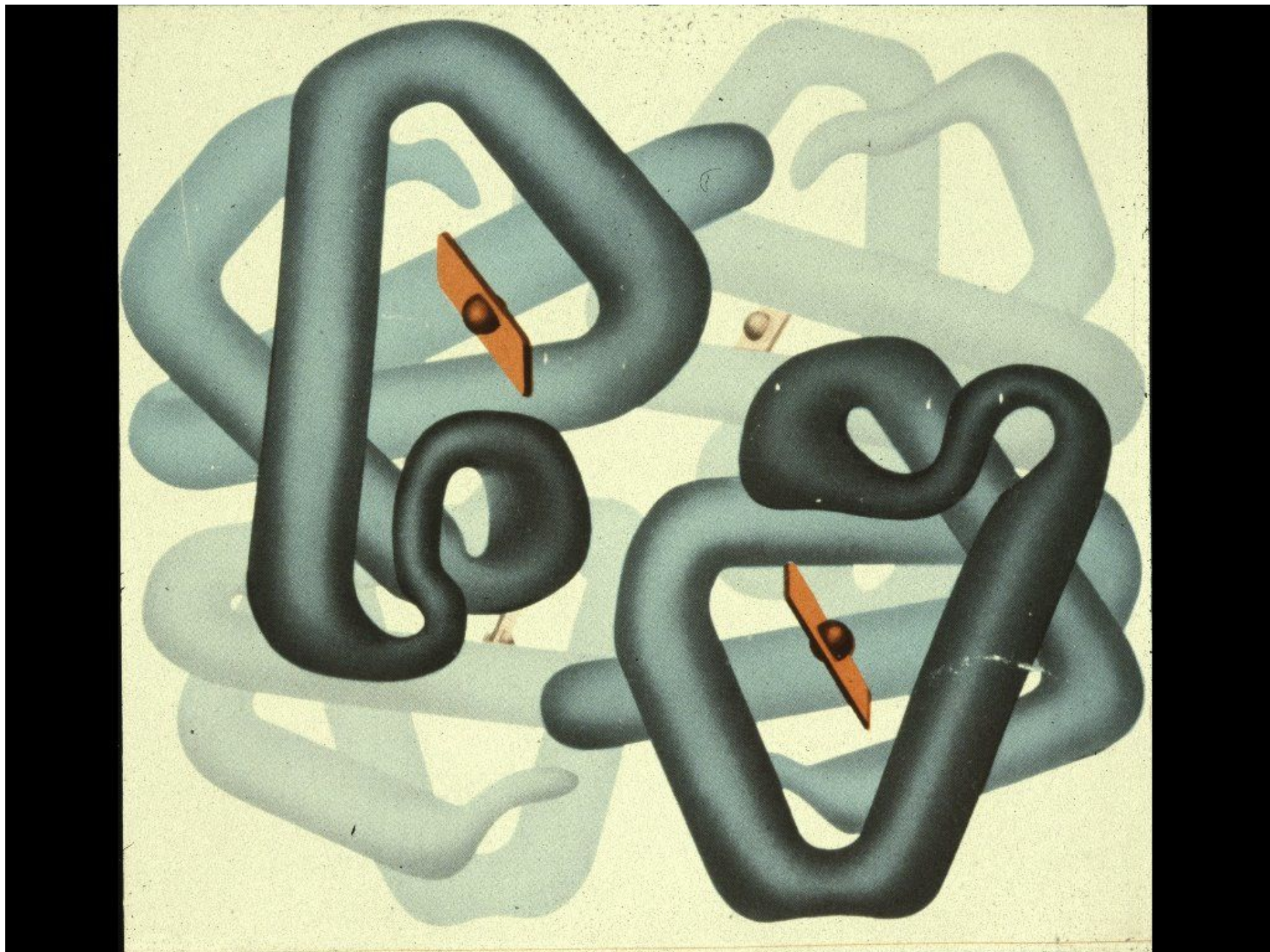


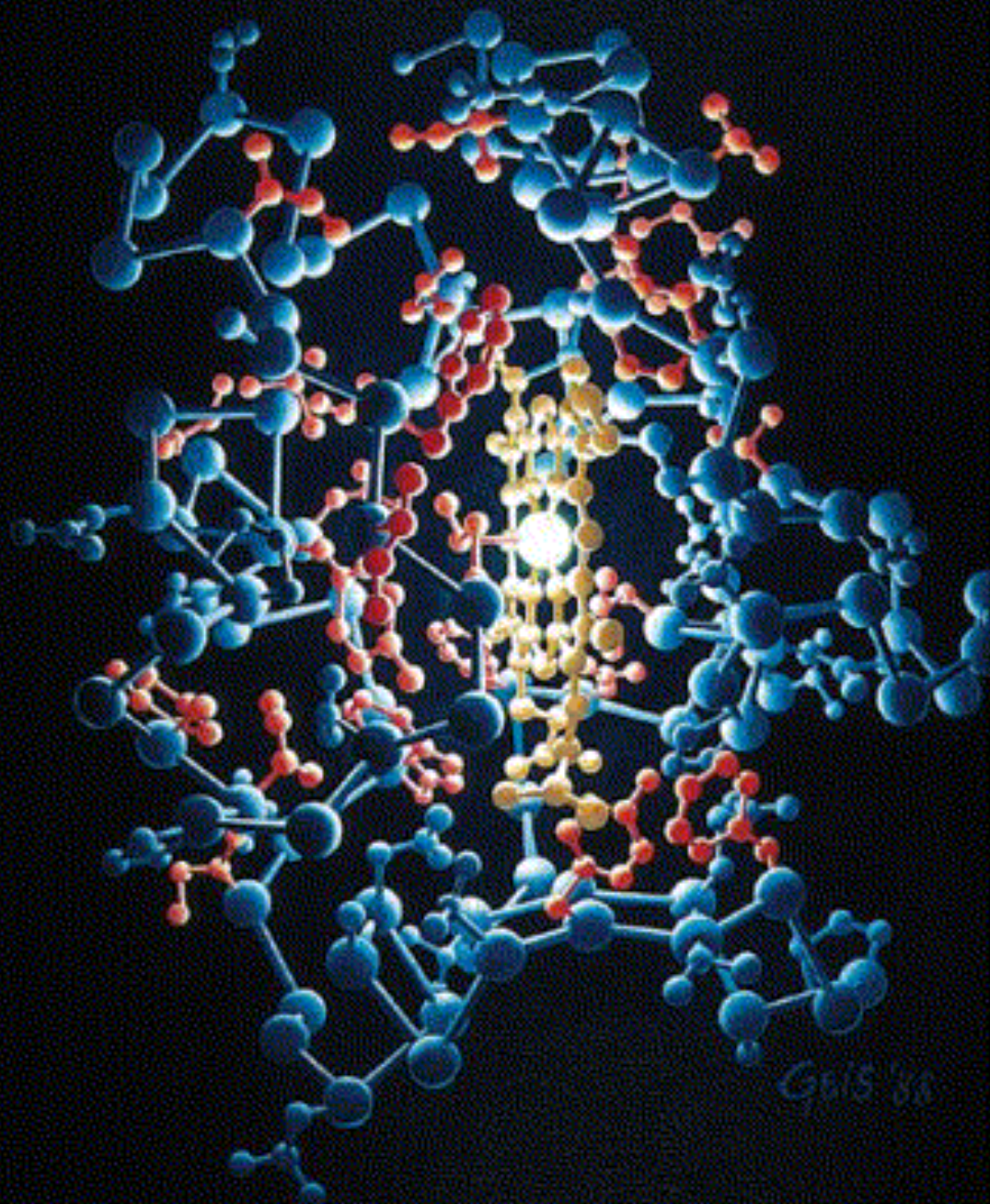
Symposium Celebrating the 40th Anniversary of the Protein Data Bank,
Cold Spring Harbor, NY, USA, October 29, 2011

Structural Biology by NMR and the Protein Data Bank

Kurt Wüthrich

The Scripps Research Institute, La Jolla, CA, USA and
ETH Zürich, Zürich, Switzerland



