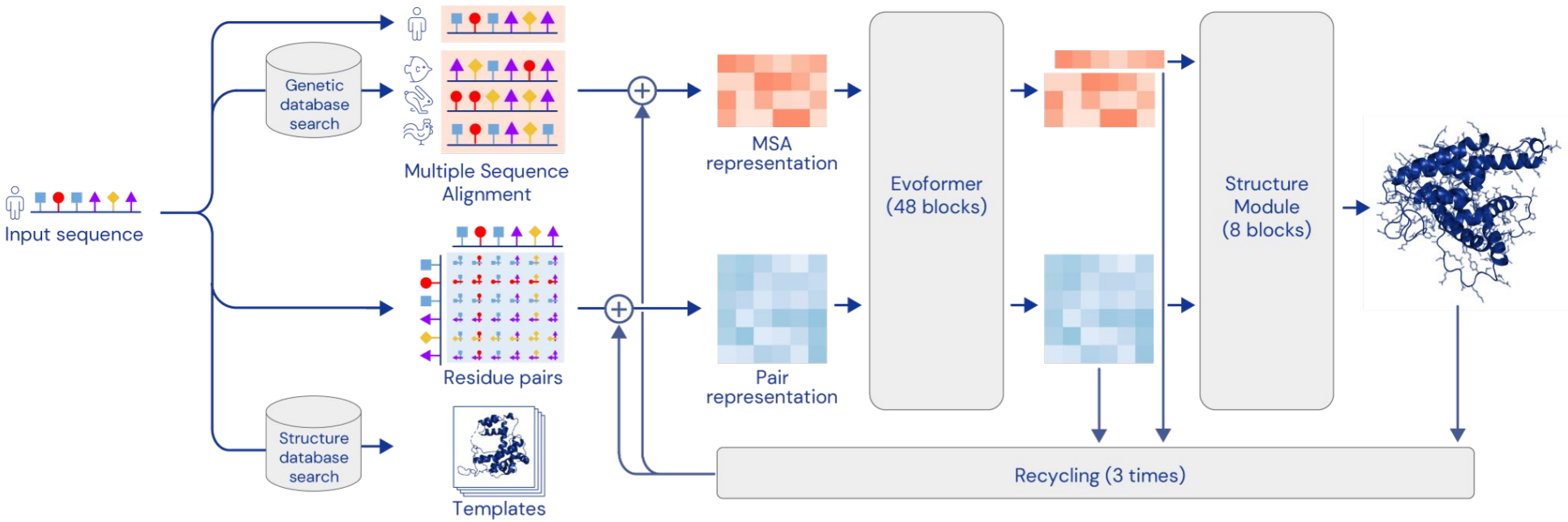
The Evoformer blocks extract information about the relationship between residues.
The MSA representation can update the pair representation and vice versa.

Network



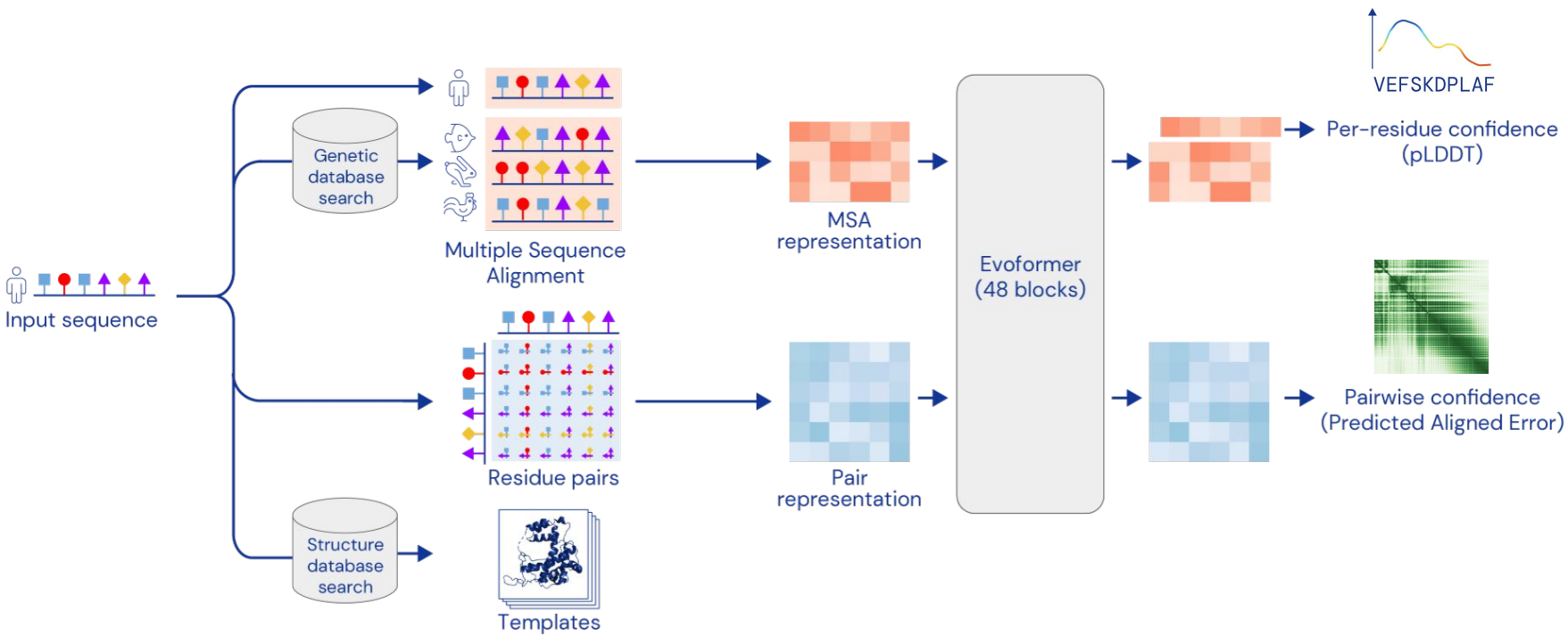
The Structure Module predicts a rotation + translation to place each residue.
A small network predicts side chain chi angles. The final structure is run through a relaxation process.

Network



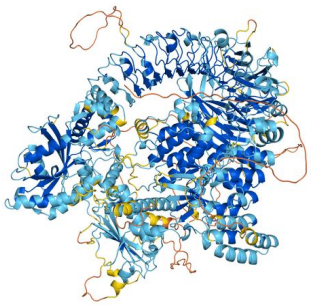
Feeding certain outputs back through the network again improves performance

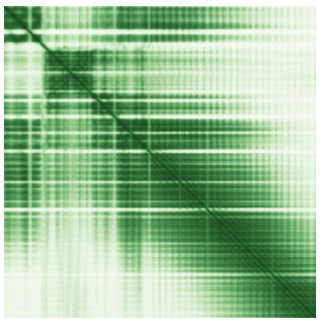
Other outputs



As well as a predicted structure, AlphaFold produces two confidence estimates

Interpreting predictions





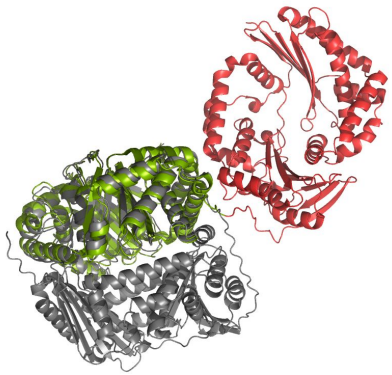
The short version: use **both** confidence metrics

Predicted LDDT: definition

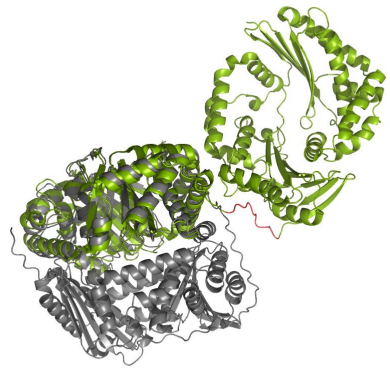
AlphaFold's per-residue prediction of its lDDT-Cα score*

Roughly, lDDT measures the percentage of correctly predicted interatomic distances, not how well the predicted and true structures can be superimposed.

It rewards **locally correct** structures, and **getting individual domains right**.
pLDDT behaves similarly, as a measure of **local confidence**



Alignment-based metric



lDDT

*Mariani et al. 2013

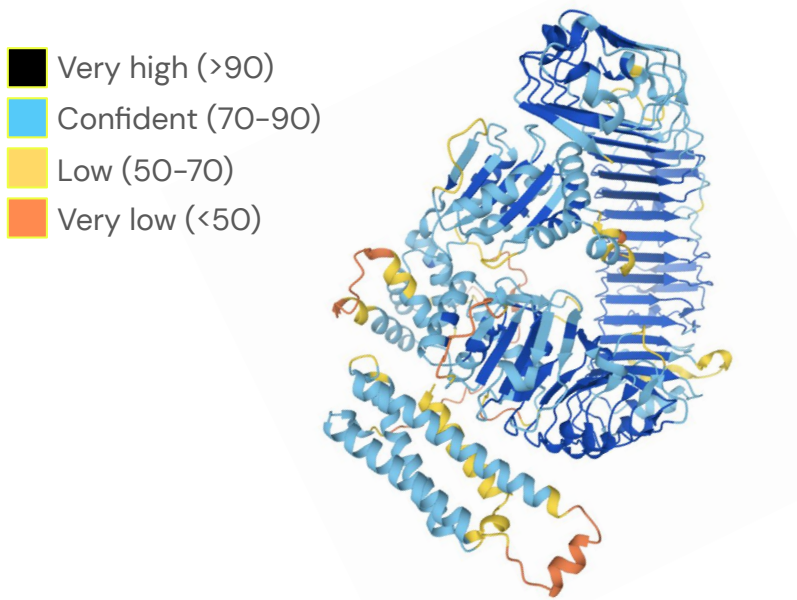
Predicted LDDT: format

pLDDT ranges from 0 to 100 (100 is most confident)

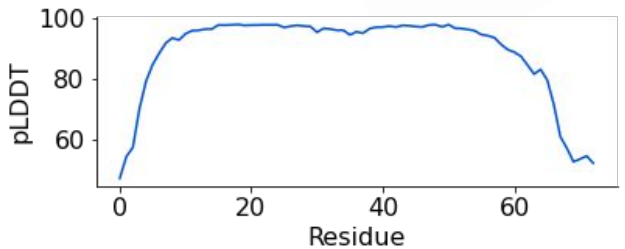
We use a consistent “confidence bands” color scheme when displaying predictions

A pLDDT plot is also displayed by some of our tools

Prediction files always contain pLDDT in the B-factors
Therefore a **higher** B-factor is better!




```
MODEL        0
ATOM      1  N   MET A   1      -9.212  -5.798  33.490  1.00 63.75           N
ATOM      2  CA  MET A   1     -10.075  -6.130  32.322  1.00 63.75           C
ATOM      3  C   MET A   1     -11.469  -6.615  32.714  1.00 63.75           C
ATOM      4  CB  MET A   1      -9.419  -7.112  31.341  1.00 63.75           C
ATOM      5  O   MET A   1     -12.429  -6.075  32.184  1.00 63.75           O
ATOM      6  CG  MET A   1      -8.311  -6.411  30.547  1.00 63.75           C
ATOM      7  SD  MET A   1      -7.766  -7.280  29.061  1.00 63.75           S
ATOM      8  CE  MET A   1      -7.045  -8.751  29.798  1.00 63.75           C
ATOM      9  N   ALA A   2     -11.624  -7.579  33.634  1.00 66.38           N
ATOM     10  CA  ALA A   2     -12.951  -8.096  34.007  1.00 66.38           C
```

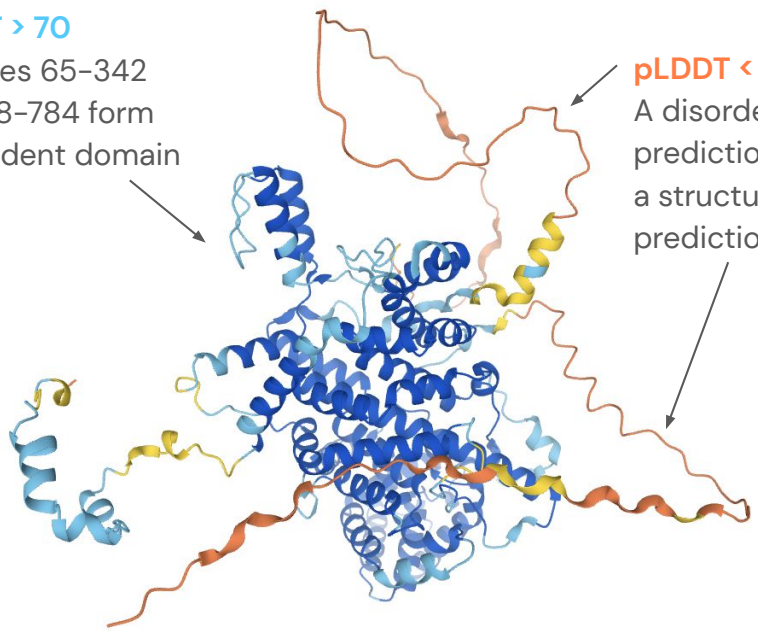


Predicted LDDT: usage

Identifying domains & possible disordered regions

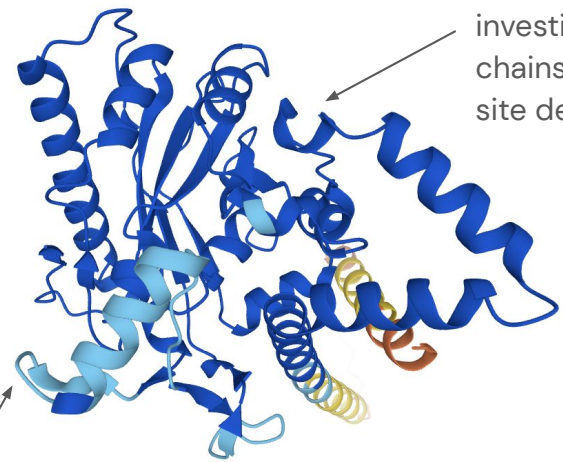
pLDDT > 70
Residues 65-342 and 418-784 form a confident domain

pLDDT < 50
A disorder prediction not a structure prediction



Assessing confidence within a domain

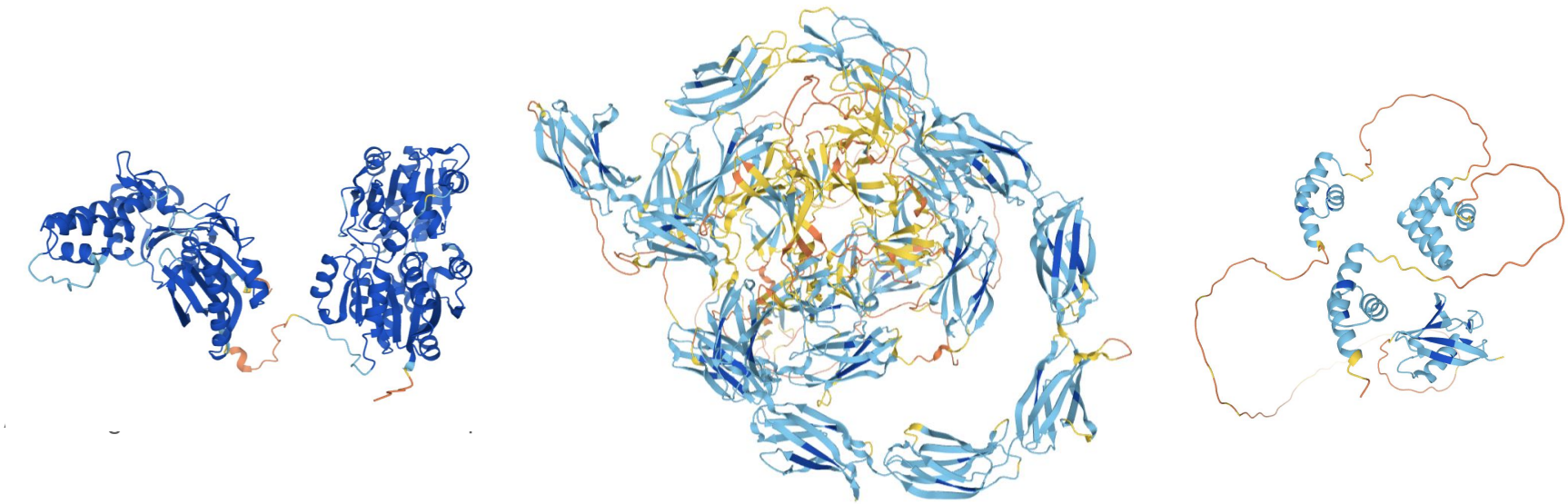
pLDDT > 90
Reasonable to investigate side chains / active site details



pLDDT > 70
Lower confidence on these specific parts

Predicted LDDT: pitfalls

High pLDDT on all domains does **not** imply AlphaFold is confident of their relative positions



Predicted Aligned Error: definition

AlphaFold's prediction of its position error at residue *x*,
if the predicted and the true structures were aligned on residue *y*

PAE aims to measure confidence in the **relative positions** of **pairs of residues**

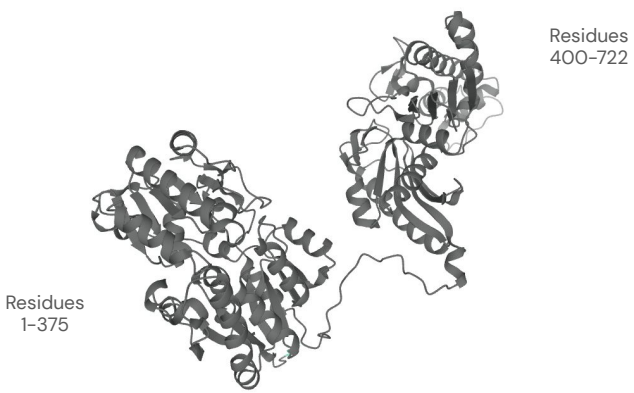
Mainly used to assess relative domain positions, but applicable whenever pairwise confidence is relevant

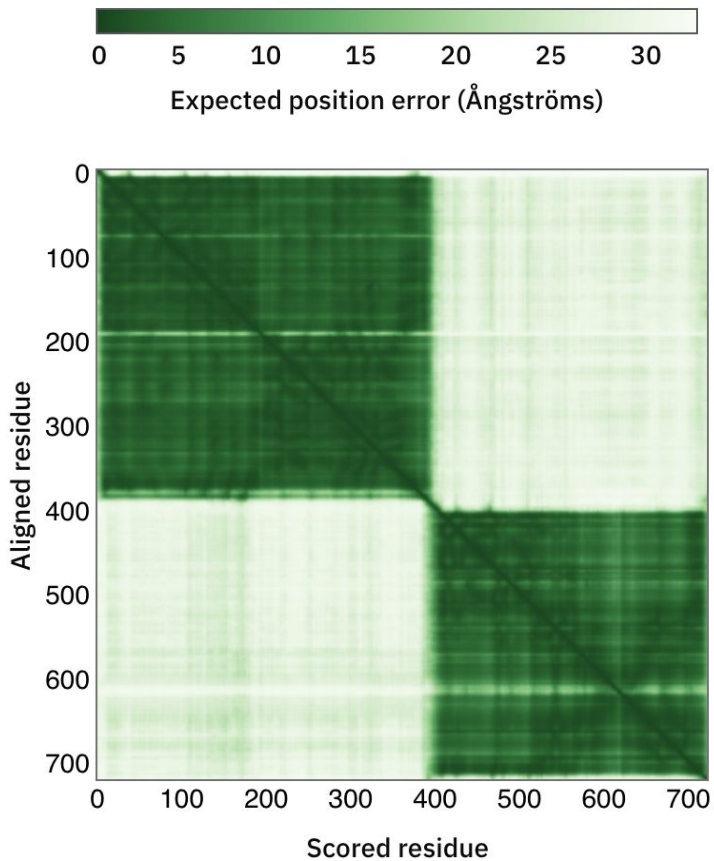
Predicted Aligned Error: format

PAE is displayed as a 2D plot.

Suppose residue *y* were aligned to the true structure and we measured the position error at residue *x*
The color at (*x*, *y*) is AlphaFold's prediction of that error

In this case the squares correspond to two domains



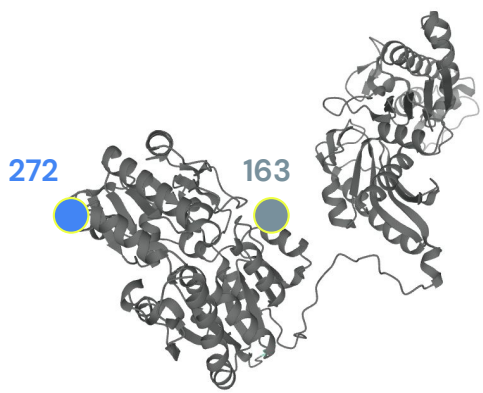


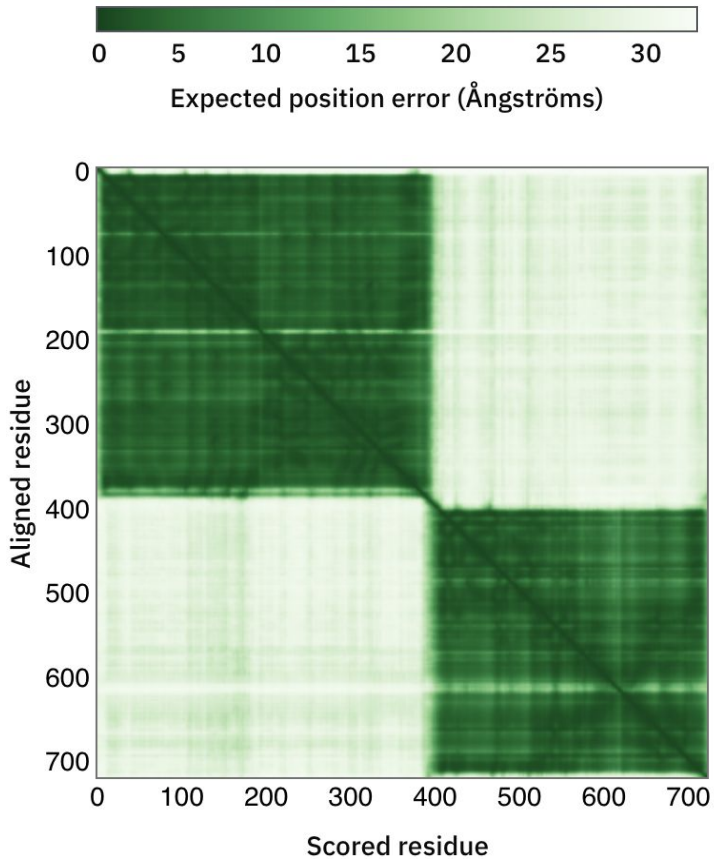
Predicted Aligned Error: format

PAE is displayed as a 2D plot.