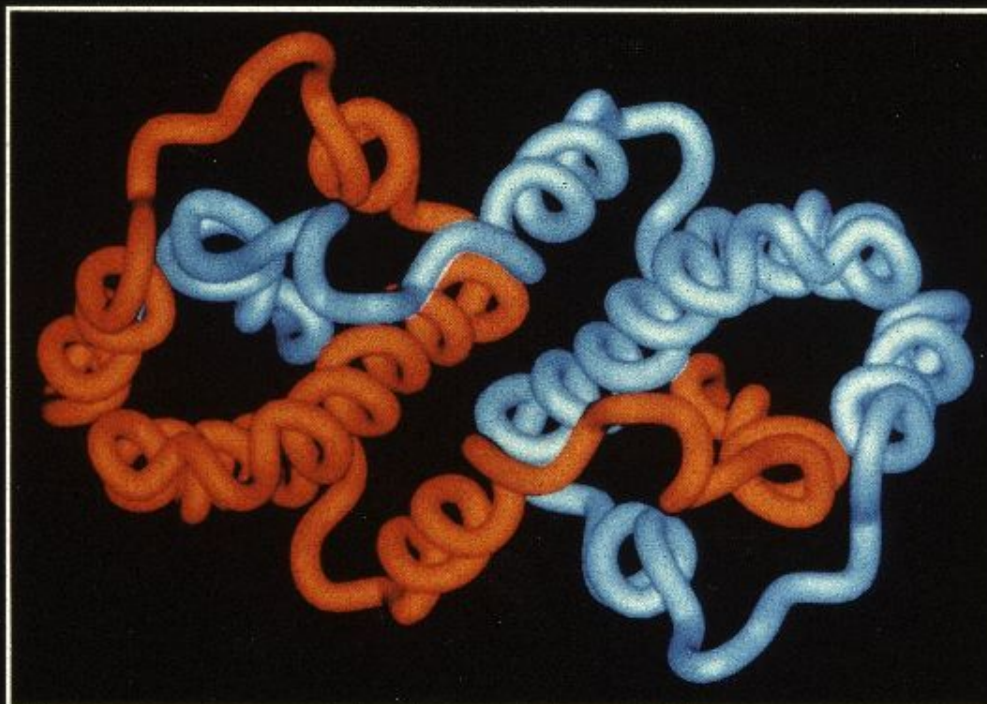


GUS '88

MACROMOLECULAR STRUCTURES

Atomic structures of
biological macromolecules
reported during 1991

1992

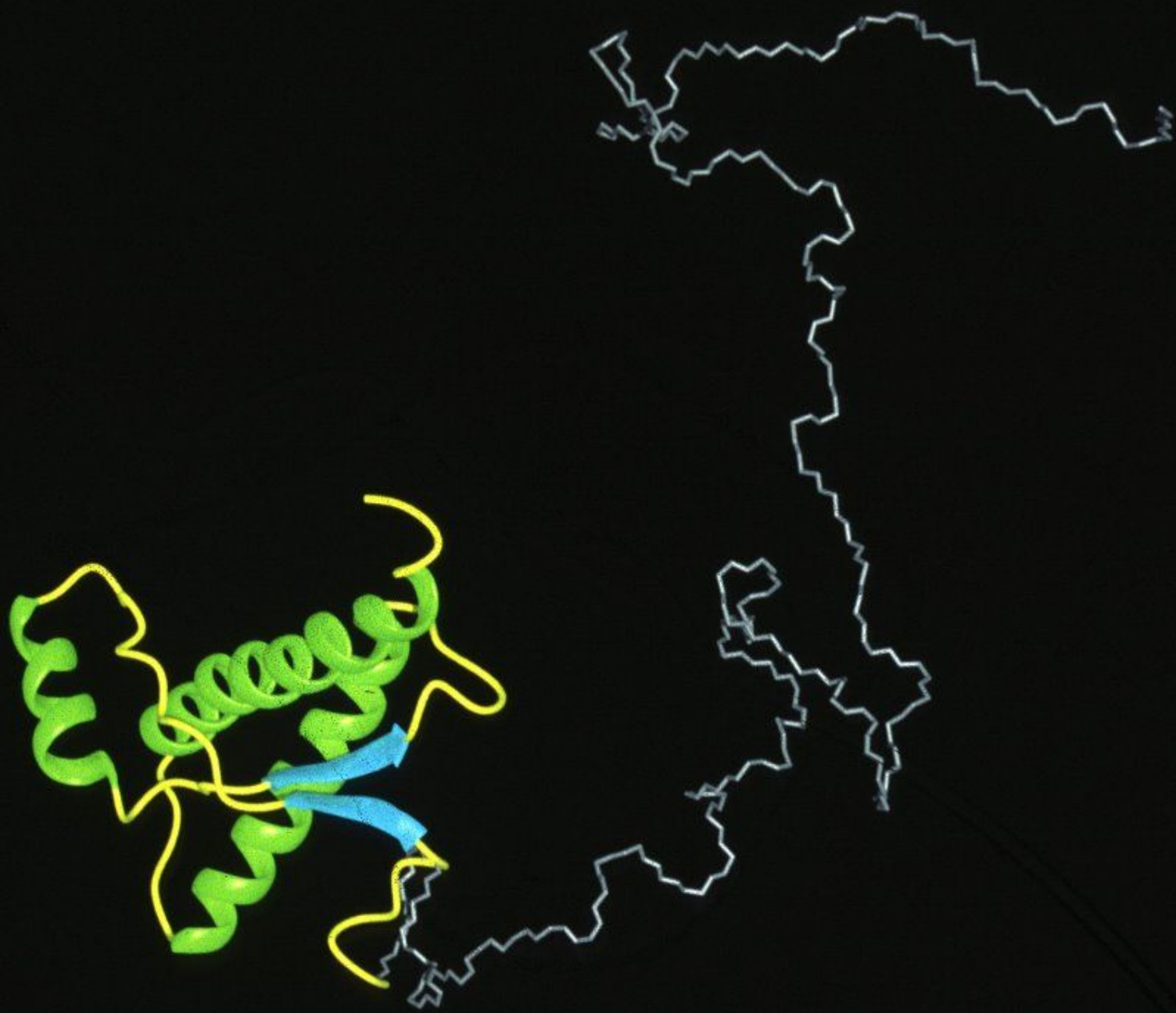


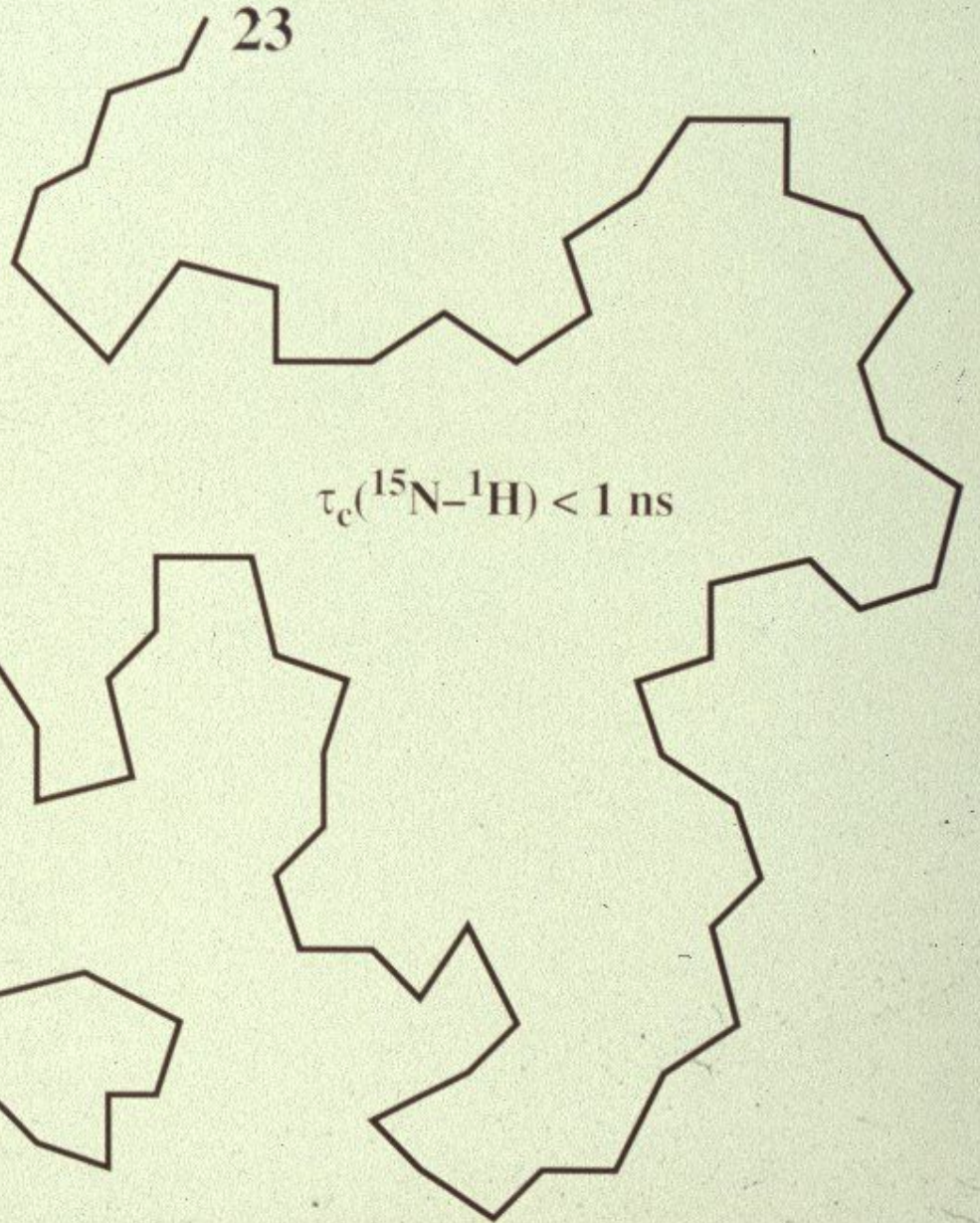
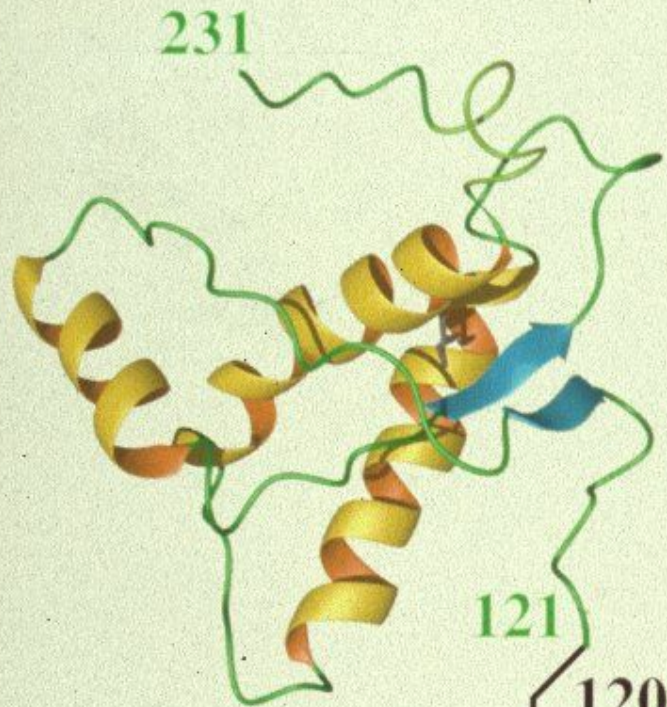
Edited by Wayne A Hendrickson and Kurt Wüthrich

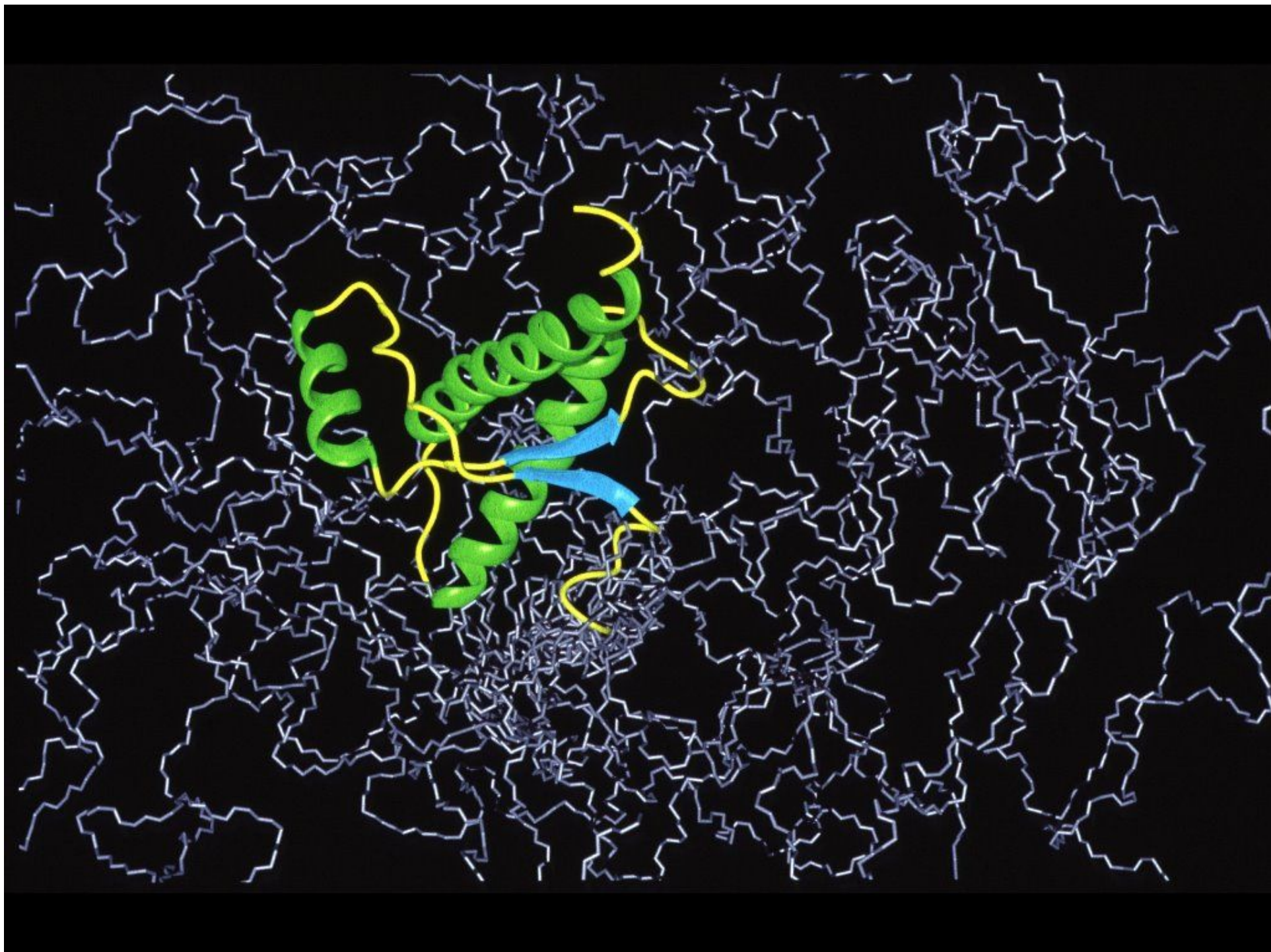
New 3D Structures of Proteins and Nucleic Acids

(Source: Macromolecular Structures 1991,...., 1999)

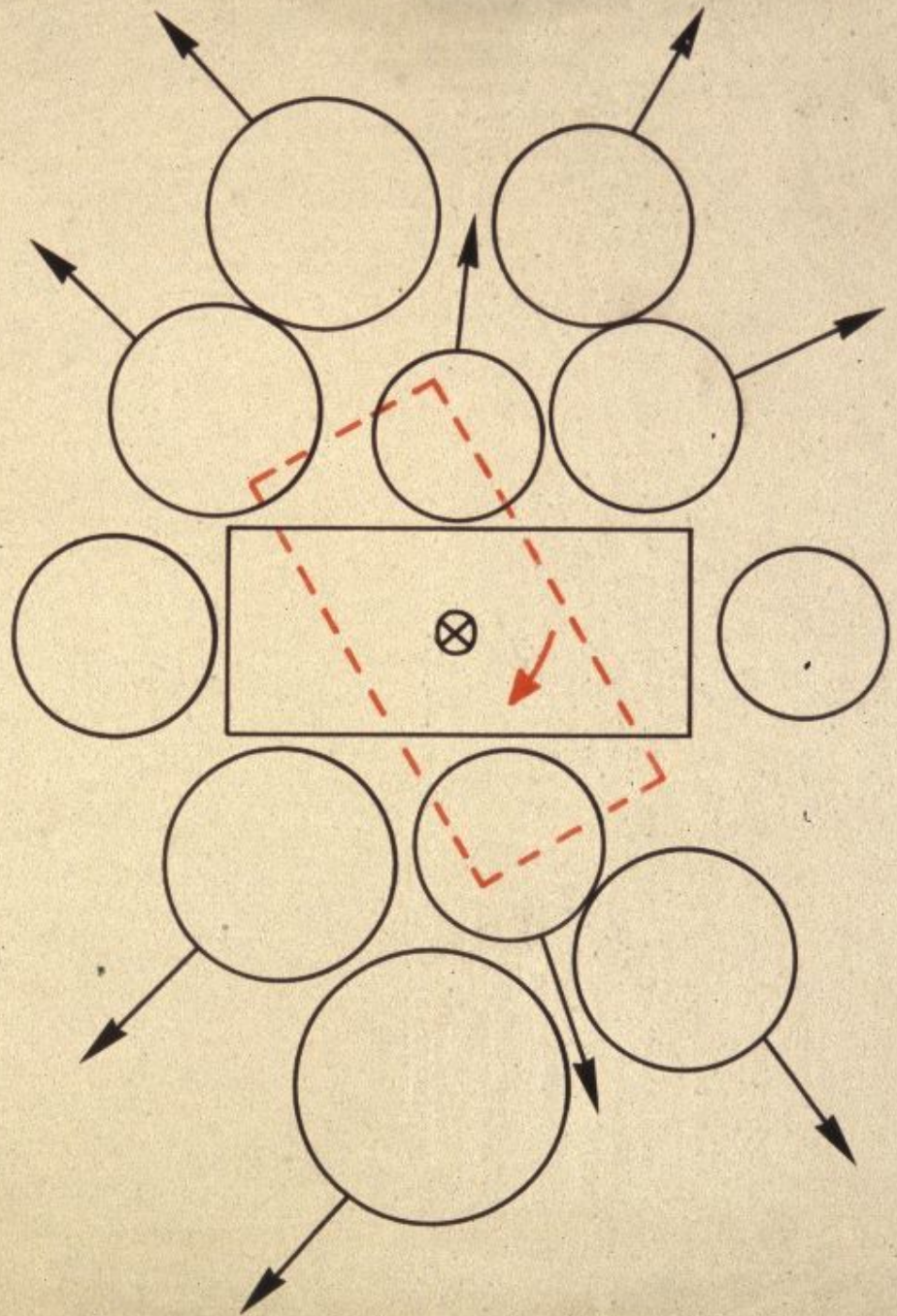
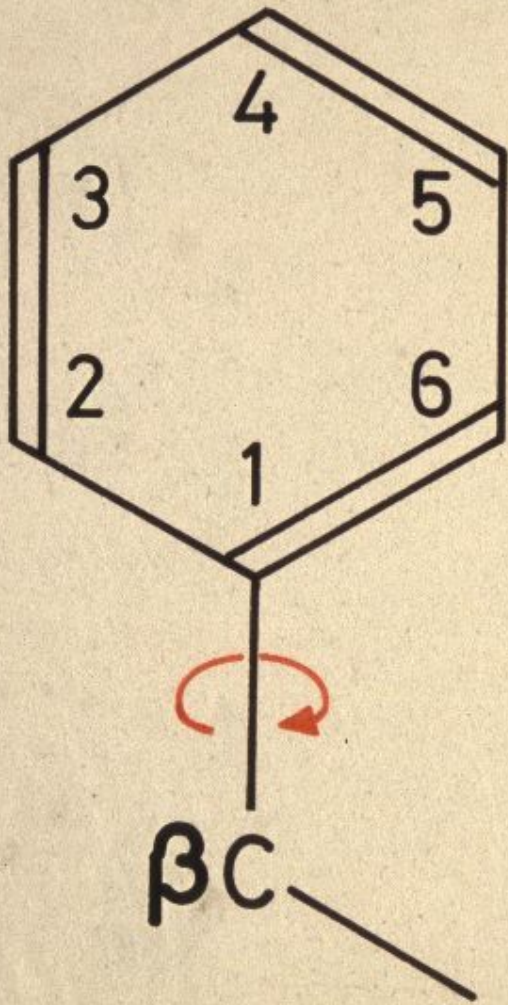
Year	X-rays (single crystals)	NMR (solution)	Other methods (crystals, fibers)
1990	109	23	2
1991	123	36	-
1992	168	61	-
1993	207	59	-
1994	377	100	2
1995	394	102	-
1996	461	112	-
1997	540	99	-
1998	747	125	2

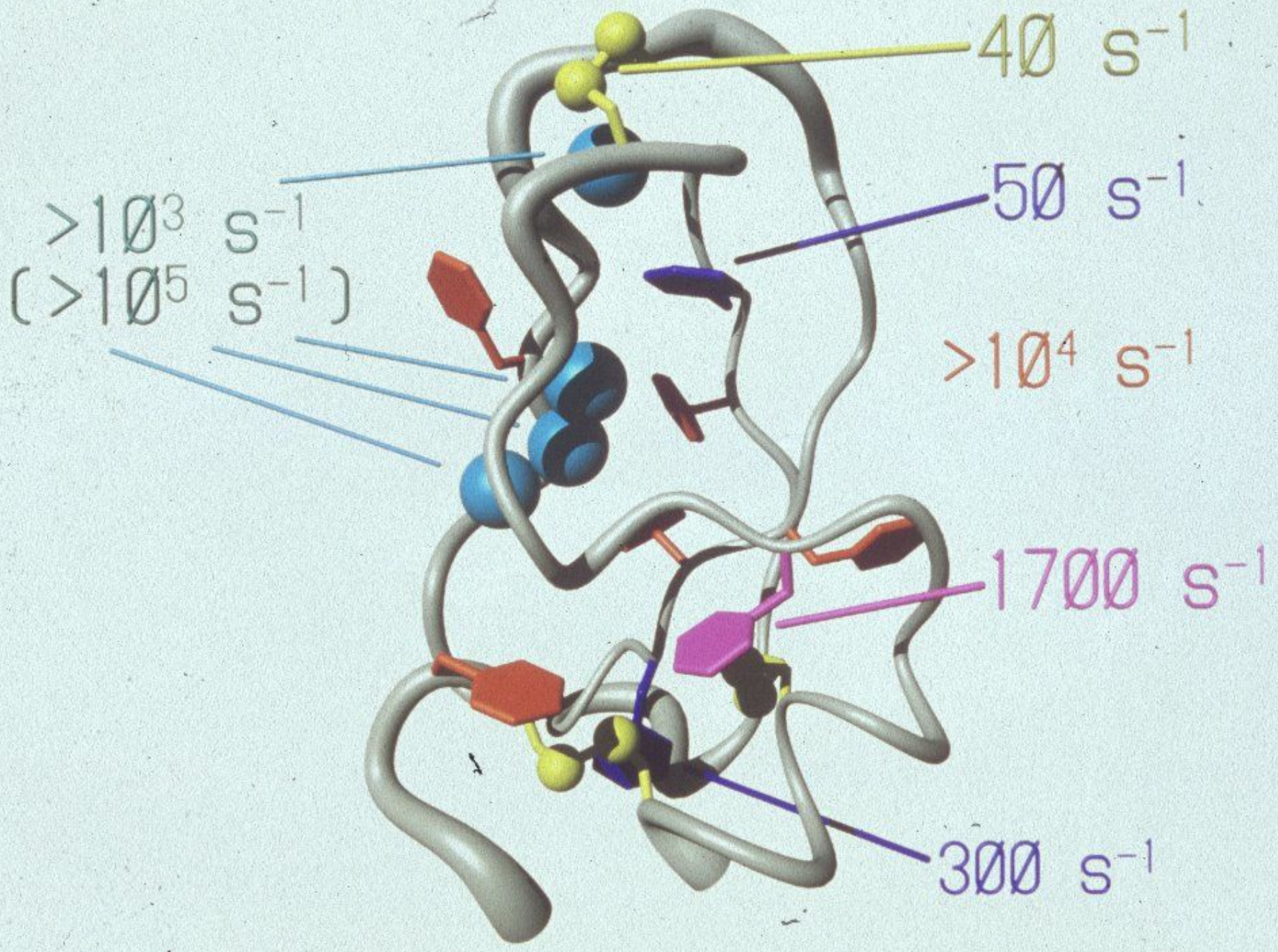






RING FLIPS OF PHE AND TYR





JCSG

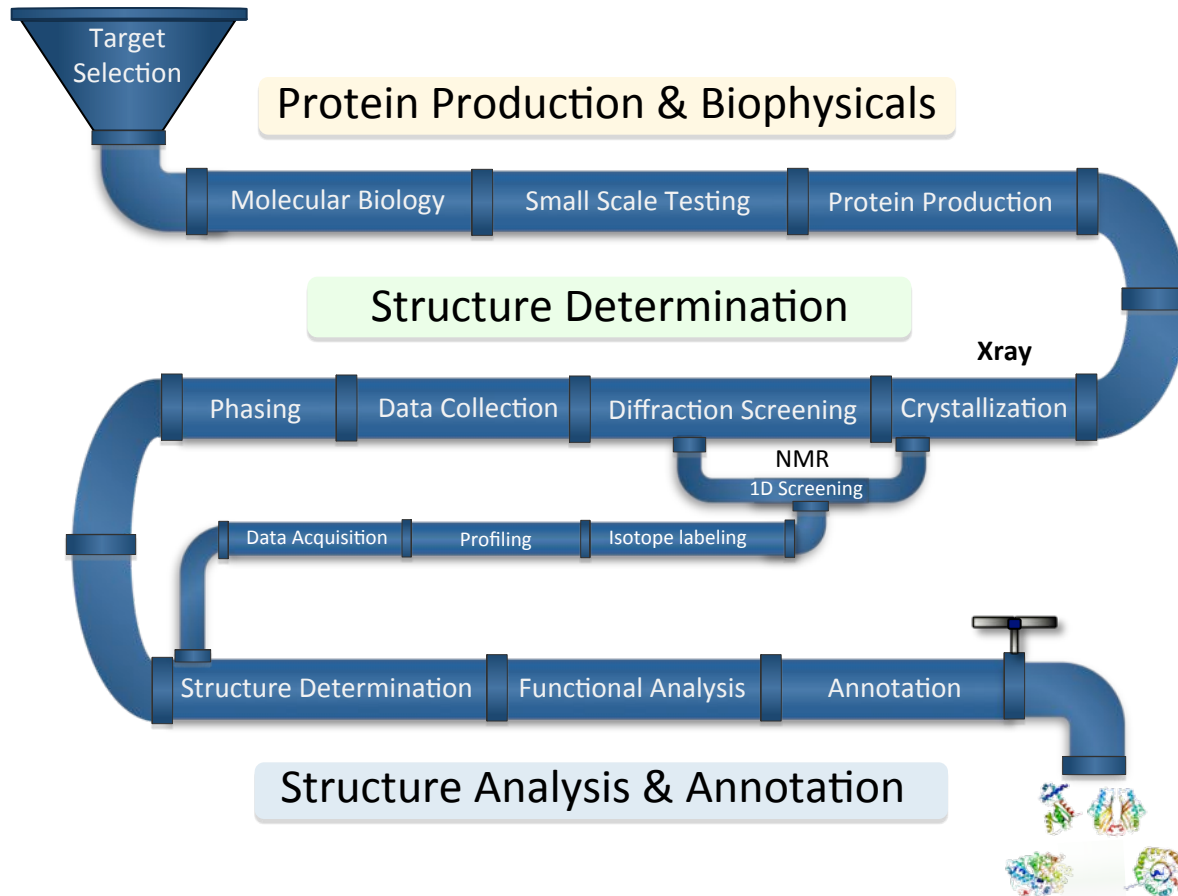


Joint Center for Structural Genomics

Developing HT methods for Gene to Structure and Function

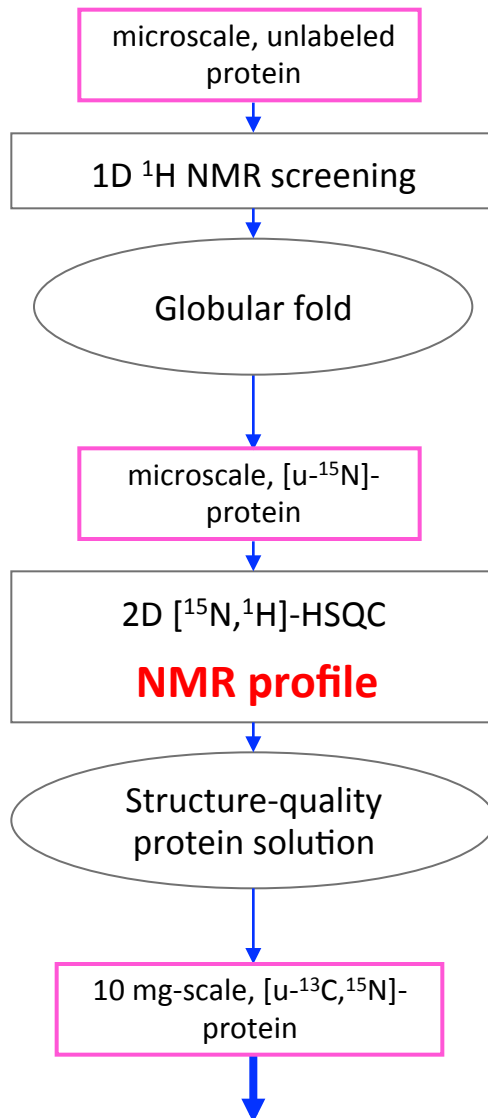


HT Structure Determination Pipeline

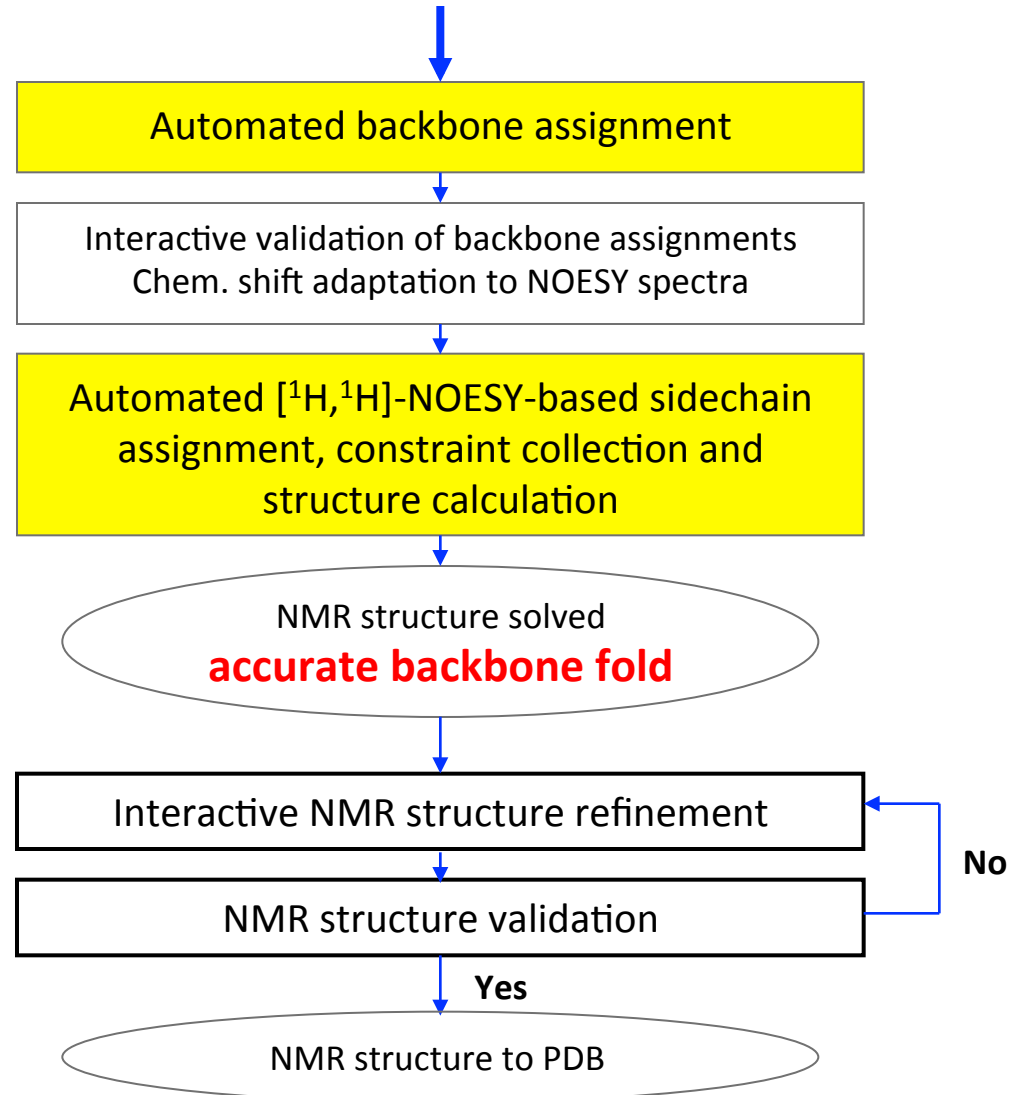


Protocol for automated NMR structure determination

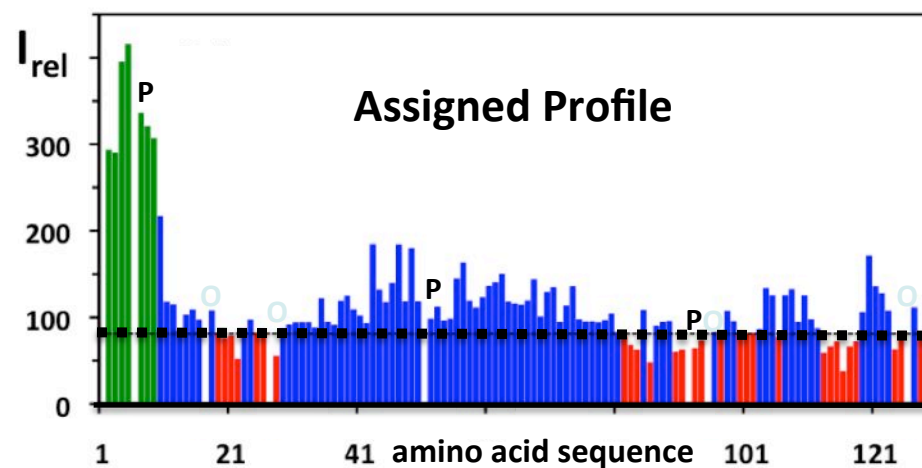
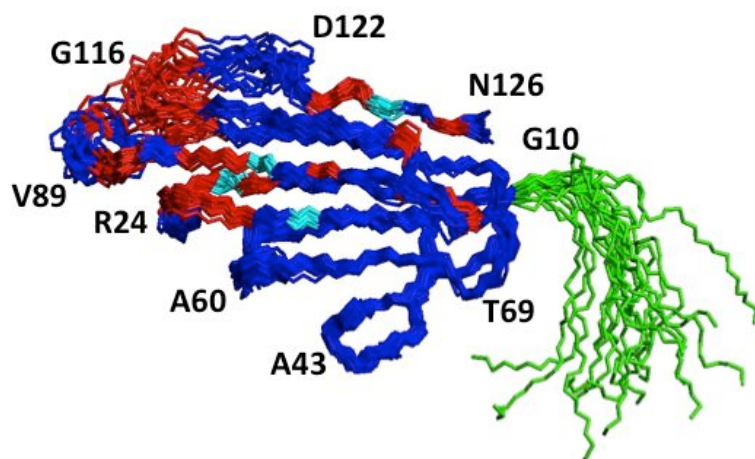
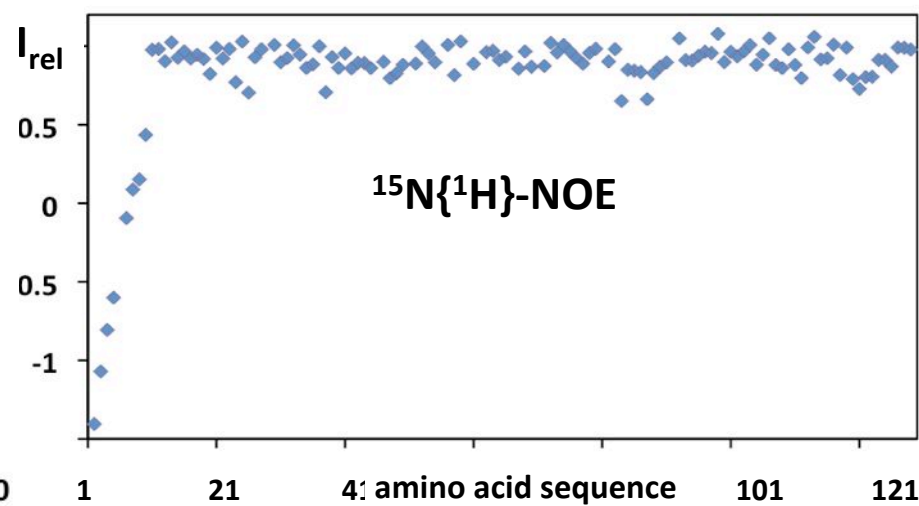
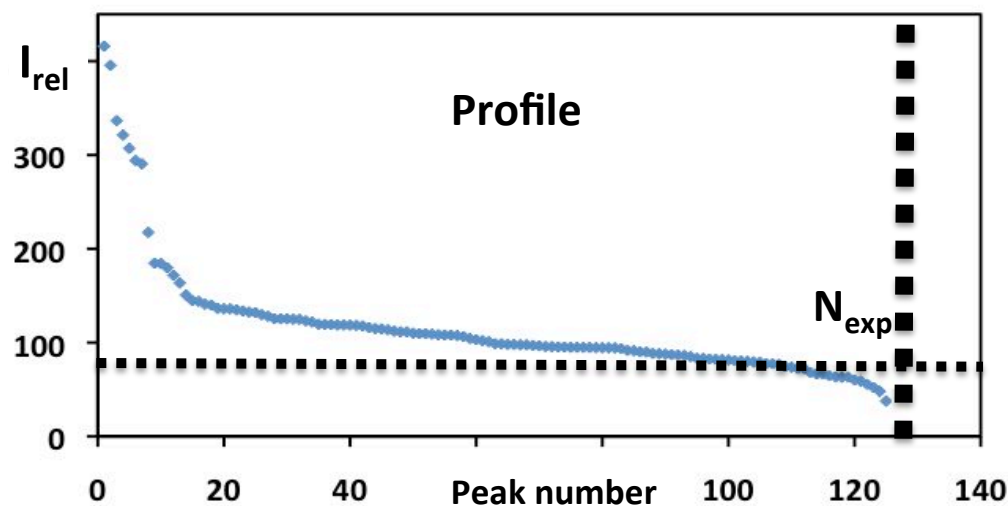
1. NMR sample preparation