Suppose residue *y* were aligned to the true structure
And we measured the position error at residue *x*
The color at (*x*, *y*) is AlphaFold's prediction of that error

AlphaFold is confident in relative positions within each domain...





Predicted Aligned Error: format

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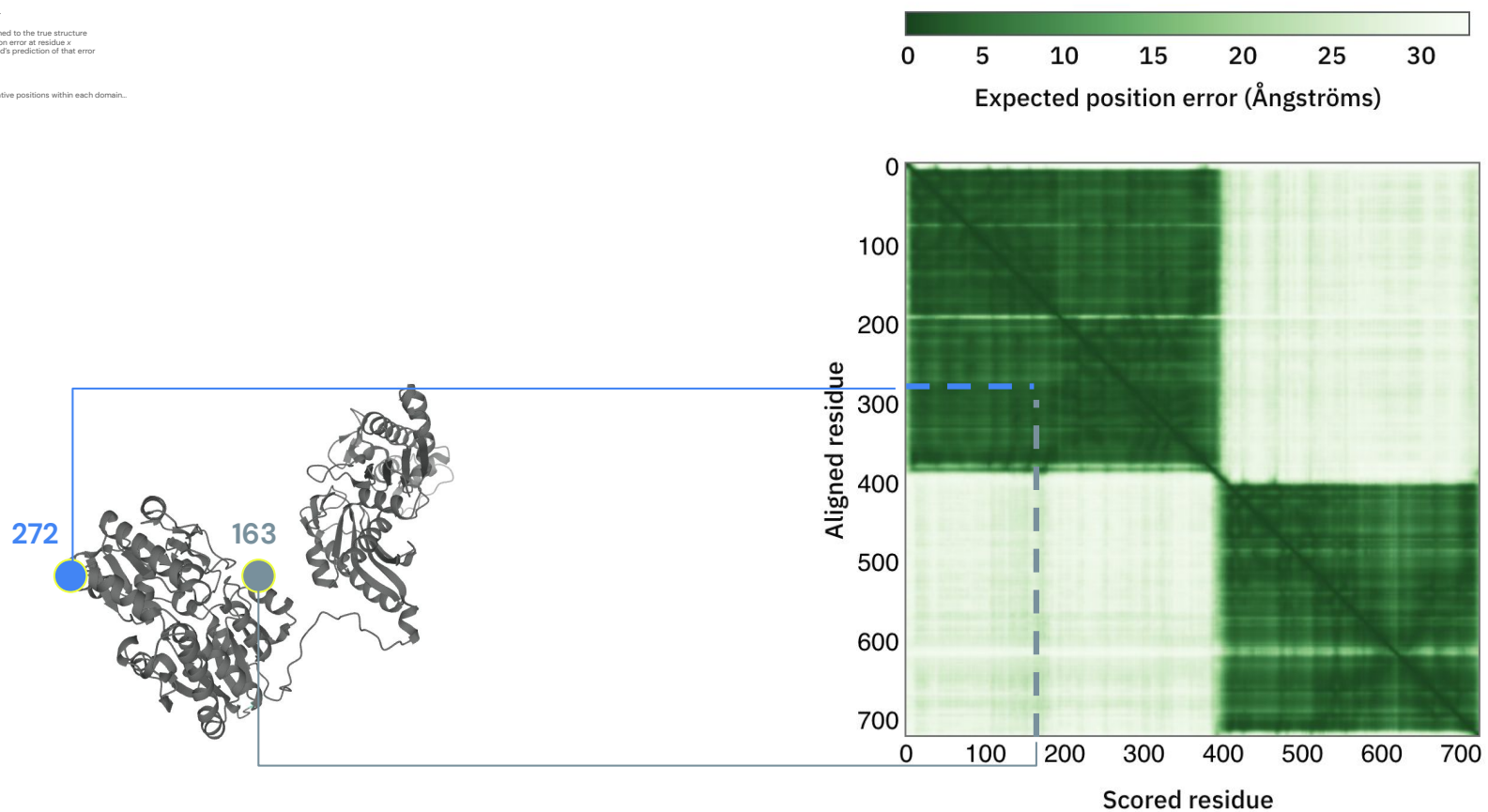
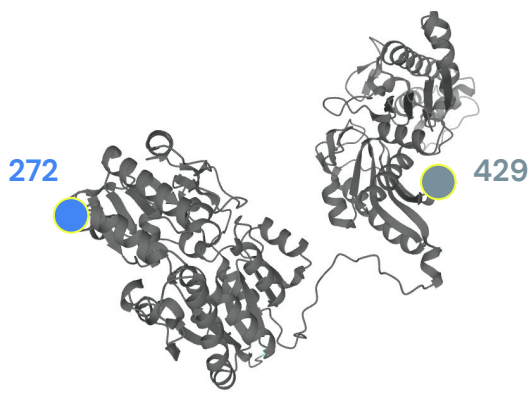


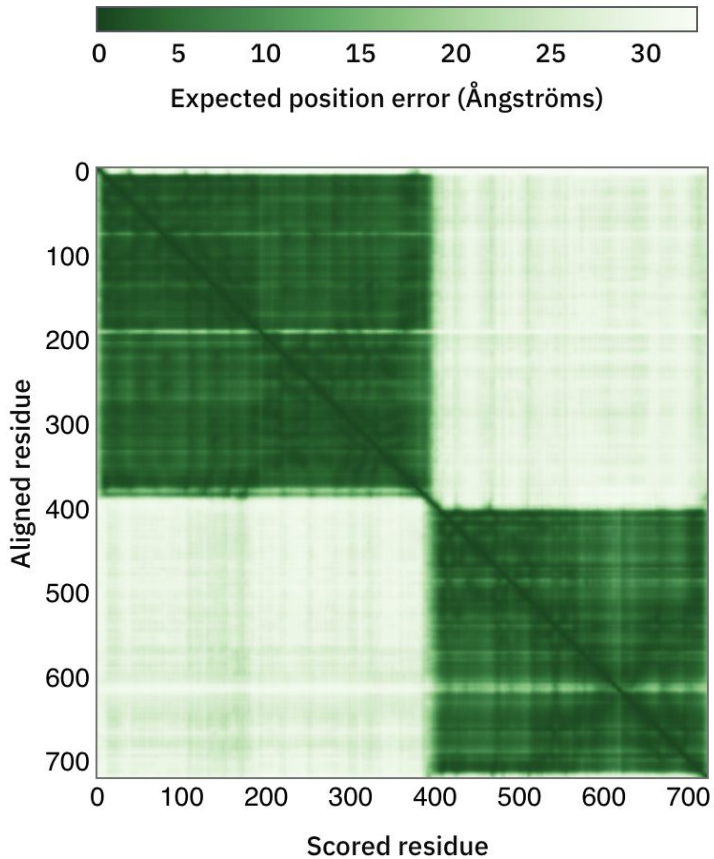
Predicted Aligned Error: format

PAE is displayed as a 2D plot.

Suppose residue *y* were aligned to the true structure
And we measured the position error at residue *x*
The color at (*x*, *y*) is AlphaFold's prediction of that error

...but not between domains.