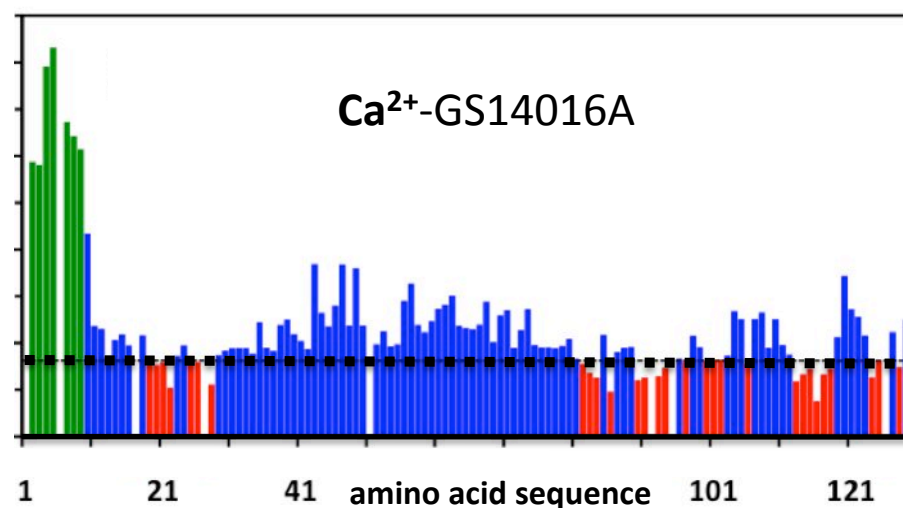
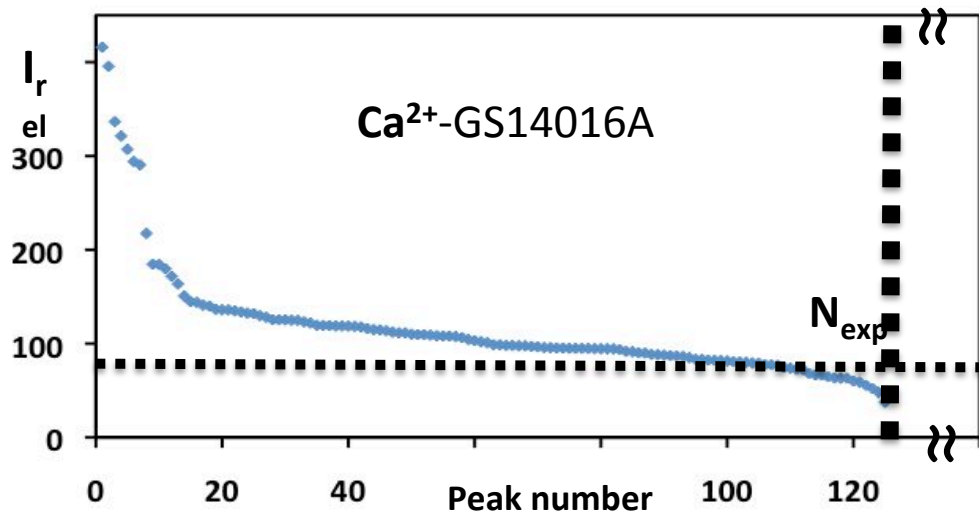
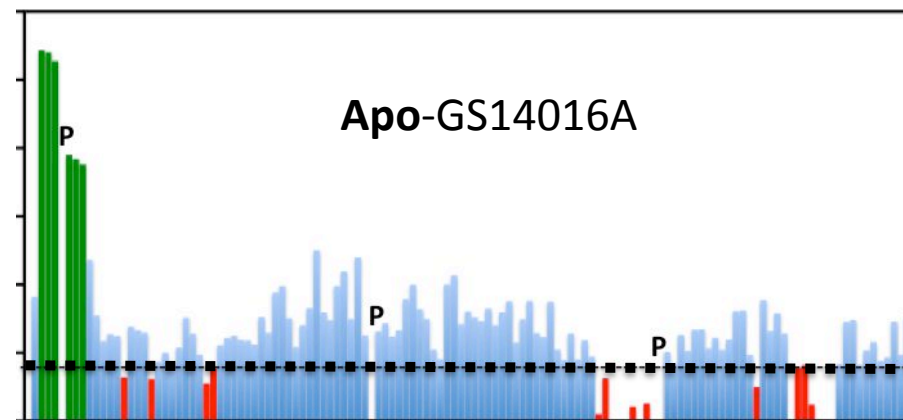
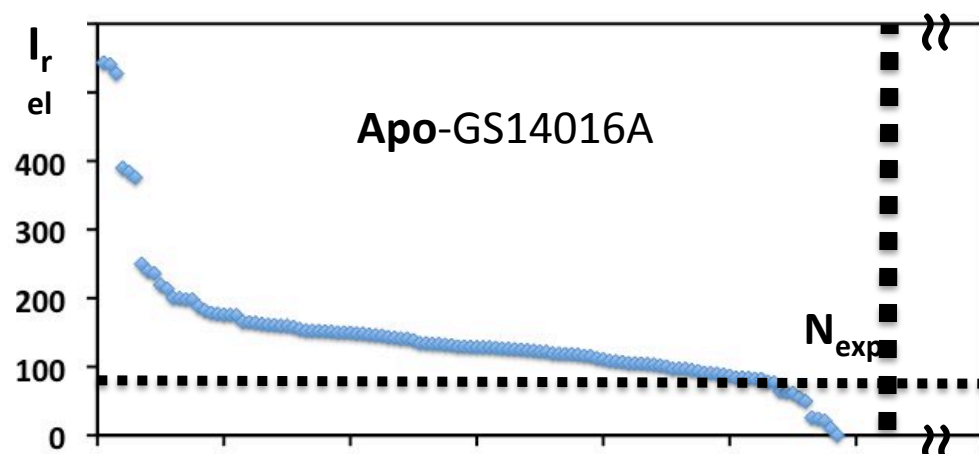
2. NMR structure determination



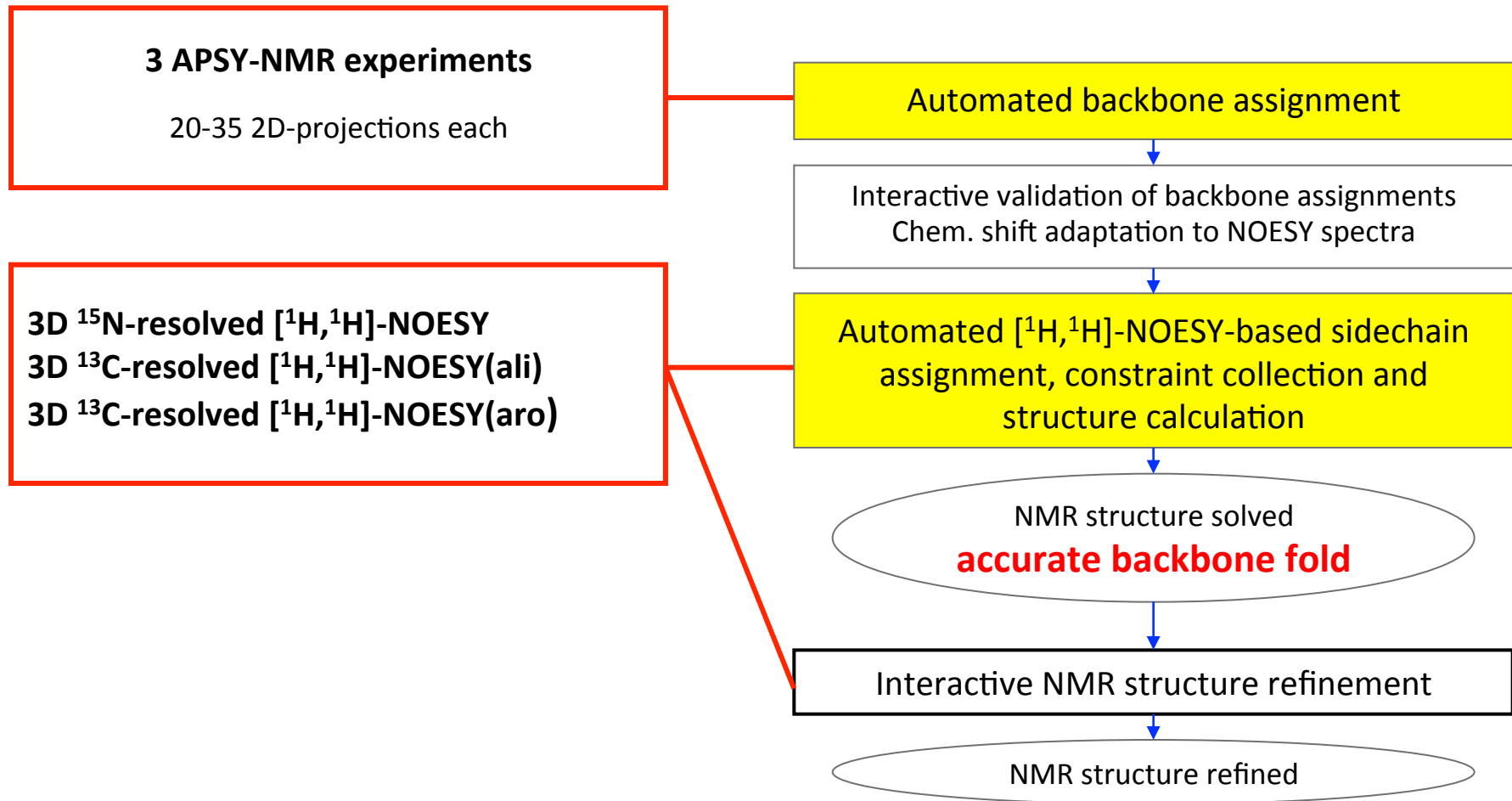
[¹⁵N,¹H]-NMR-Profiles and Protein Structure