Predicted Aligned Error: format

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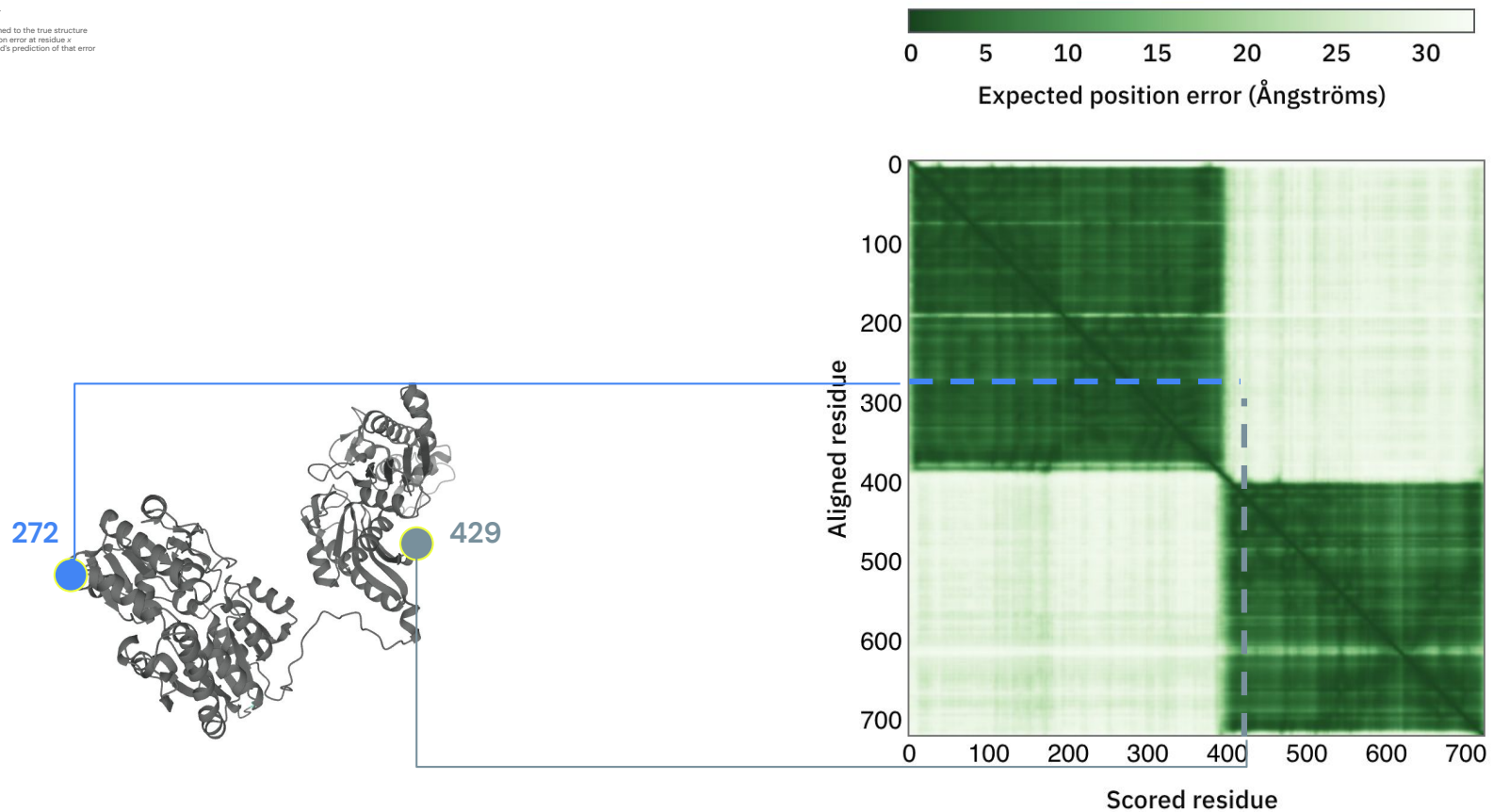
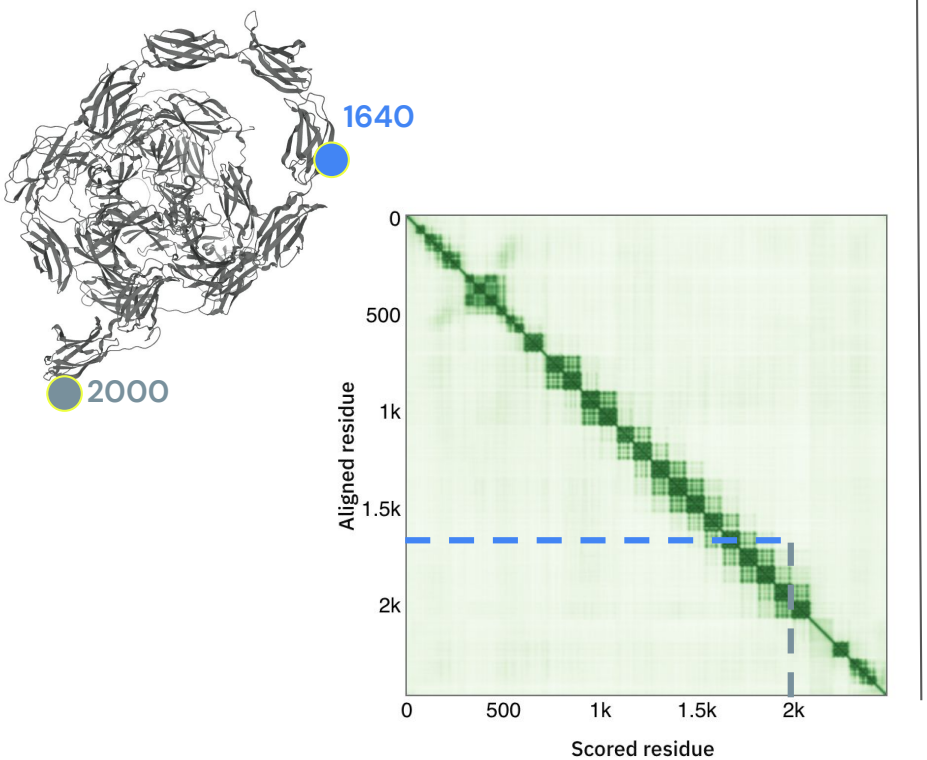
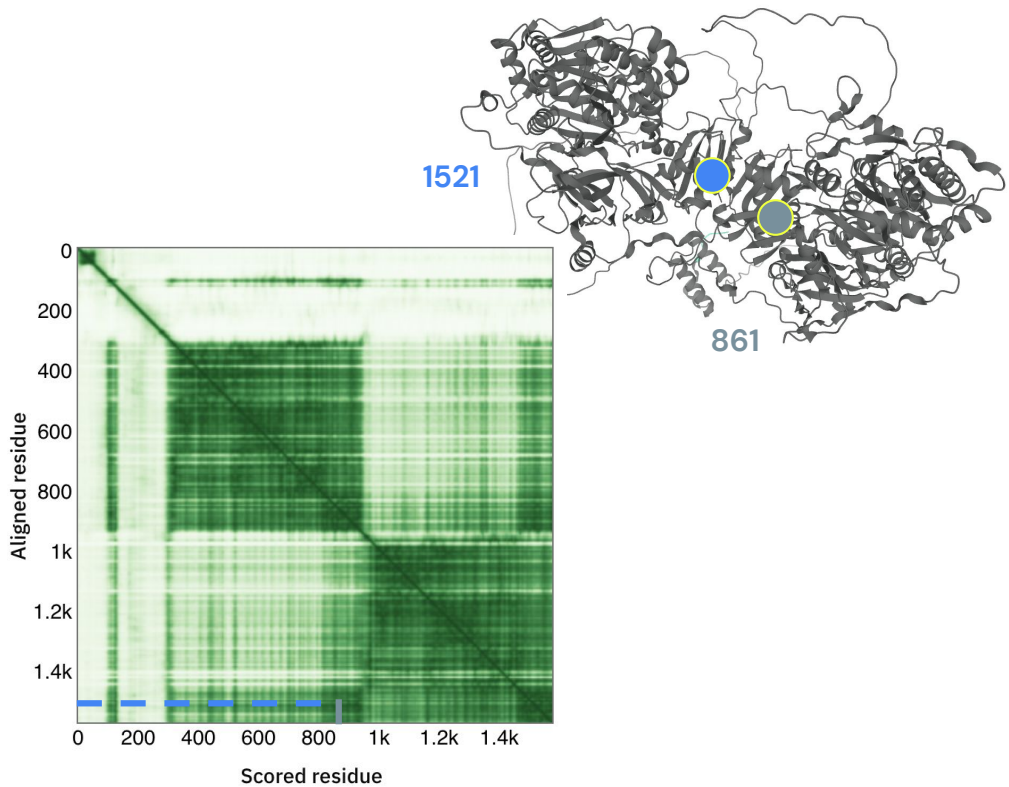


Predicted Aligned Error: usage

No confidence in relative domain positions



Predicted domain packing



Things to be aware of

Uncertain domain placement

- If AlphaFold is uncertain, it won't necessarily place domains sensibly relative to each other
  - Membrane proteins won't leave space for the cell membrane
  - Clashes can occur

Complexes

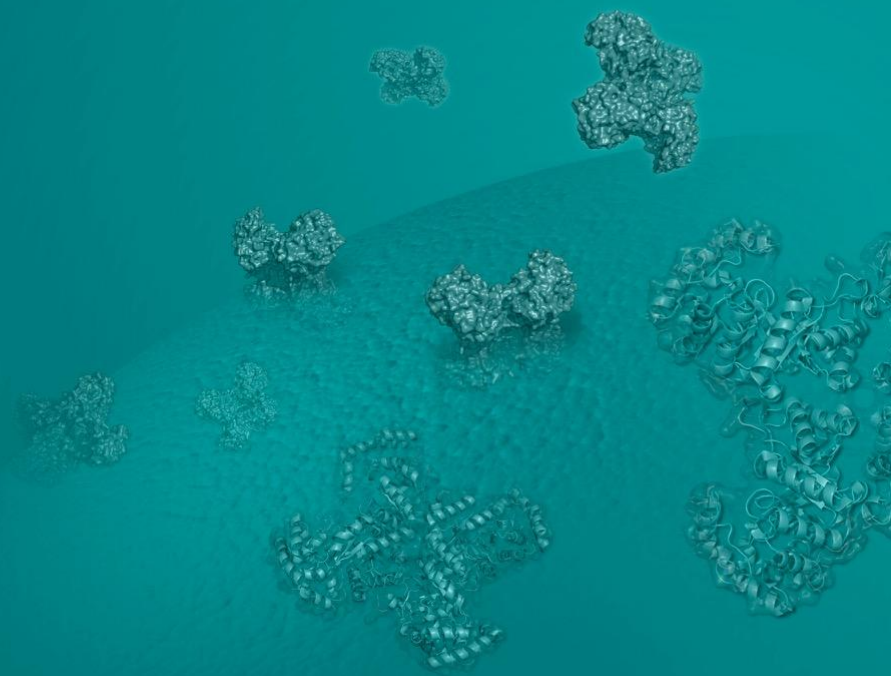
- For proteins that exist in complex, AlphaFold is missing context about their binding partners
  - Heteromers more problematic than homomers
  - Worst case: the protein is flexible in isolation

- Some have had success predicting complexes by joining 2 sequences with a linker
  - We think it is possible to extend the ideas in AlphaFold to complexes
  - However, this linker setup remains to be benchmarked

AlphaFold database

Sameer Velankar

EMBL-EBI



DeepMind

EMBL-EBI

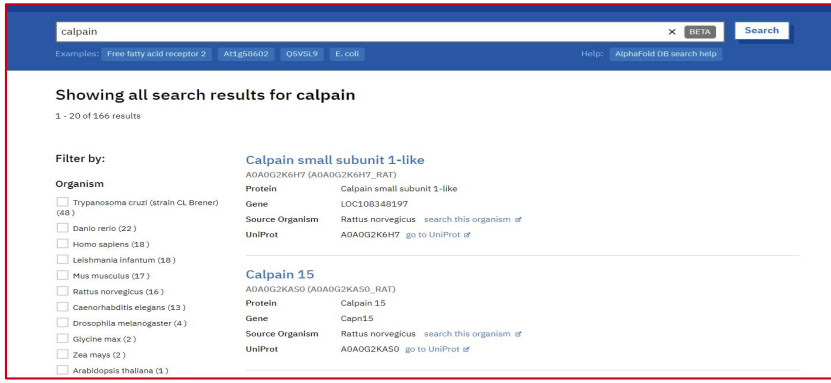
AlphaFold Database

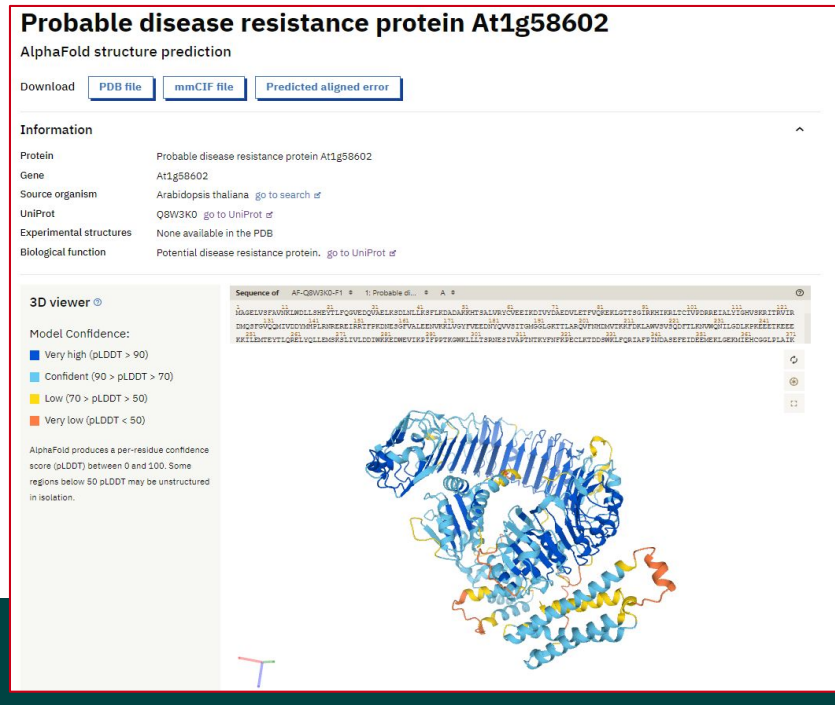
- ~365K predicted structures for proteins from 21 model organisms
- For the organisms currently covered, predicted structures available for sequences in the UniProt reference proteome that are between 16 and 2700 amino acids long and contain only standard amino acids
- Only one prediction (out of 5 independent predictions) is made available in the release
- Accession – AF-P12345-F[1-N]
- Files
  - AF-P12345-F[1-N]-model_V[1-N].[pdb, mmcif]
  - AF-P12345-F[1-N]-predicted-aligned-error_V[1-N].json

AlphaFold web pages

- Basic search system
- Allows search using UniProt accession, UniProt id, protein name, gene name and organism

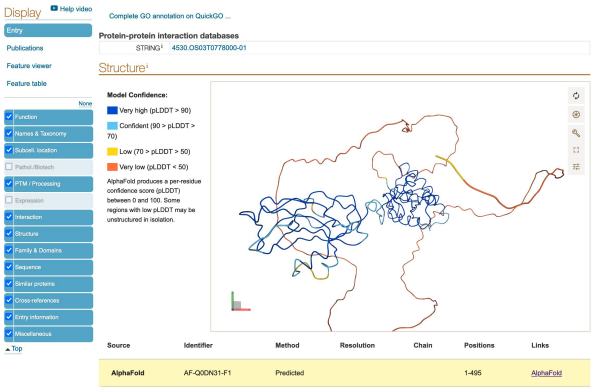
- Clear indication that the structure shown is a prediction
- Allow easy download of structure data
- Basic information about protein
- Clearly indicates if there are experimental structures available
- Display residue-quality information in 3D viewer (pLDDT – predicted Local Distance Difference Test)
- Predicted Aligned Error (PAE viewer)



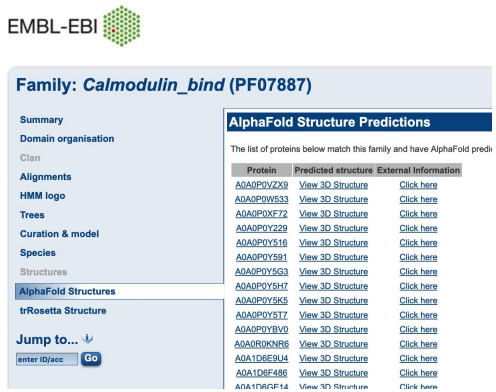


Models available across EBI resources

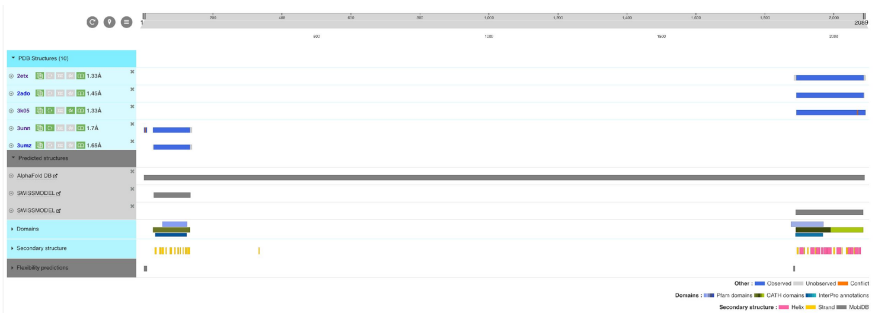
UniProt



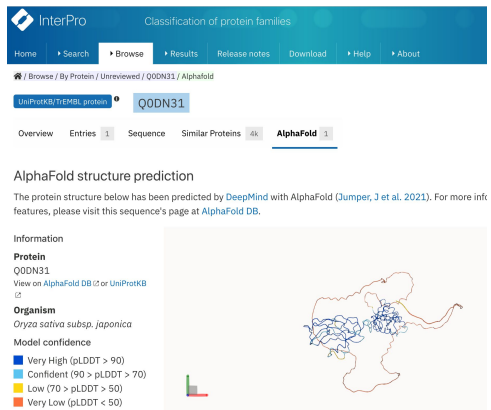
Pfam



PDBe-KB



InterPro



AlphaFold Database – limitations

- Information on complexes with other proteins, nucleic acids (DNA or RNA) or ligands. In some cases, the single-chain prediction may correspond to the structure adopted in a complex. The missing context from surrounding molecules may lead to an uninformative prediction
  - AlphaFold does not make any predictions about any of the non-protein components such as cofactors, metals, ligands including drug-like molecules, ions, carbohydrates and other post-translational modifications
- Protein dynamics - AlphaFold will usually only produce one of multiple conformations
- AlphaFold has not been trained or validated for predicting the effect of mutations
- May (or may not) lead to hypotheses about protein function – any hypotheses have to be tested by further experimentation

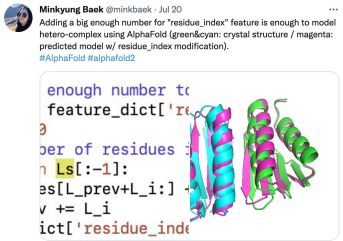
What's next – under discussion

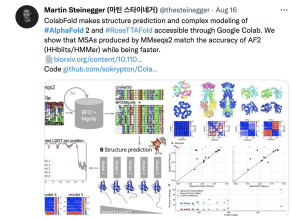
- Remove signal peptides from predictions
- Making 5 independent predictions available for each protein
- Additional metadata
  - MSA – need to consider data size
  - information on templates
  - quality criteria e.g. predicted TM score
- Updating database to UniRef90 dataset (~130 million structures)

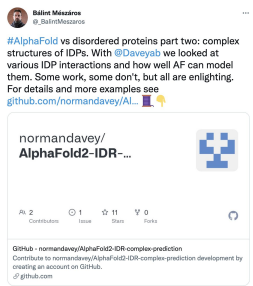
Impact of AlphaFold database on life science research

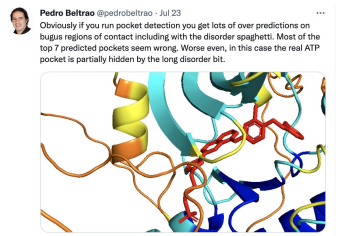
Structural bioinformatics – (structure/function)

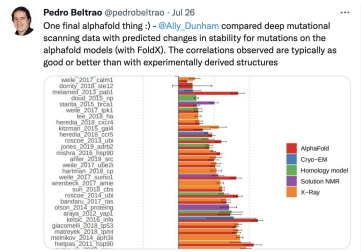
- Predicting complexes between macromolecules
  - Homo- and Hetero- Protein-protein; Protein-nucleic acid complexes
  - Intrinsically disordered proteins
- Provide information on protein dynamics
  - Relevant conformational states
- Functionally important residues
  - Impact of mutation; Binding sites; Conformationally important residues
  - Interfaces
- Ligand prediction – What binds?
  - What might bind in a pocket





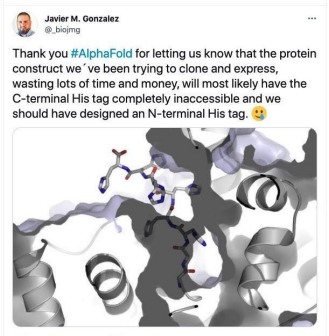


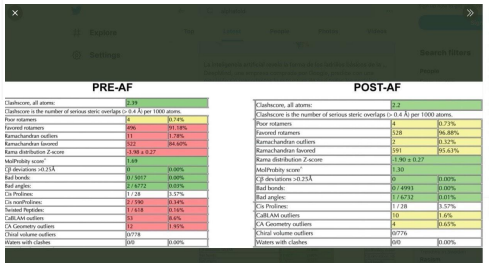


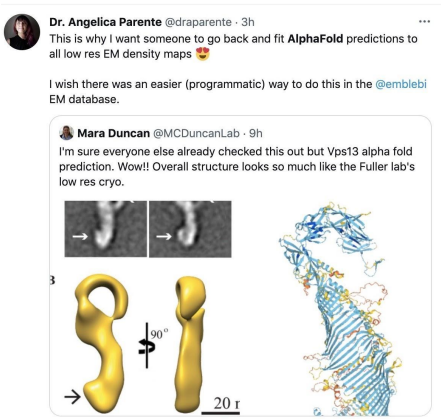


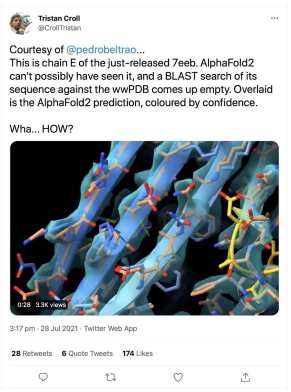
Structural biology

- Accelerating structure studies
  - Improved construct design
  - Starting model for structure determination
  - Fitting models in low resolution EM maps
  - Time resolved studies to understand mechanism





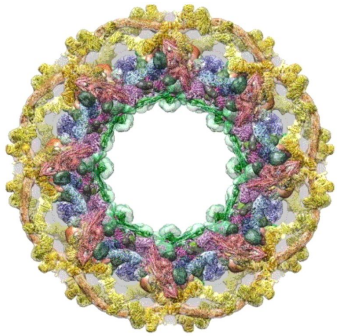




Structural biology

- Integrative/hybrid methods
  - Models for individual components

- Combination of sparse experimental data and predicted model may lead to actionable data to test hypothesis
  - Chemical foot printing
  - Hydrogen-Deuterium exchange
  - smFRET - Single molecule fluorescence resonance energy transfer



I/H Methods Structures
552-protein yeast Nuclear Pore Complex
Kim et al. (2018) *Nature 555*, 475-82
PDBDEV_00000010; PDBDEV_00000011; PDBDEV_00000012

Validation et impact d'AlphaFold en 2024

"AlphaFold two years on: Validation and impact"

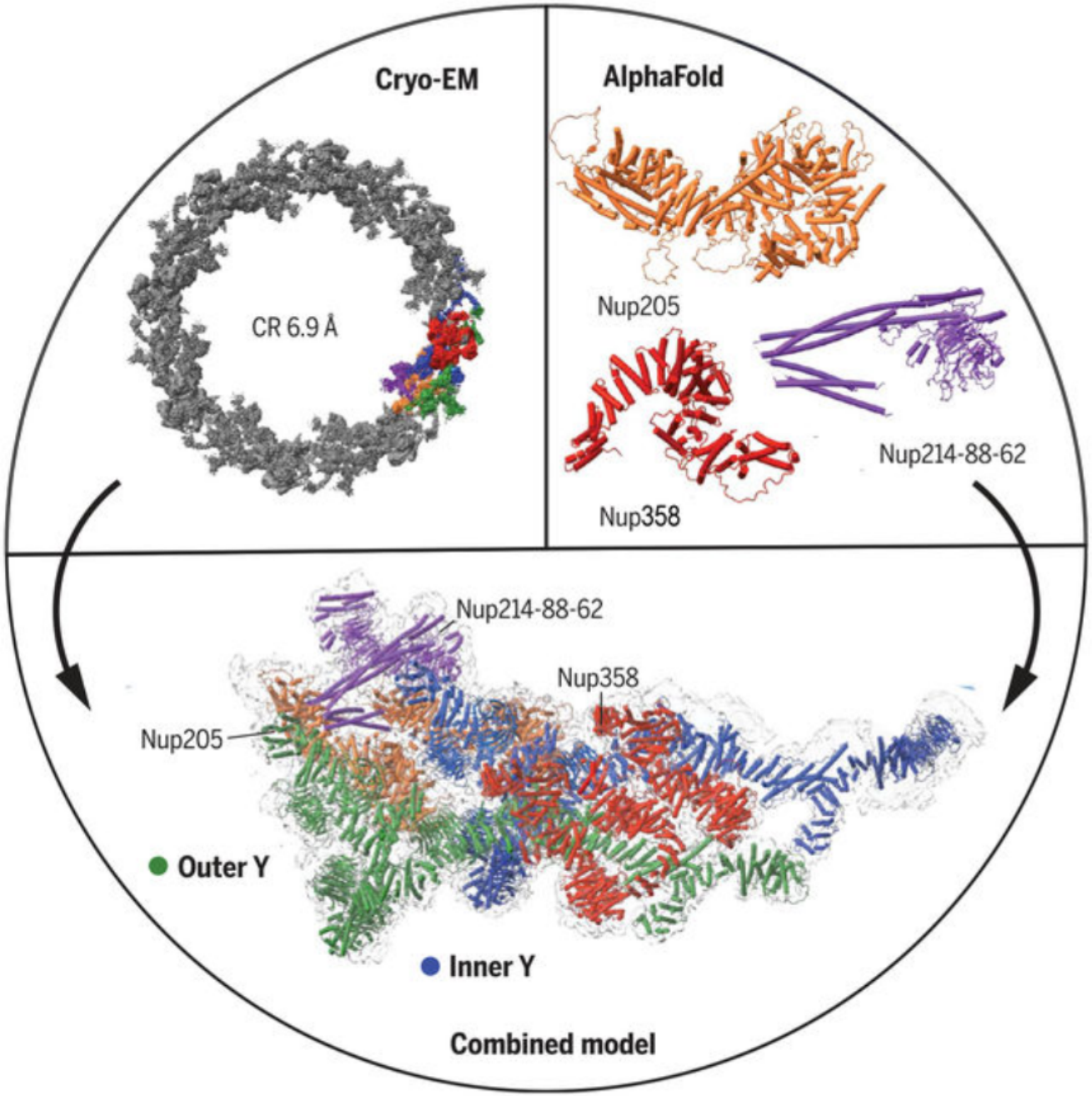
revue de Kovalevskiy et al. dans PNAS de mars 2024 (soumis octobre 2023)