[¹⁵N,¹H]-NMR-Profiles and Protein Complexation

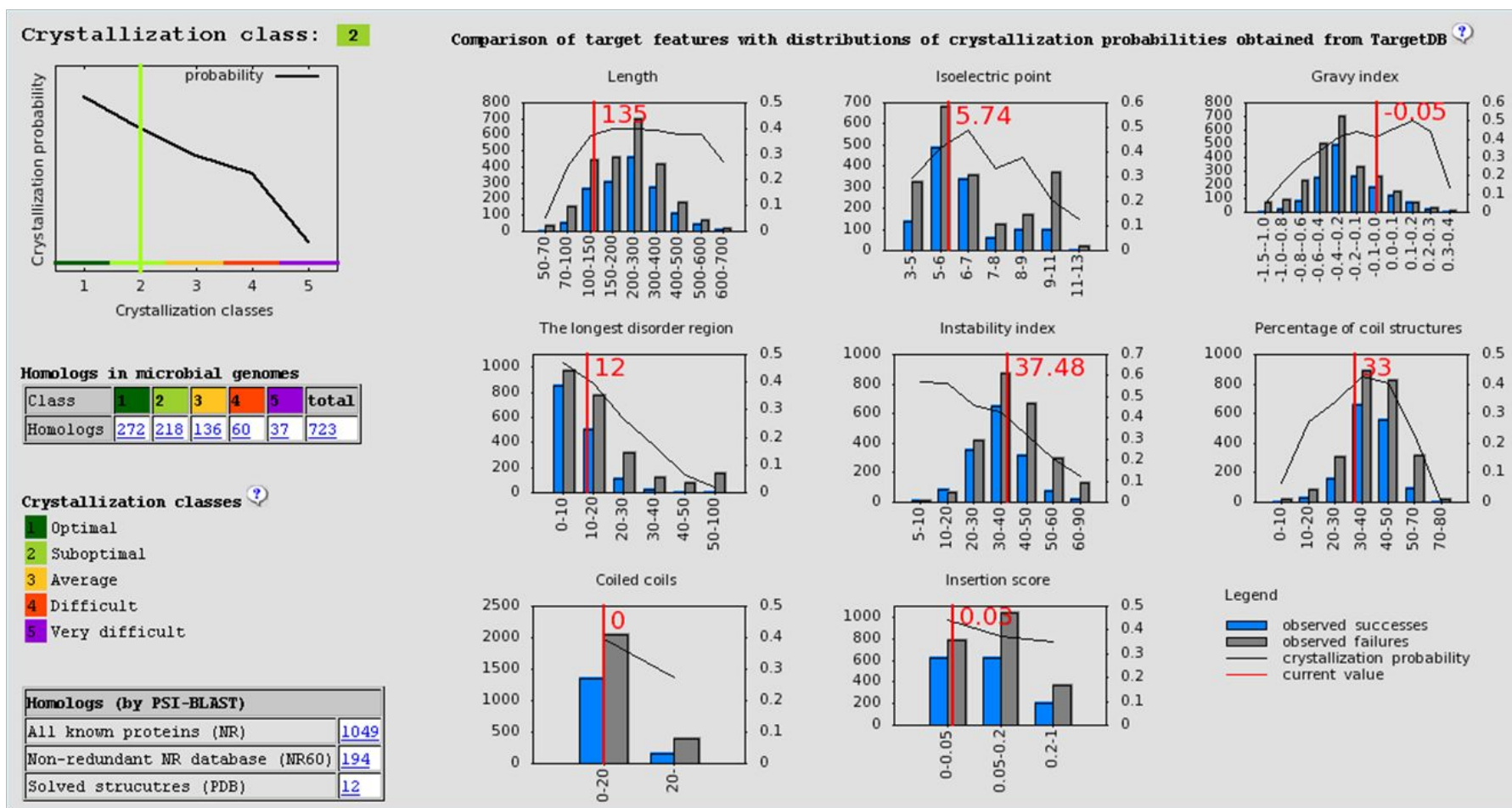


NMR experiments

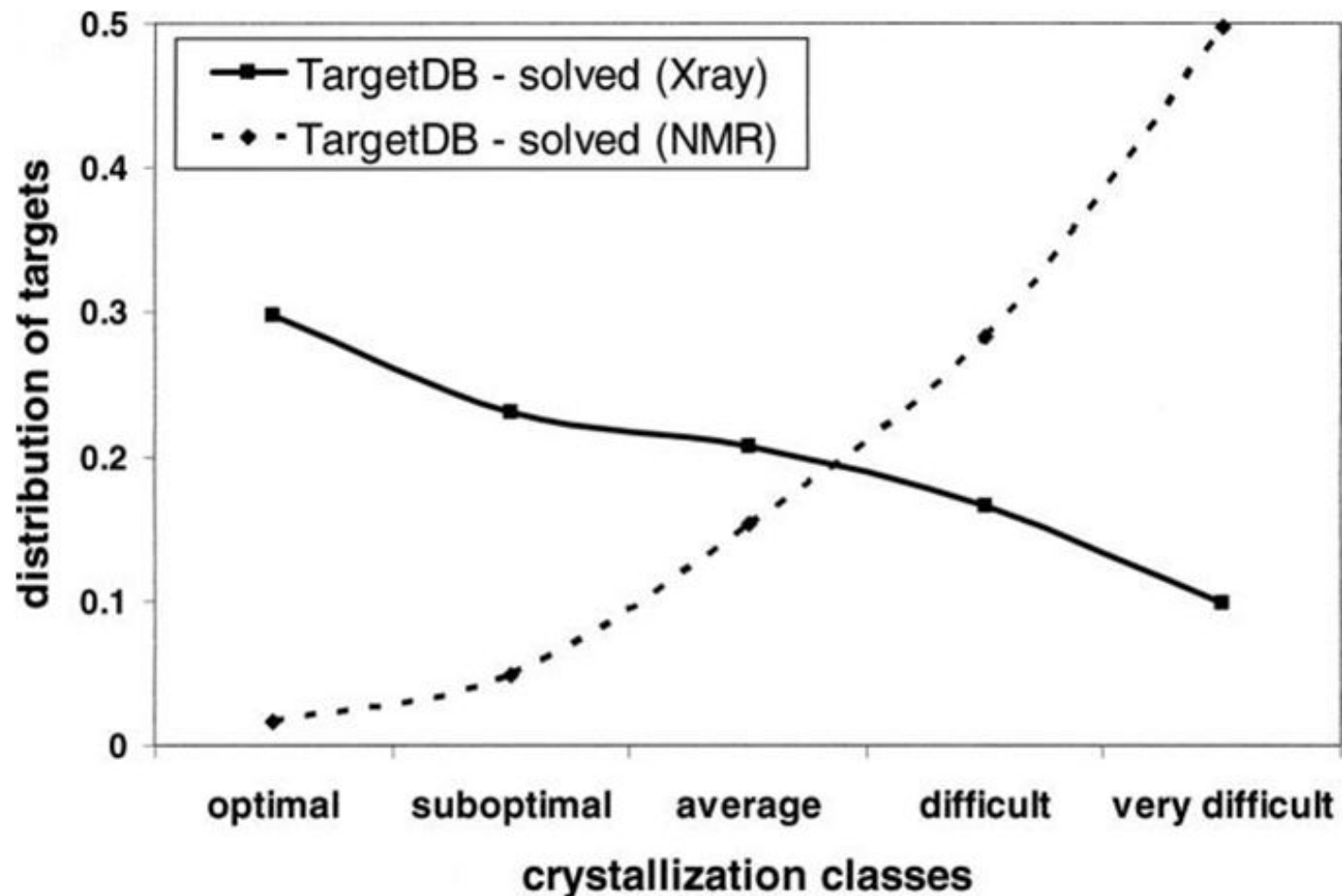


XtalPred: a web server for prediction of protein crystallizability

(<http://ffas.burnham.org/XtalPred-cgi/xtal.pl>)

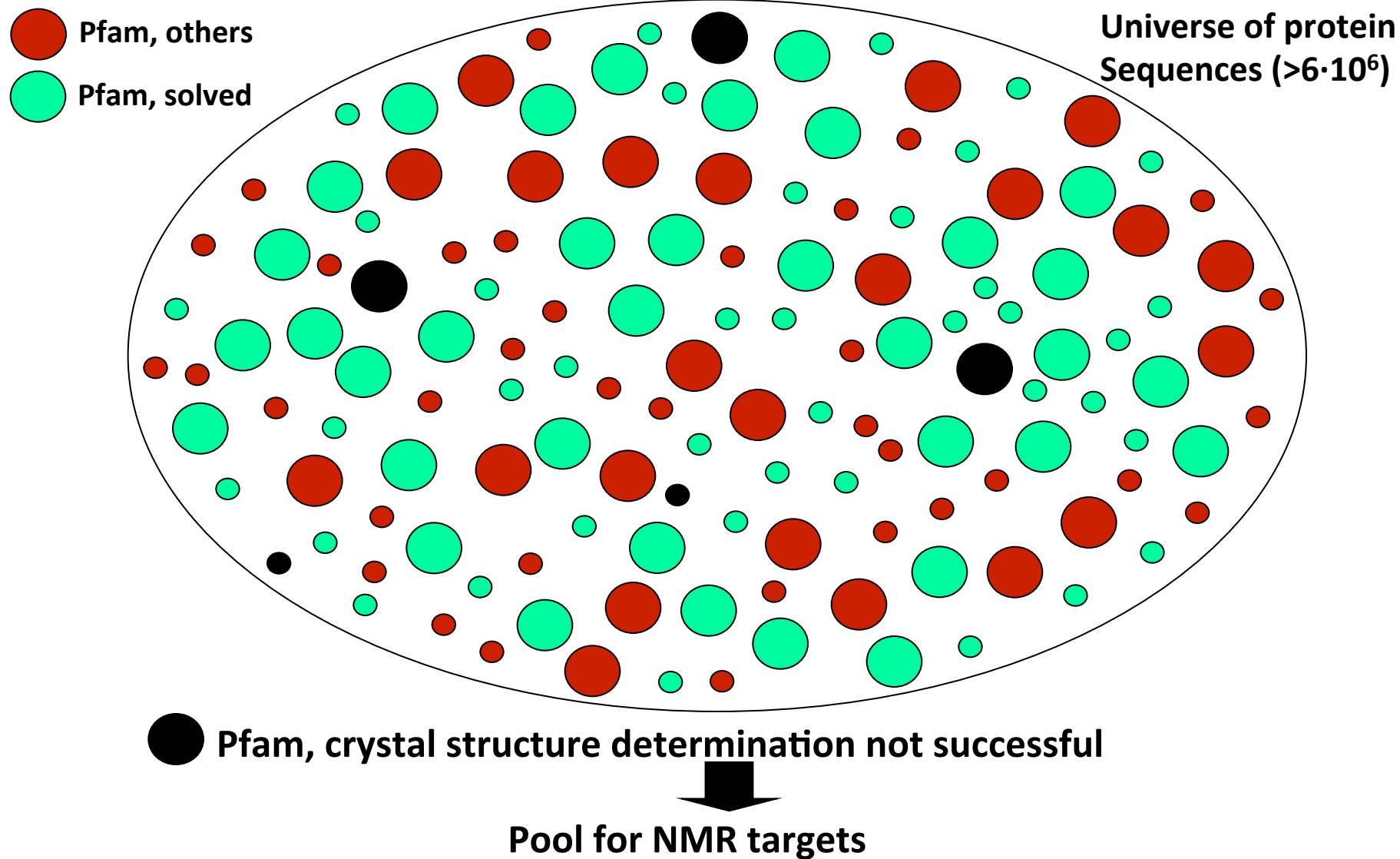


Xray and NMR highly complementary

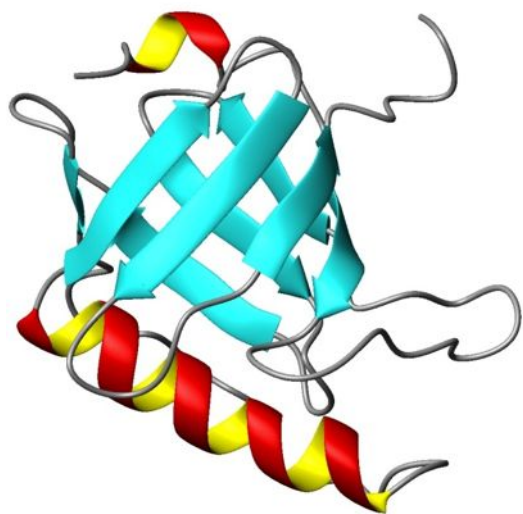


Slabinski L, Jaroszewski L, Rodrigues AP, Rychlewski L, Wilson IA, Lesley SA, Godzik A. "The challenge of protein structure determination- lessons from structural genomics," *Protein Science*, 16: 2472-2482 (2007).

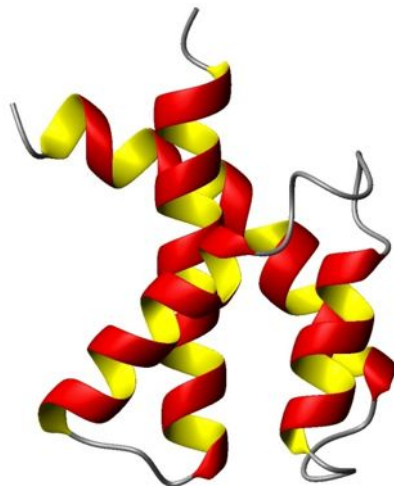
JCSG NMR Target Selection on the HT-Crystallography Background