Oleg Kovalevskiy, Juan Mateos-Garcia, and Kathryn Tunyasuvunakool (Aug. 2024). In: *Proceedings of the National Academy of Sciences* 121.34

1. Impact on Experimental Structure Determination
2. Predicting Protein-Protein Interactions
3. Use in Protein Design
4. Enabling New Computational Work
5. Experimental Validation of AlphaFold Models
   - Single Chains
   - Complexes
   - Biologically Relevant States

Impact on Experimental Structure Determination

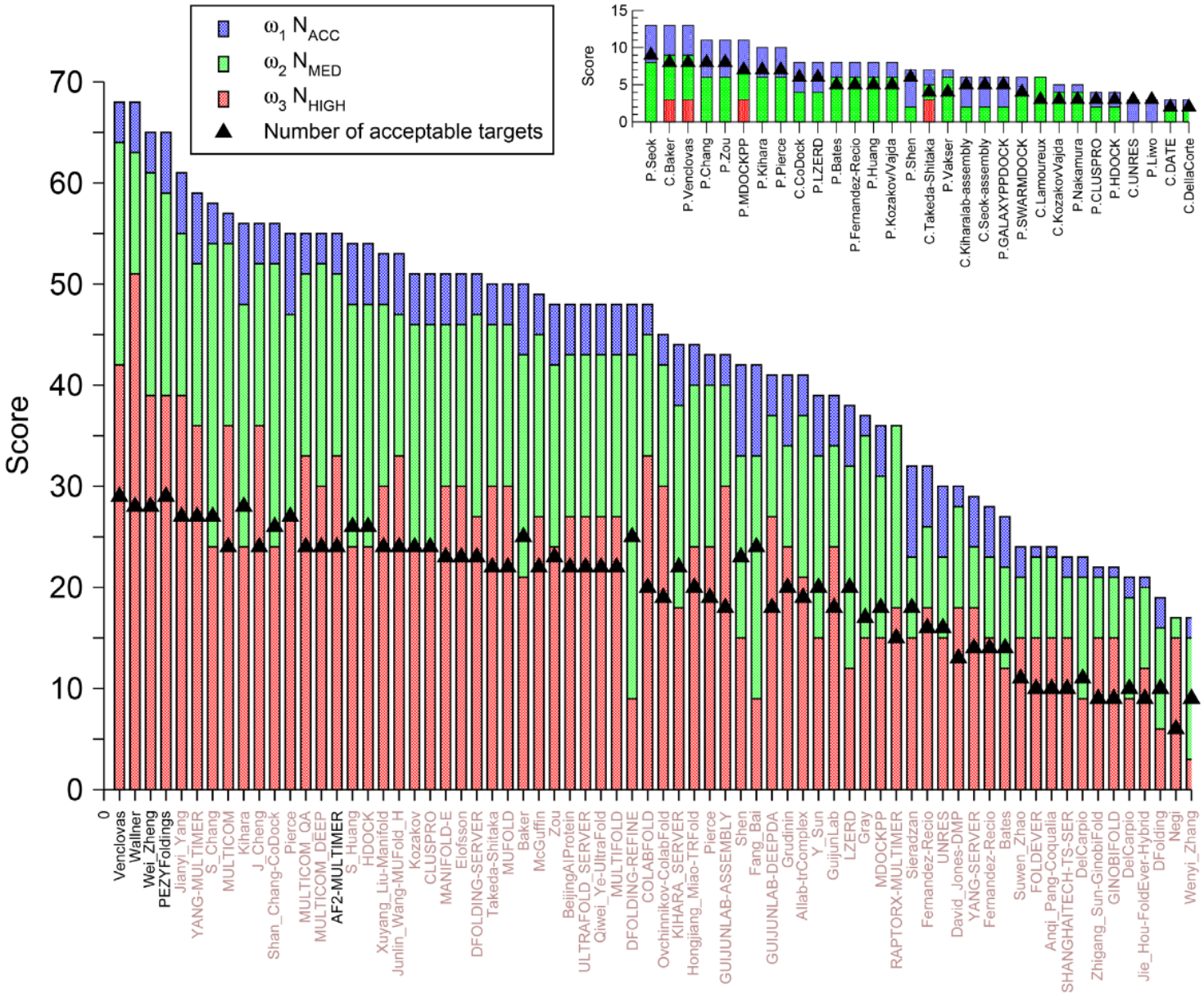
1. X-ray crystallography
   - Phase problem => Molecular Replacement solved by AlphaFold
   - on challenging cases where no PDB template can be found
   - on novel folds
   - de novo design proteins
   - automated process with AlphaFold
2. Cryo-EM
   - Combine cryo-EM with AlphaFold
   - example : Nuclear pore complex (120 MDa)
   - find structure in AlphaFoldDB for unknown subunits in cryo-EM map


Cryo-EM
CR 6.9 Å
AlphaFold
Nup205
Nup214-88-62
Nup358
Nup214-88-62
Nup358
Nup205
Outer Y
Inner Y
Combined model

Predicting protein-protein Interactions

- AlphaFold-Multimer (2021)
- AlphaFold3

CASP 15 (2022): joint CASP-CAPRI experiment



CASP 15 (2022): joint CASP-CAPRI experiment

- Huge improvement from previous CASP-CAPRI (2020) experiment, due to AlphaFold 2 and AlphaFold-Multimer
- for 40% of the targets high quality models have been obtained (previously: only 8%)
- Webservers are on par with human groups
- Antibodies and Nanobodies remain challenging
- Targets with important conformational flexibility remain challenging

Marc F. Lensink et al. (2023). en. In: *Proteins: Structure, Function, and Bioinformatics* 91.12

Conclusion

Is the protein-protein docking problem solved ?
Challenges:

- Better sampling and scoring
- Conformational changes upon binding
- Predicting domain motions
- Folding upon binding
- Large scale docking => Interactome, Large molecular assemblies
- Predicting which proteins interact => Predicting binding affinities

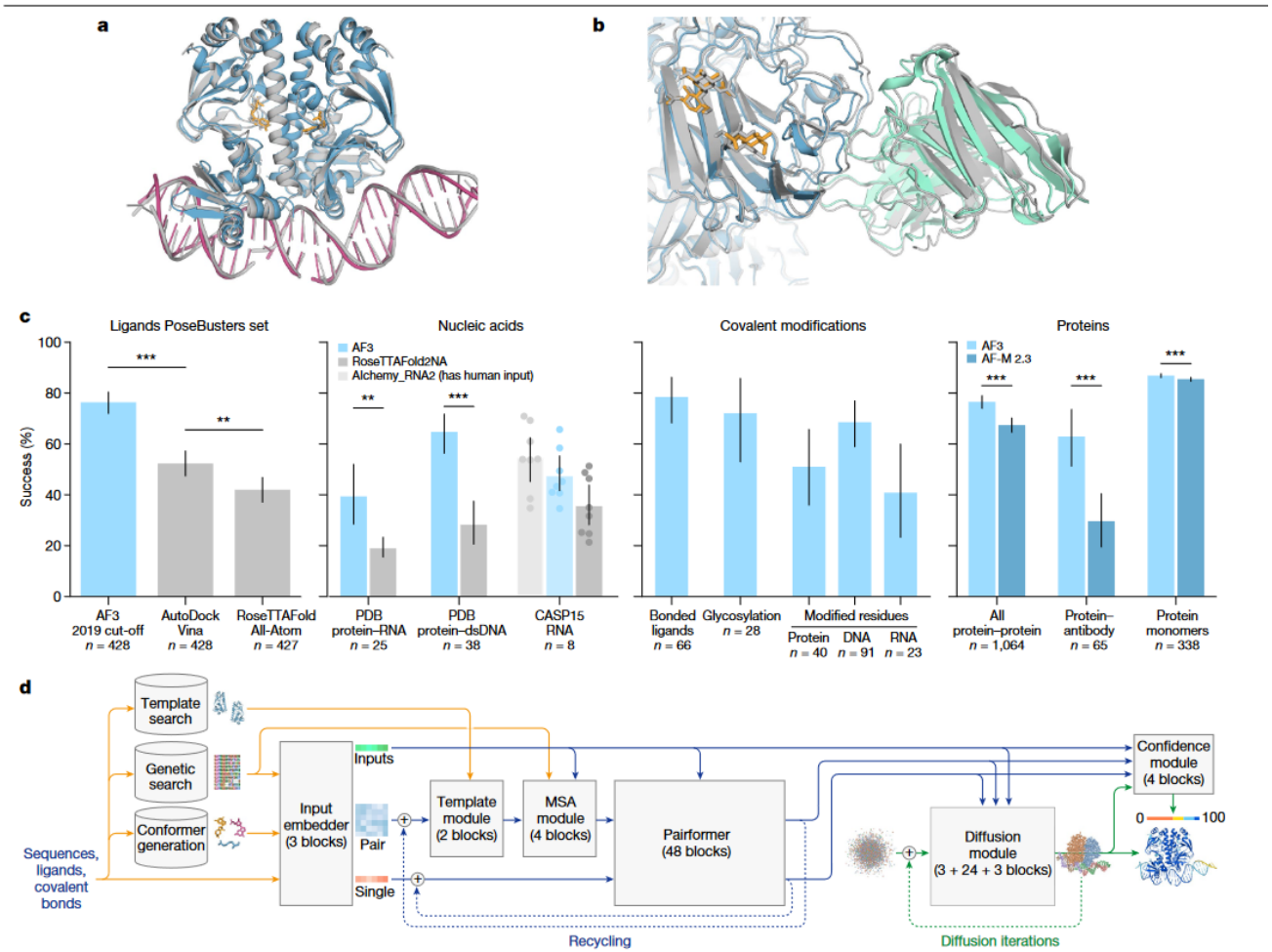
AlphaFold3 et co.

AlphaFold3

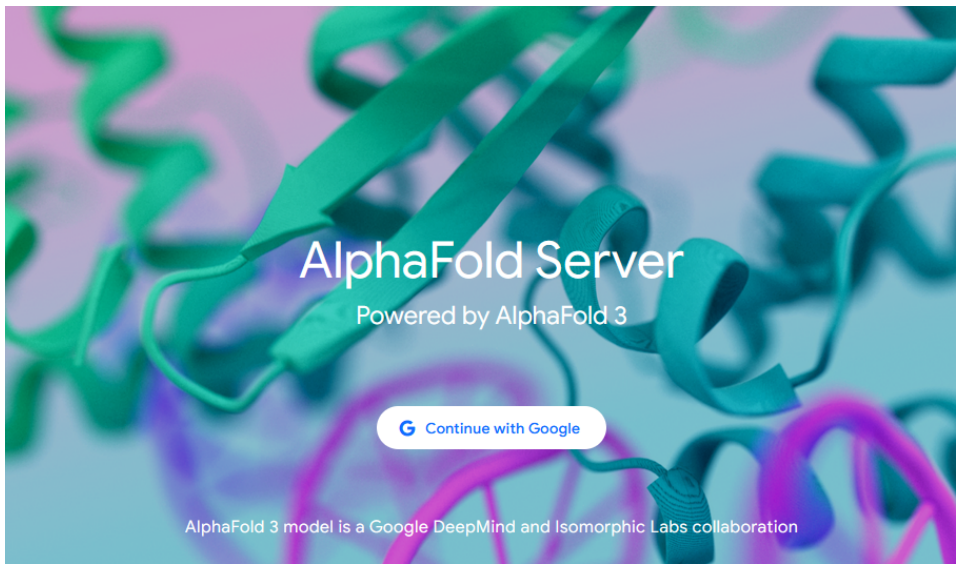
collaboration DeepMind et Isomorphic Labs

Josh Abramson et al. (June 2024). en. In: *Nature* 630.8016

cité 770 fois depuis mai 2024 (8 oct 2024)



AlphaFold3



Terms of use and attribution

AlphaFold Server is for **non-commercial use only, subject to AlphaFold Server Terms of Service**. AlphaFold Server output cannot be used **in docking or screening tools** or to train machine learning models or related technology for biomolecular structure prediction similar to AlphaFold Server.

If you use an AlphaFold Server prediction, please cite our paper: Abramson, J et al. Accurate structure prediction of biomolecular interactions with AlphaFold 3. Nature (2024).

AlphaFold3 clone: HelixFold3

Serveur:
`https://paddlehelix.baidu.com/app/all/helixfold3/forecast`
Github (= code source):
`https://github.com/PaddlePaddle/PaddleHelix`
publie en preprint:

Lihang Liu et al. (Aug. 2024). en

AlphaFold3 clone: Chai-1

Serveur:
https://www.chaidiscovery.com/blog/introducing-chai-1
Github (= code source):
https://github.com/chaidiscovery/chai-lab
publie en preprint:
https://chaiassets.com/chai-1/paper/technical_report_v1.pdf

HelixFold3, AlphaProteo, Chai-1

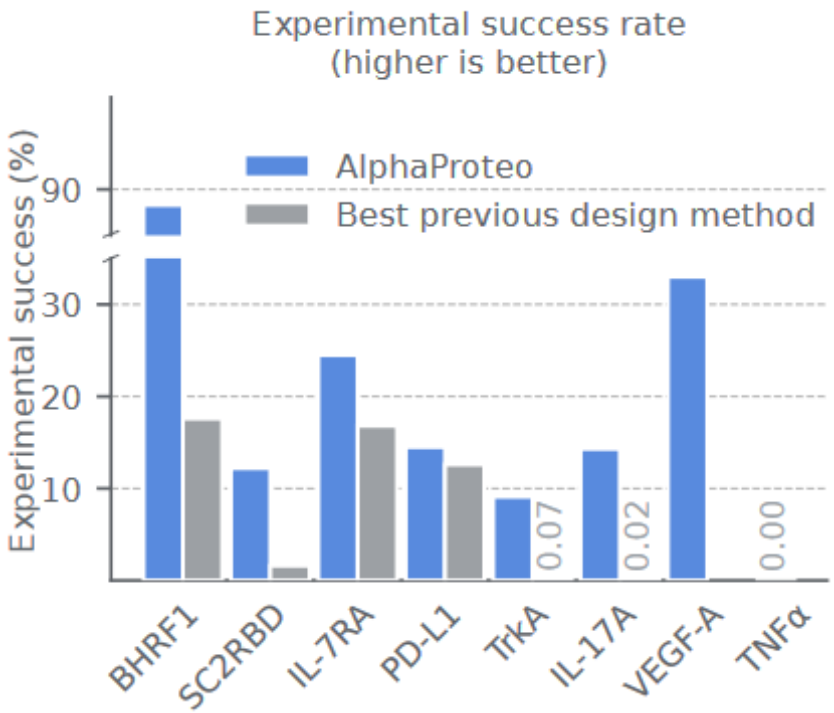
https://newsletter.towardsai.net/p/tai-116-rise-of-the-protein-foundation

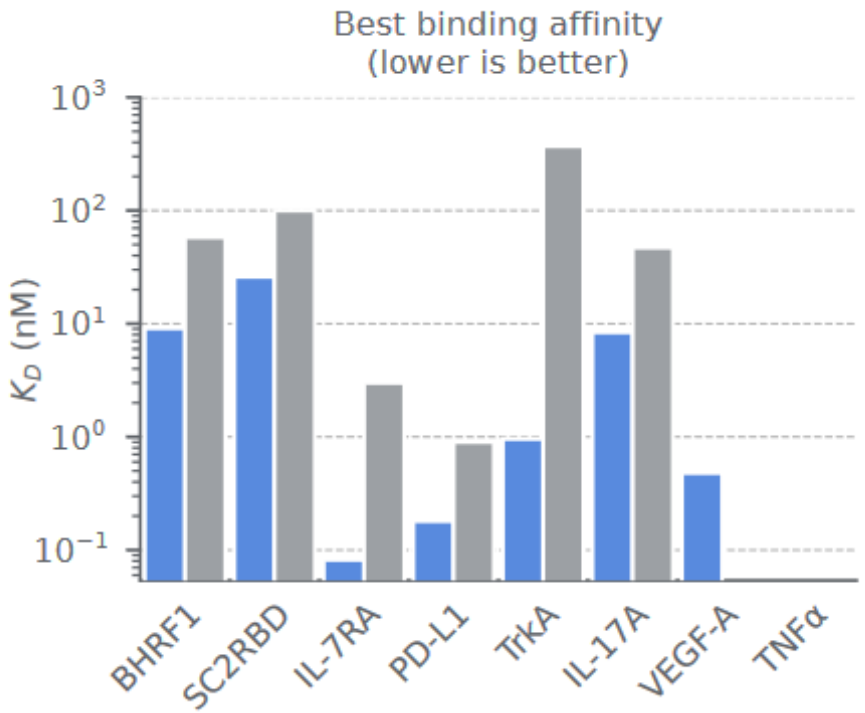
Model	AlphaFold-3 (May 2024)	HelixFold3 (Aug 2024)	AlphaProteo (Sep 2024)	Chai-1 (Sep 2024)
Focus	Predicting the structure and interactions of biomolecules, including proteins, DNA, RNA, ligands, ions.	Replicating AlphaFold-3 functionalities; structures of proteins, nucleic acids, and small molecule ligands.	Designing novel protein binders based on target molecule structure and binding locations.	Multi-modal structure prediction for proteins, small molecules, DNA, and RNA.
Architecture	New diffusion-based architecture. Uses a simplified "pairformer" module for reduced MSA processing and a diffusion module for direct prediction of atom coordinates.	Insights from AlphaFold-3 and previous HelixFold versions utilize Evoformer and diffusion networks.	Generative model. Alphafold distillation. Design scoring system and filter.	Leverages pair-bias self-attention, similar to AlphaFold-3. Incorporates language model embeddings for single-sequence prediction and constraint features to utilize experimental data.
Training Data	PDB data released before September 30, 2021, enriched with structures predicted by AlphaFold-Multimer (cross-distillation).	Protein Data Bank (PDB) targets released before September 30, 2021, self-distillation datasets.	Protein Data Bank (PDB) and over 100 million AlphaFold predicted structures.	PDB structures released before January 12, 2021, and AlphaFoldDB structures.
Key Capabilities	Improved accuracy in protein-ligand, protein-nucleic acid, and antibody-antigen predictions compared to specialized tools and AlphaFold-Multimer.	Comparable accuracy to AlphaFold-3 in predicting protein, nucleic acid, and ligand structures. Open-source for academic research.	Designs high-affinity protein binders with higher success rates than existing methods. Experimentally validated on various targets.	Achieves state-of-the-art performance on various structure prediction benchmarks. Strong protein-ligand and multimer prediction, including antibody-antigen complexes.
Limitations	Limitations in stereochemistry, the potential for hallucinations in some regions, and challenges with antibodies. Not fully open-sourced; access via AlphaFold Server. Restrictions on ligands and modifications.	Accuracy in protein-protein complex prediction still lags behind AlphaFold-3.	Unable to design successful binders for all targets (e.g., TNFα).	Sensitivity to modified residues and difficulty in accurately predicting relative orientations of chains in a complex. Model weights and code available for non-commercial use; commercial access through a web interface.

AlphaProteo

collaboration DeepMind et Isomorphic Labs

Vinicius Zambaldi et al. (Sept. 2024).





"Google DeepMind WetLab"

https://www.crick.ac.uk/news/2022-07-06_the-francis-crick-institute-and-deepmind-join-forces-t

Prix Nobel 2024 en Chimie

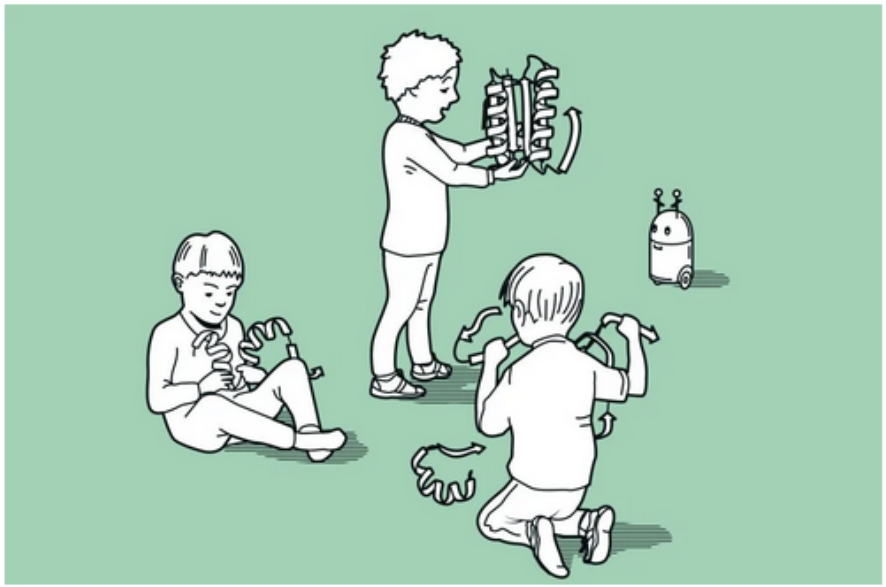
The Nobel Prize in Chemistry 2024

They cracked the code for proteins' amazing structures

The Nobel Prize in Chemistry 2024 is about proteins, life's ingenious chemical tools. David Baker has succeeded with the almost impossible feat of building entirely new kinds of proteins. Demis Hassabis and John Jumper have developed an AI model to solve a 50-year-old problem: predicting proteins' complex structures. These discoveries hold enormous potential.