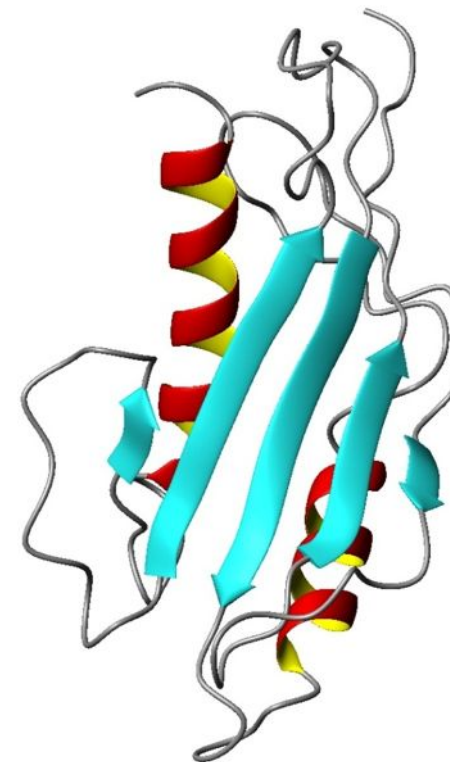
Structural Coverage of New Pfams by NMR



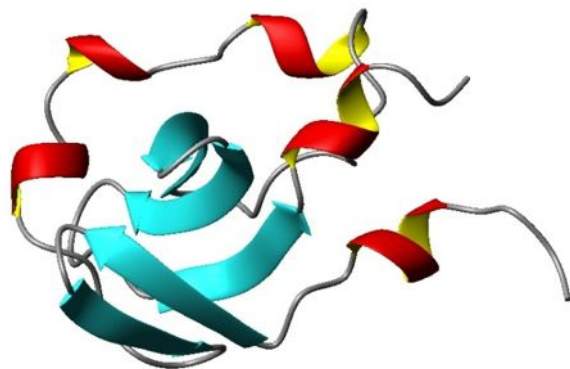
2KTS / PF03724



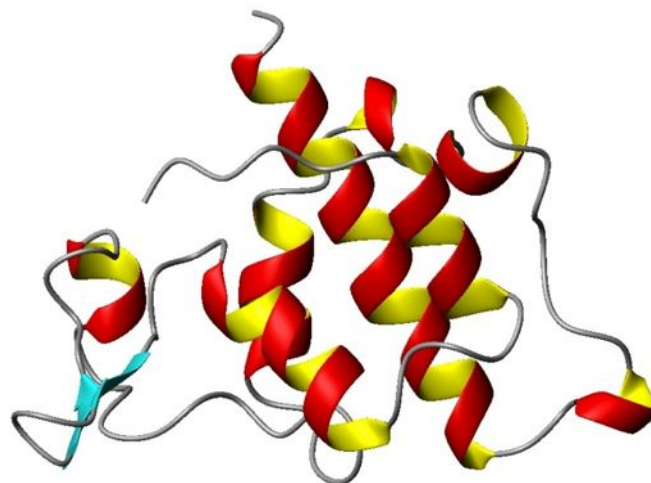
2KZC / PF07345



2L25 / BIG_19



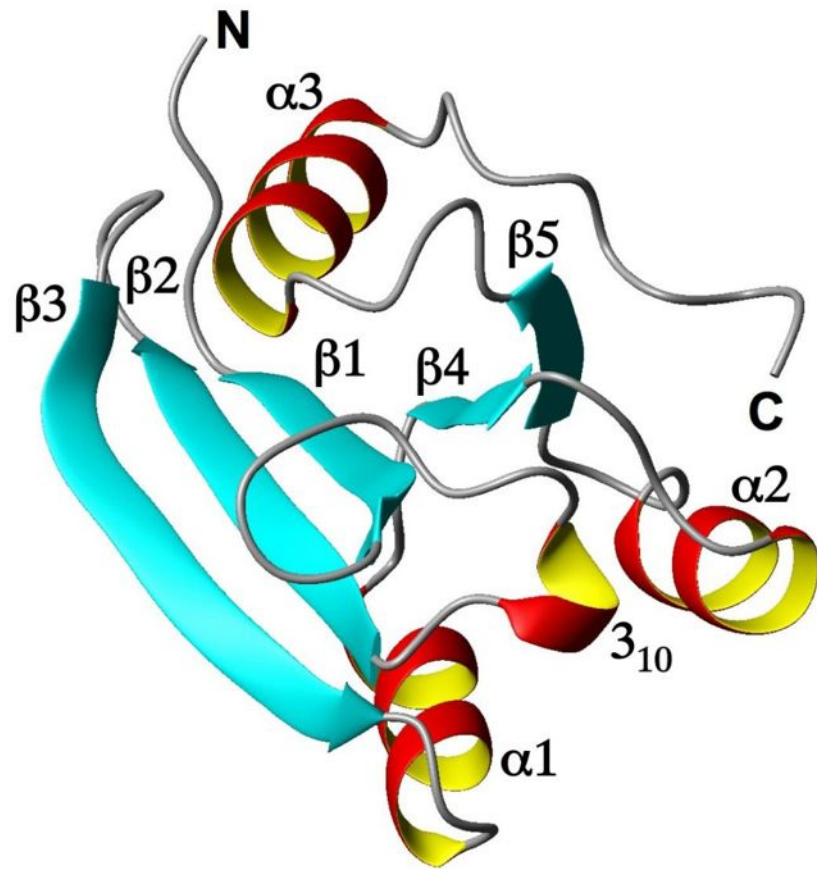
2L1S / DUF3315



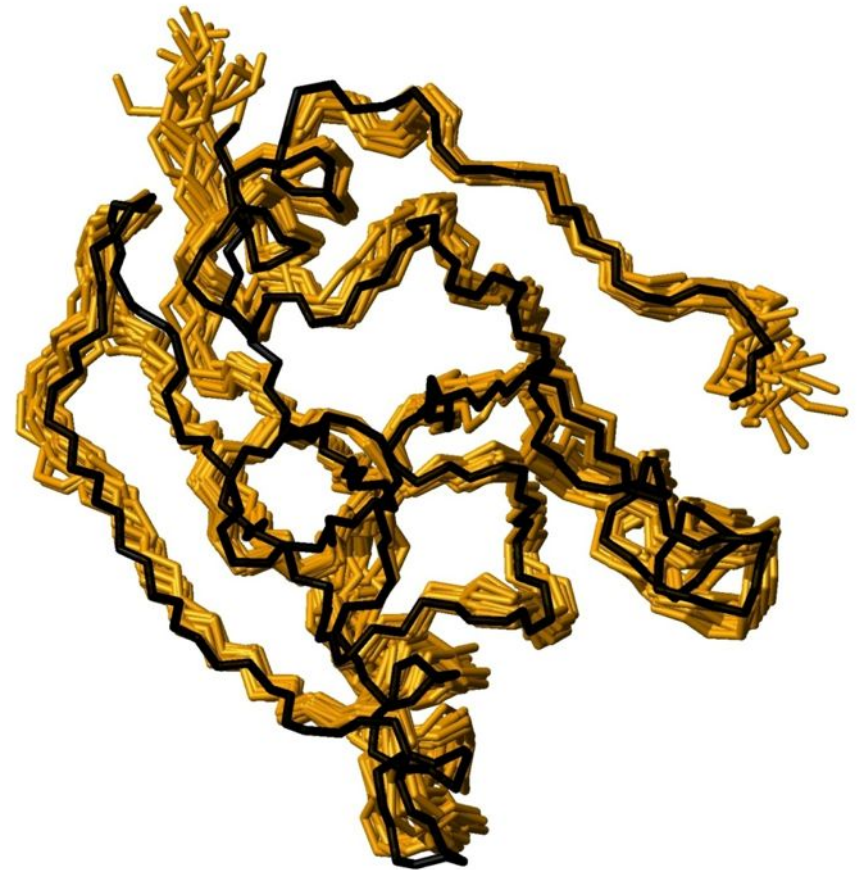
2L1N / DUF1823

NP_247299.1 (*Methanococcus jannaschii*)

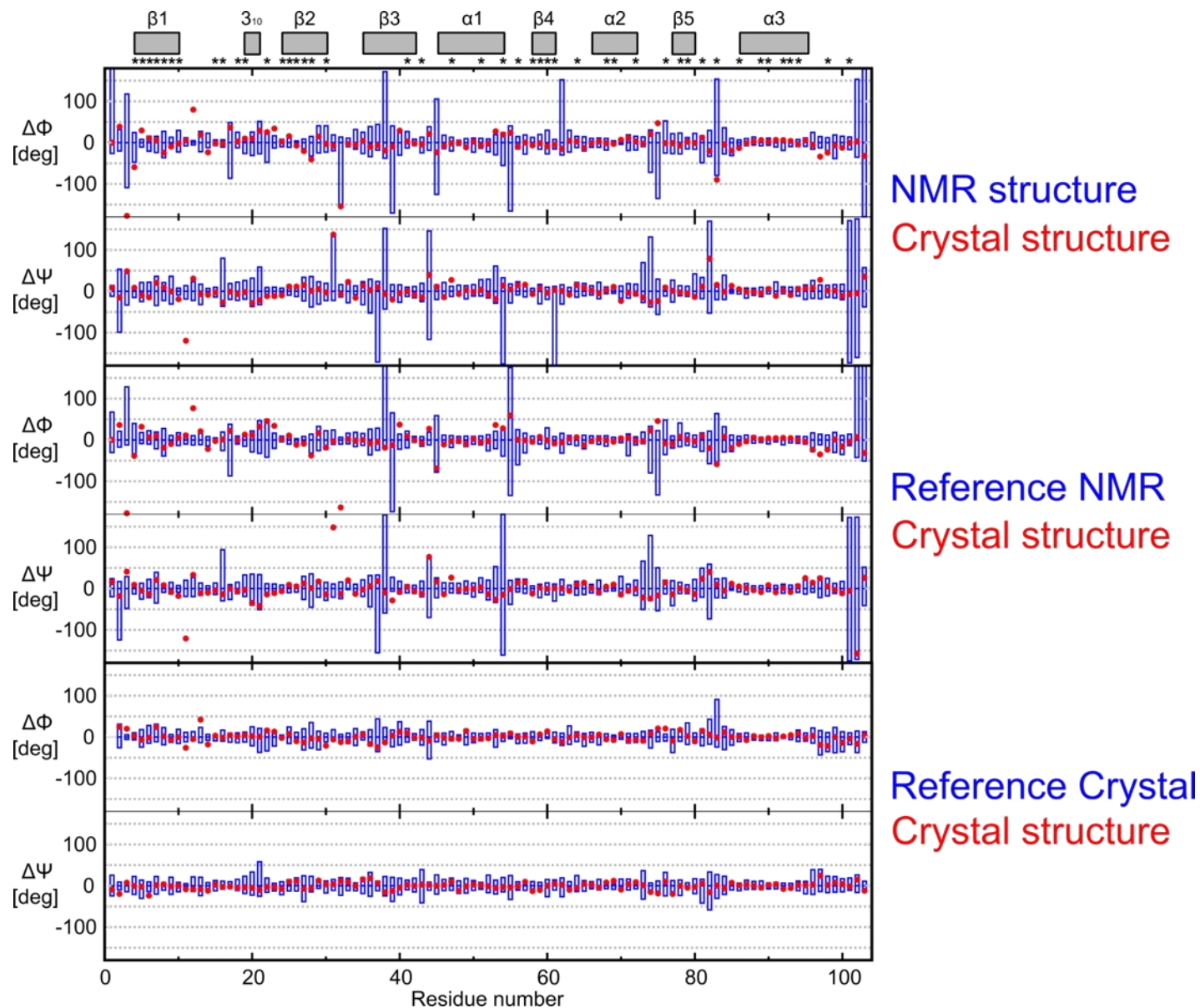
NMR



NMR/Crystal

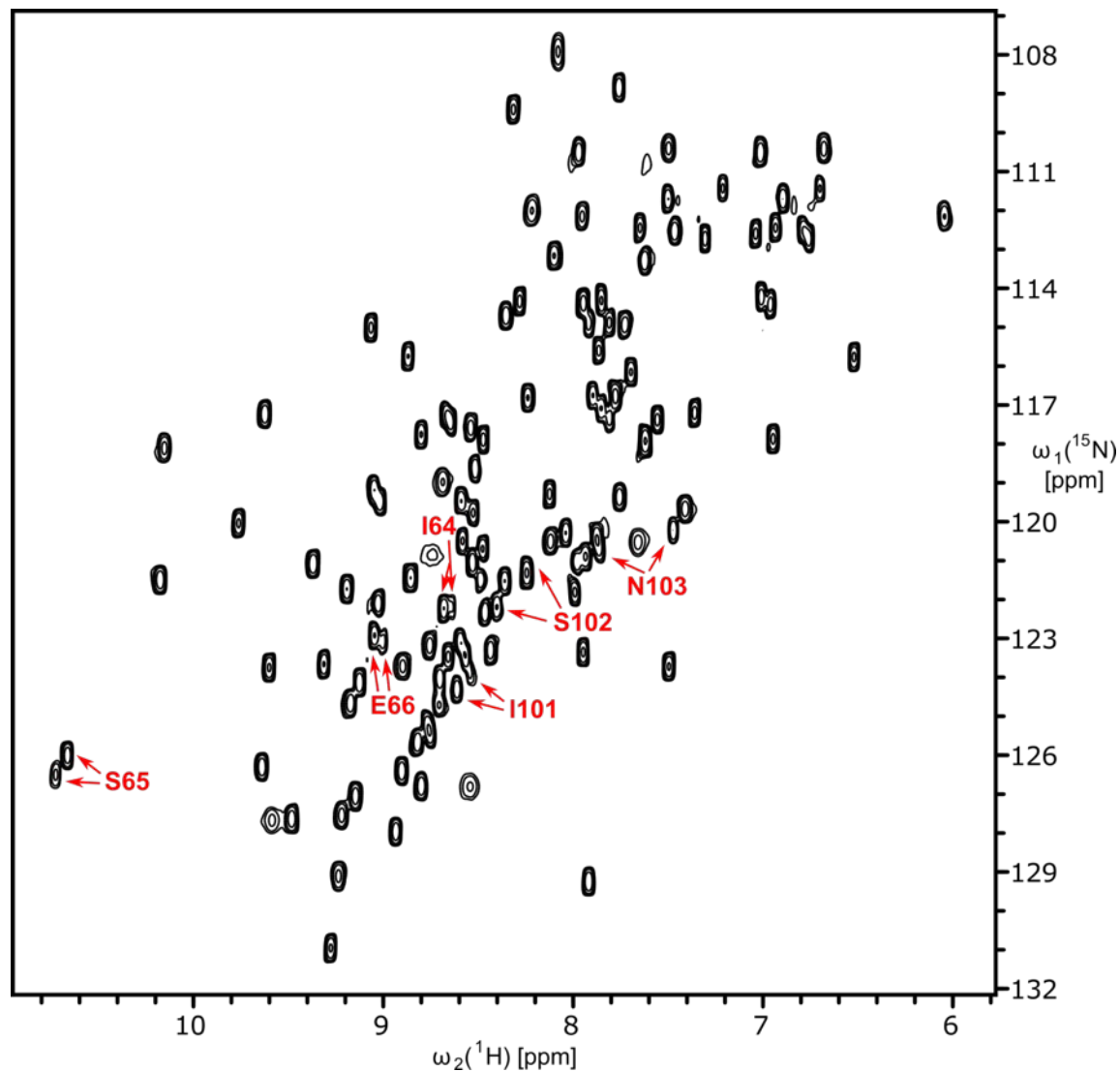


Backbone Dihedral Angles Φ and Ψ



NP_247299.1: *cis/trans* isomers of N103–P104

NMR chemical shifts: two conformations for 64–66 and 101–104

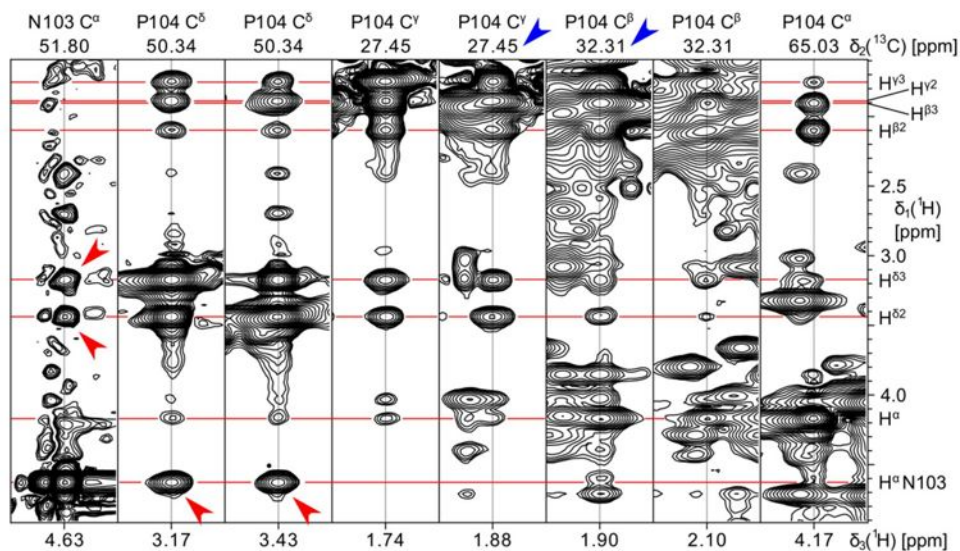


NP_247299.1: *cis/trans* isomers of N103–P104

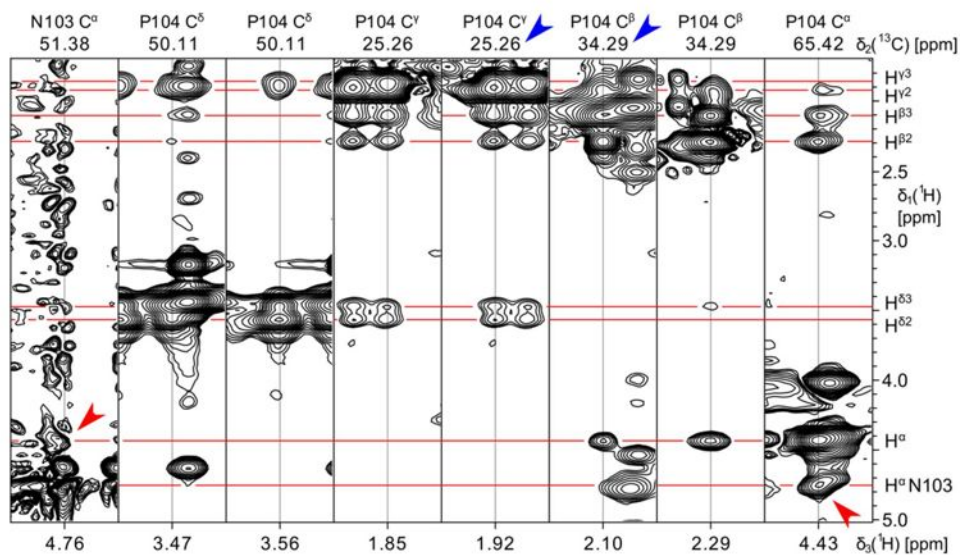
Identification from ^{13}C chemical shifts (▶) and ^1H – ^1H NOEs (▶)