Related articles

Press release
Popular information: They have revealed proteins' secrets through computing and artificial intelligence
Scientific background: Computational protein design and protein structure prediction



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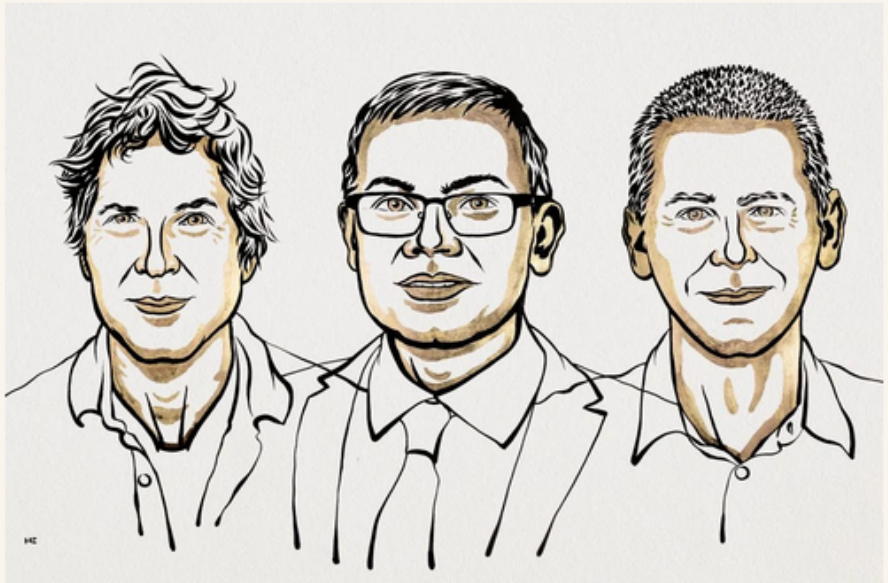
https://www.nobelprize.org/prizes/chemistry/2024

Prix Nobel 2024 en Chimie

The 2024 chemistry laureates

The Nobel Prize in Chemistry 2024 was awarded with one half to David Baker "for computational protein design" and the other half jointly to Demis Hassabis and John M. Jumper "for protein structure prediction".

Demis Hassabis and John Jumper have successfully utilised artificial intelligence to predict the structure of almost all known proteins. David Baker has learned how to master life's building blocks and create entirely new proteins.



David Baker, Demis Hassabis and John Jumper. Ill. Niklas Elmehed © Nobel Prize Outreach

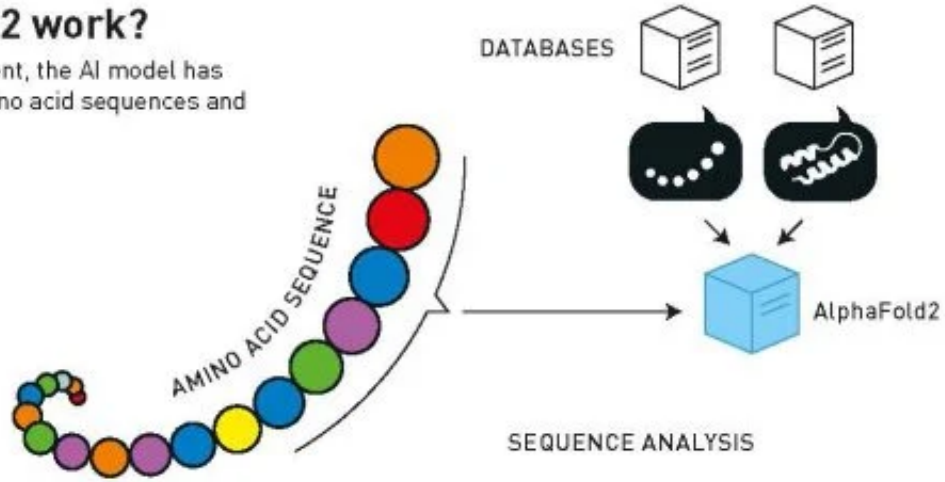
https://www.nobelprize.org/prizes/chemistry/2024

Prix Nobel 2024 en Chimie

https://www.nobelprize.org/prizes/chemistry/2024/popular-information/

How does AlphaFold2 work?

As part of AlphaFold2's development, the AI model has been trained on all the known amino acid sequences and determined protein structures.



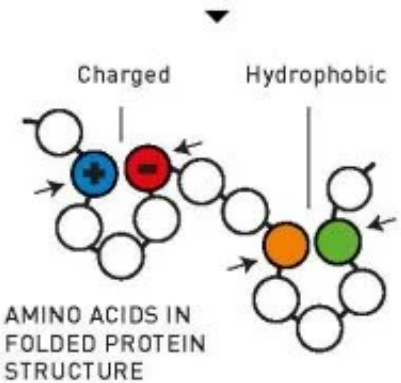
1. DATA ENTRY AND DATABASE SEARCHES

An amino acid sequence with unknown structure is fed into AlphaFold2, which searches databases for similar amino acid sequences and protein structures.

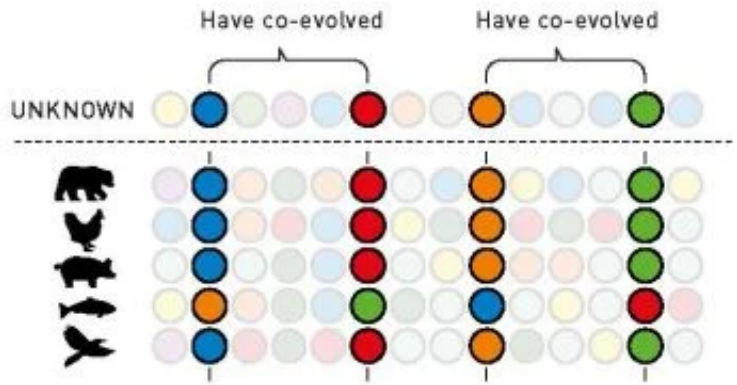
2. SEQUENCE ANALYSIS

The AI model aligns all the similar amino acid sequences – often from different species – and investigates which parts have been preserved during evolution.

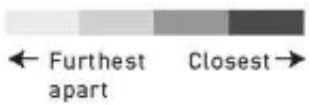
In the next step, AlphaFold2 explores which amino acids could interact with each other in the three-dimensional protein structure. Interacting amino acids co-evolve. If one is charged, the other has the opposite charge, so they are attracted to each other. If one is replaced by a water-repellent (hydrophobic) amino acid, the other also becomes hydrophobic.

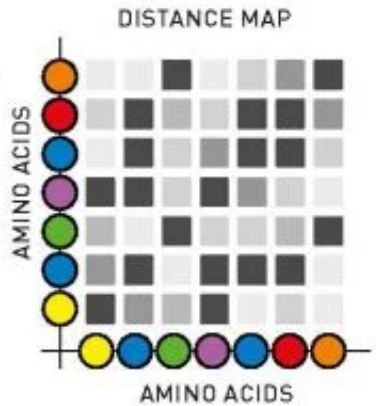


SEQUENCE ANALYSIS



Using this analysis, AlphaFold2 produces a distance map that estimates how close amino acids are to each other in the structure.



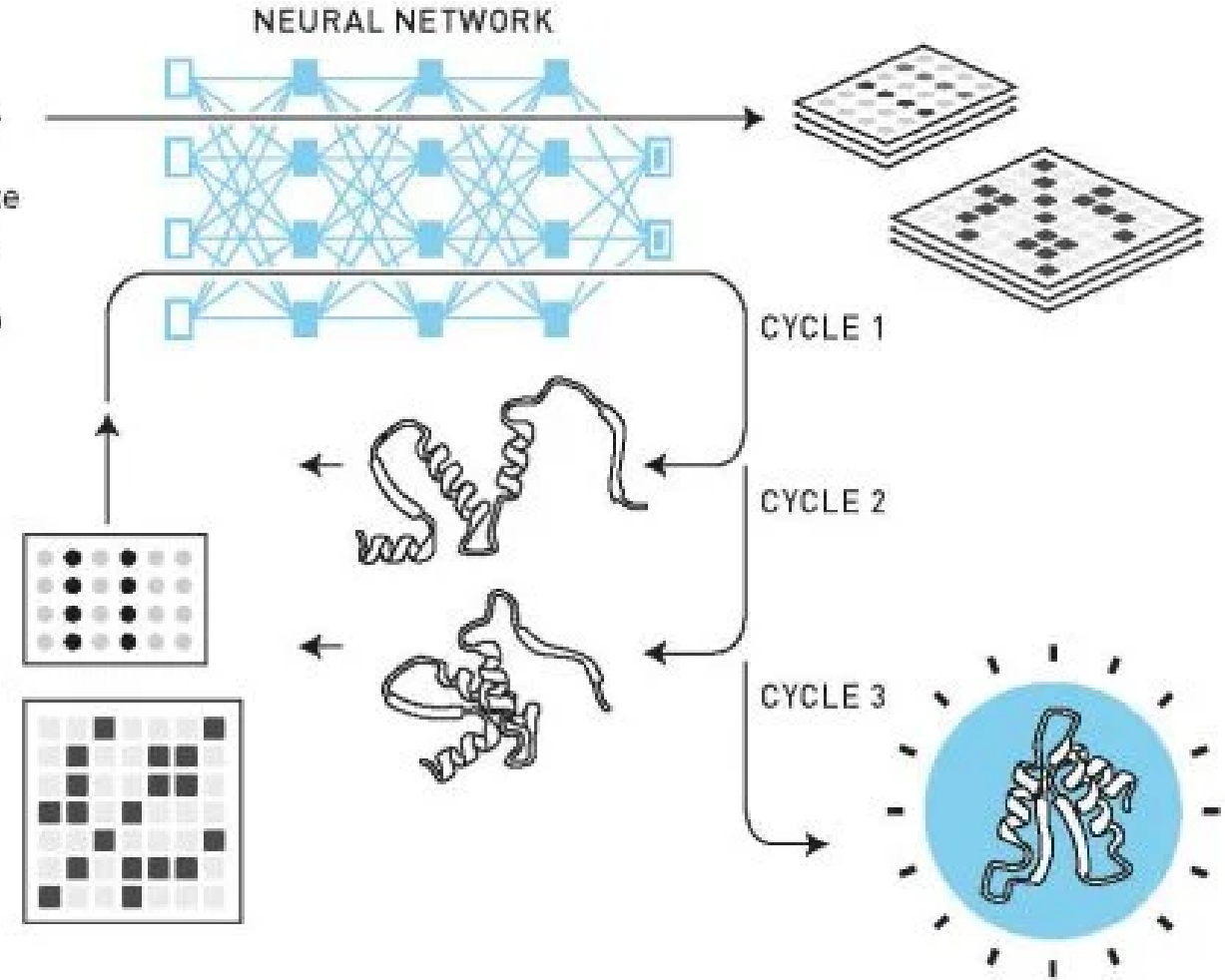


3. AI ANALYSIS

Using an iterative process, AlphaFold2 refines the sequence analysis and distance map. The AI model uses neural networks called transformers, which have a great capacity to identify important elements to focus on. Data about other protein structures – if they were found in step 1 – is also utilised.

4. HYPOTHETICAL STRUCTURE

AlphaFold2 puts together a puzzle of all the amino acids and tests pathways to produce a hypothetical protein structure. This is re-run through step 3. After three cycles, AlphaFold2 arrives at a particular structure. The AI model calculates the probability that different parts of this structure correspond to reality.



Prix Nobel 2024 en Chimie

https://www.nobelprize.org/uploads/2024/10/advanced-chemistryprize2024.pdf

The end

- MERCI pour votre attention!

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