3D ^{13}C -resolved
[^1H , ^1H]-NOESY

trans - Pro



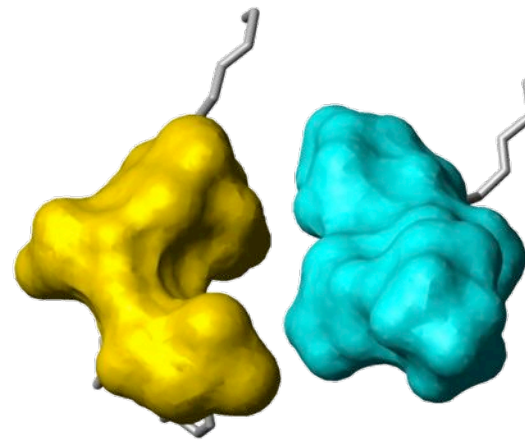
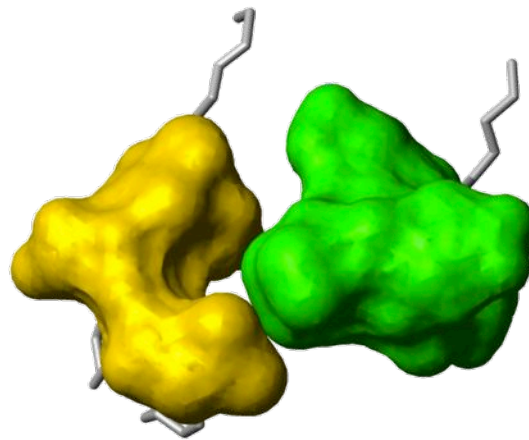
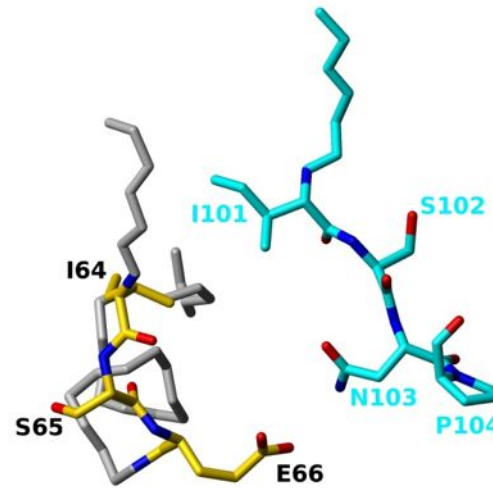
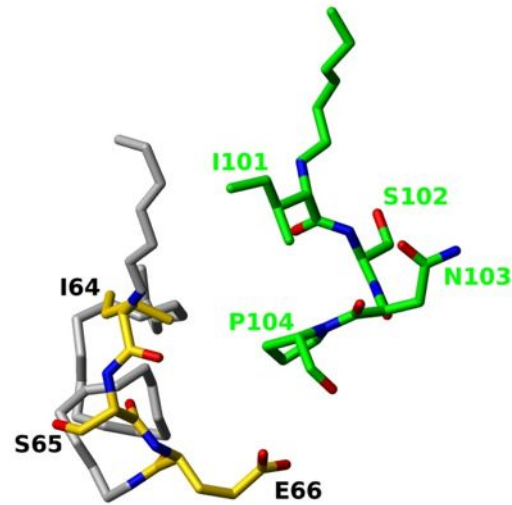
cis - Pro



NP_247299.1: segments 64–66 and 101–104

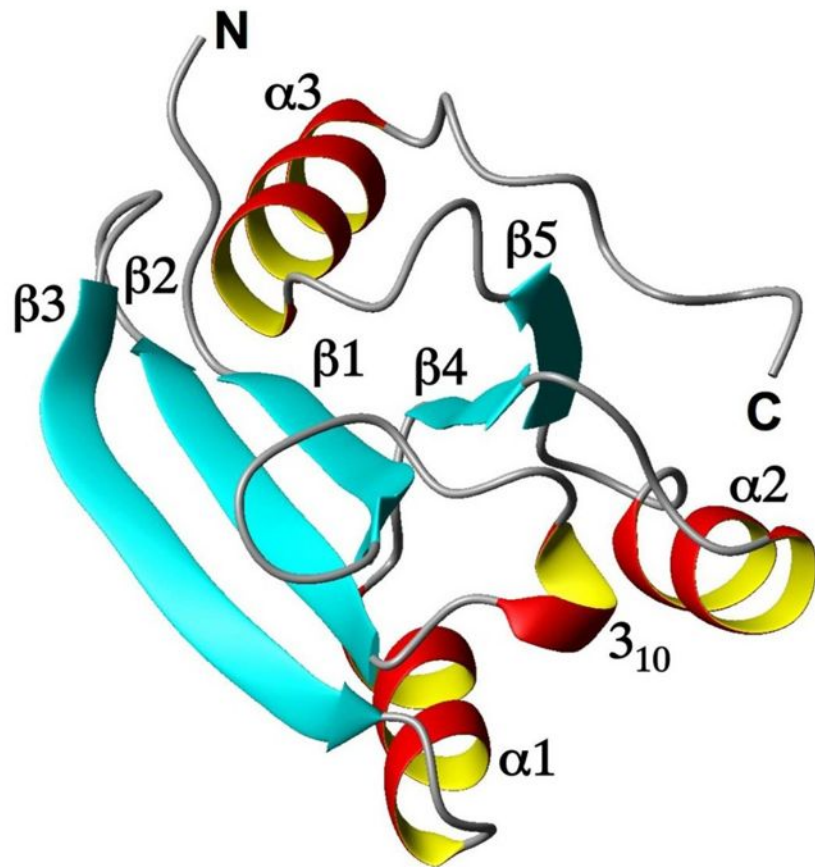
trans-P104

cis-P104

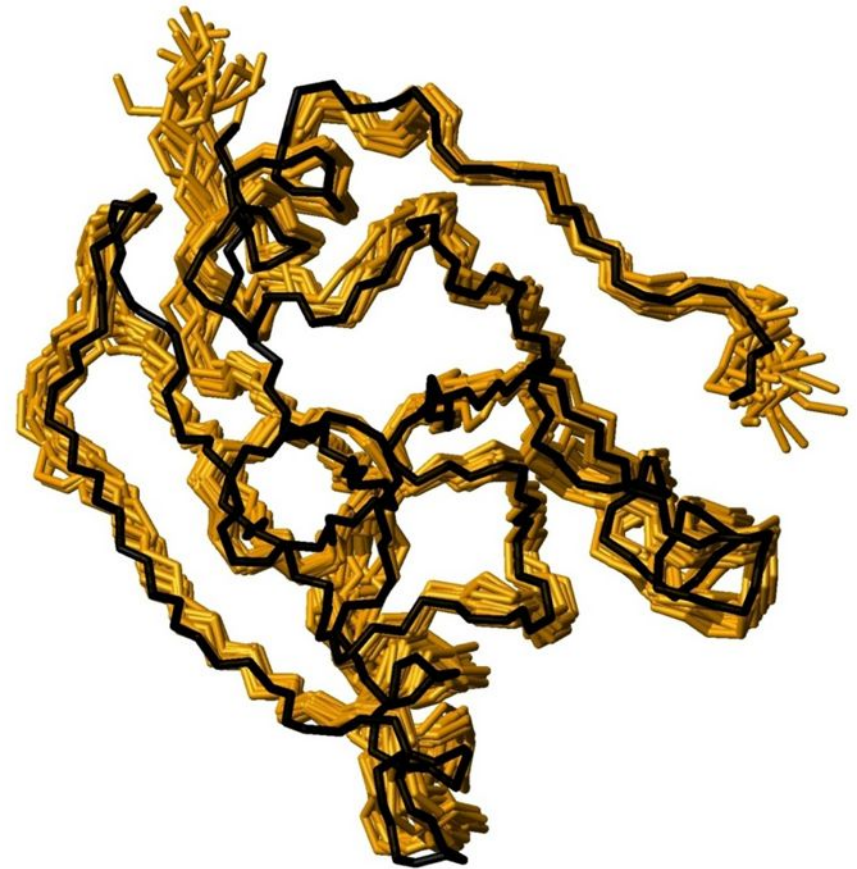


NP_247299.1 (*Methanococcus jannaschii*)

NMR



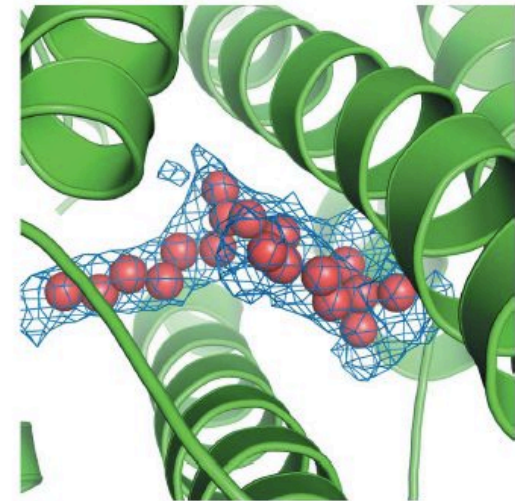
NMR/Crystal



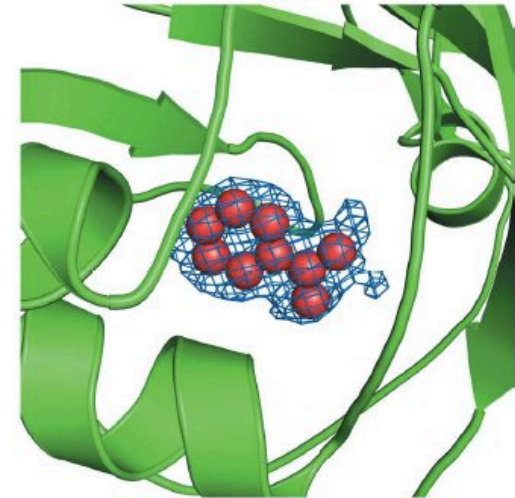
Bound Ligands Aid in Functional Annotation of Proteins

- Much diversity of ligands is found in protein crystal structures of the JCSG and other PSI centers.
- Interesting role for NMR in solution:
 - The identity of the bound ligand is often difficult to confirm by X-ray crystallography alone.
 - Scarcity of Apo-protein crystal structures.

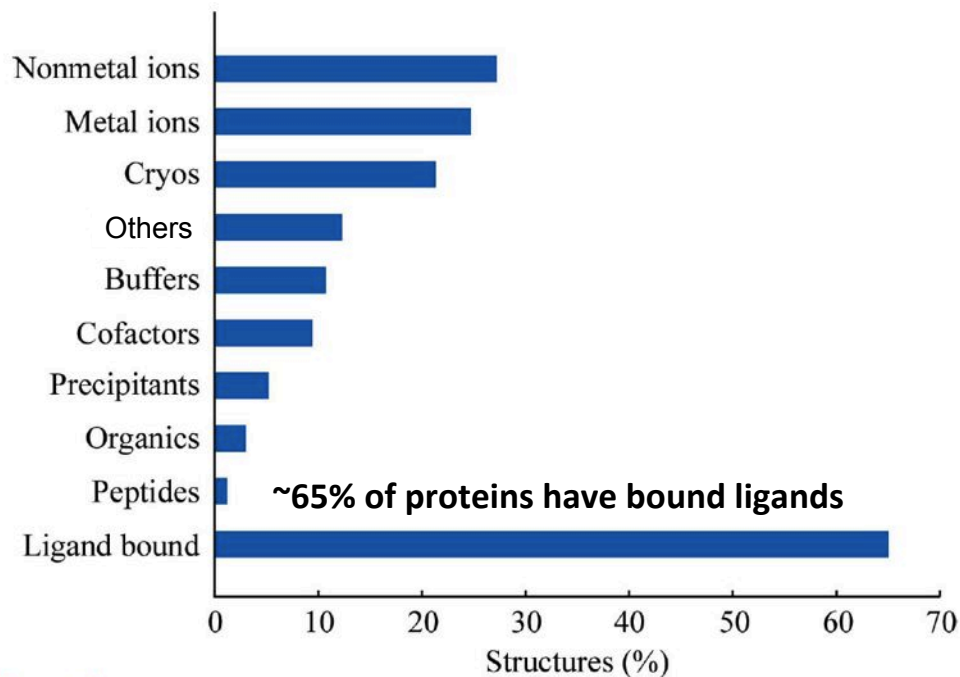
Examples of unknown ligands bound to JCSG structures



(b)

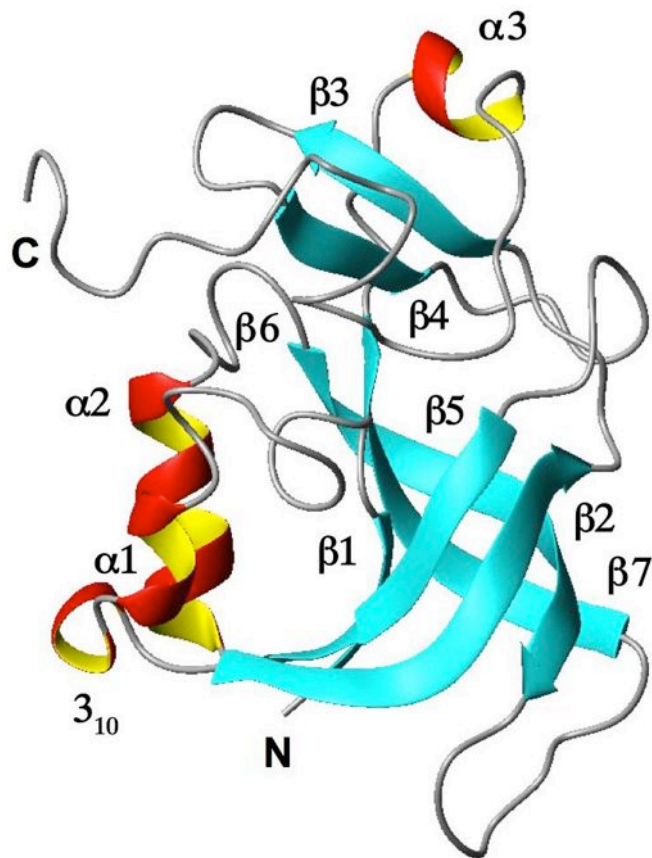


Types of bound ligands identified in PSI structures

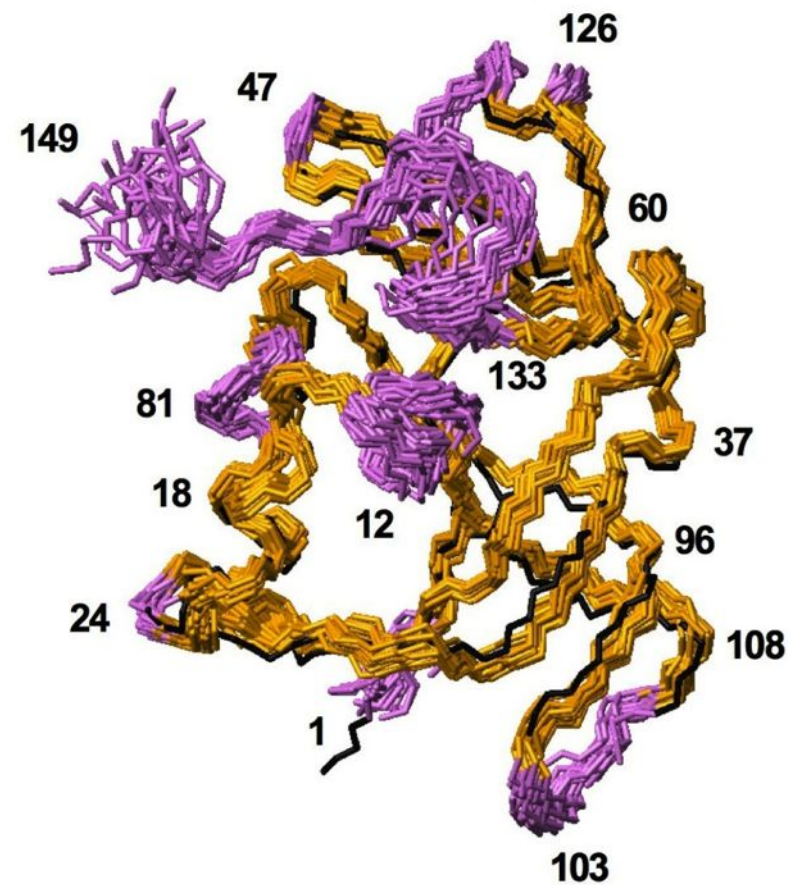


A2LD1 (*Mus musculus*)

NMR

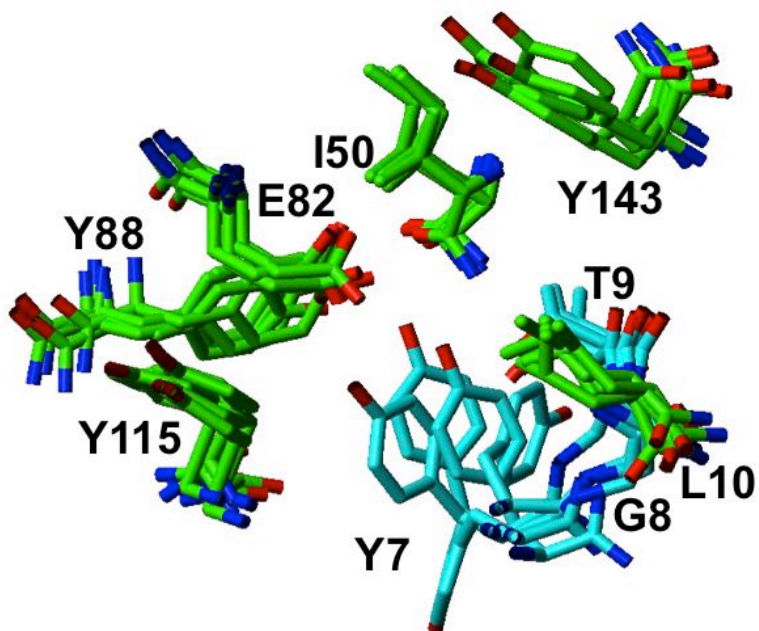


NMR/Crystal



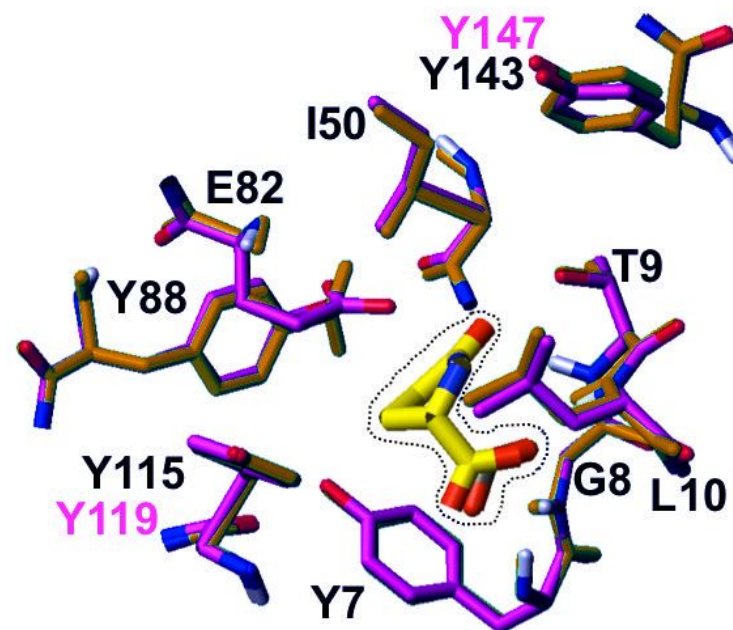
A2LD1 Active Site

NMR



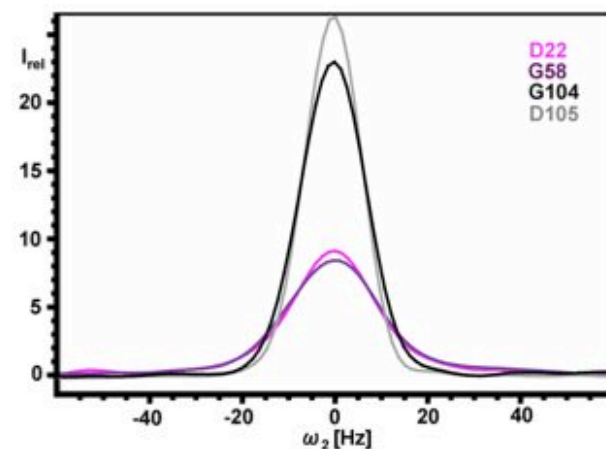
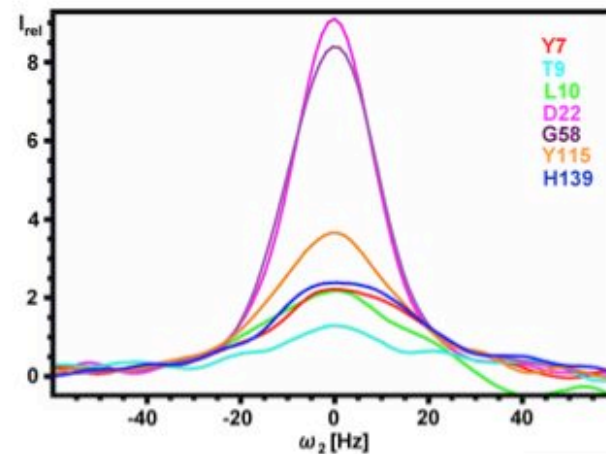
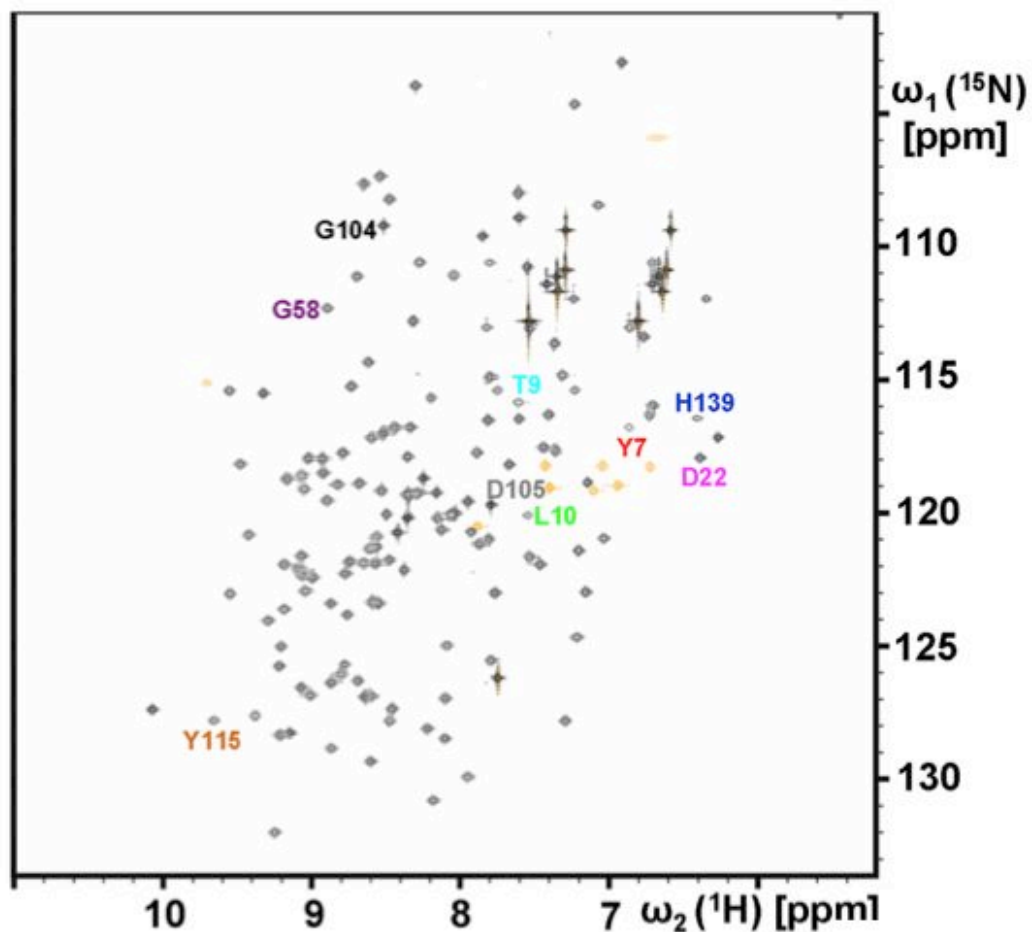
Crystal Structures

A2LD1 (formate)
GGACT (5-oxo-L- α -proline)



A2LD1

[¹⁵N,¹H]-HSQC Lineshapes



JCSG NMR Core

NMR Core Leader

Kurt Wüthrich

NMR Core Manager

Pedro Serrano

NMR Core Protein Production Manager

Michael Geralt

NMR Specialist

Reto Horst

Automation of Protein Structure Determination

Samit Dutta

Kristaps Jaudzems

Biswaranjan Mohanty

Lukas Susac

Atia-tul-Wahab

Collaborations:

Bill Pedrini

ETH Zürich

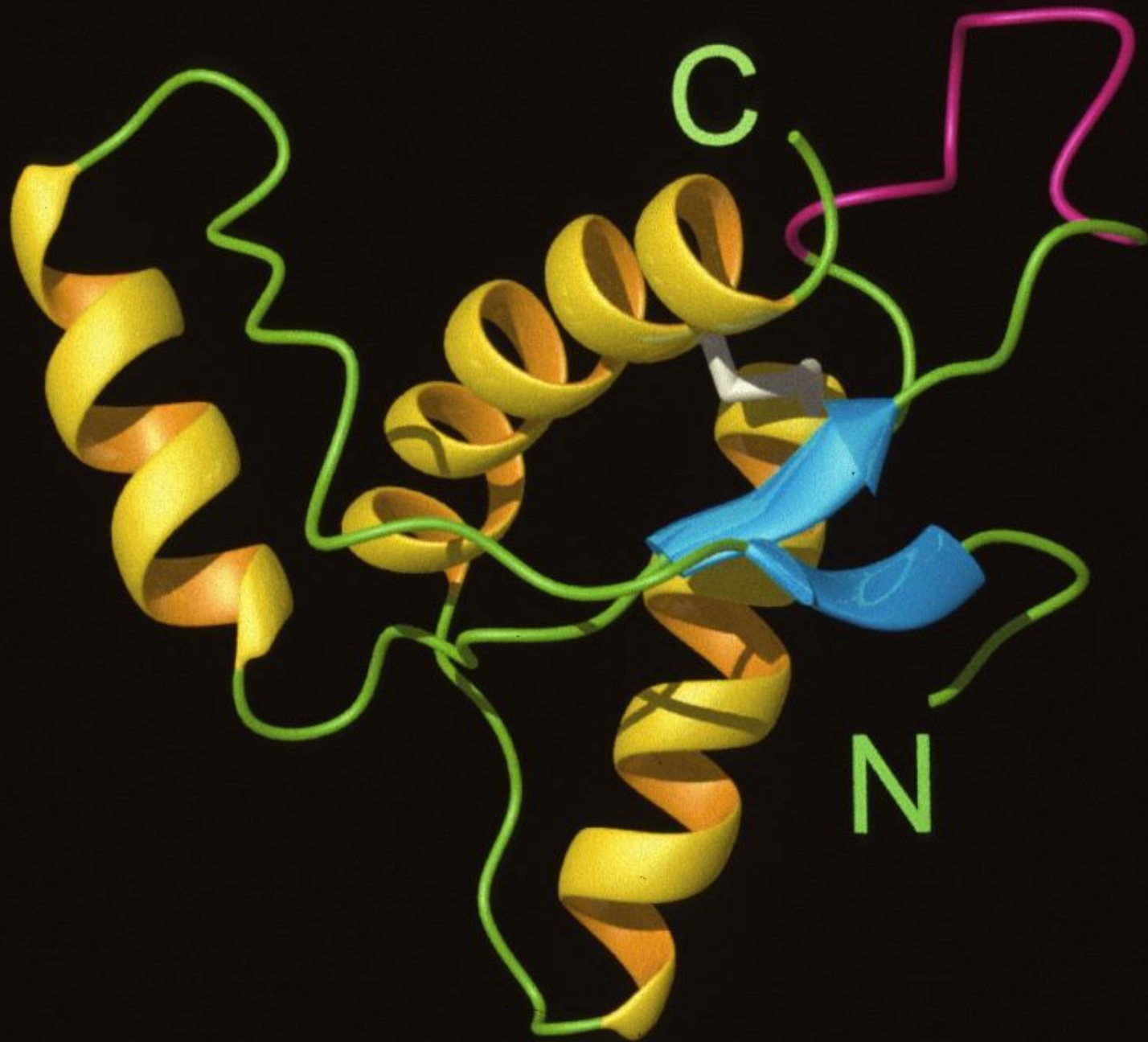
Torsten Herrmann

UCB Lyon

Prion Proteins

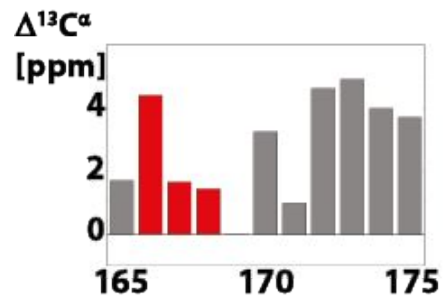
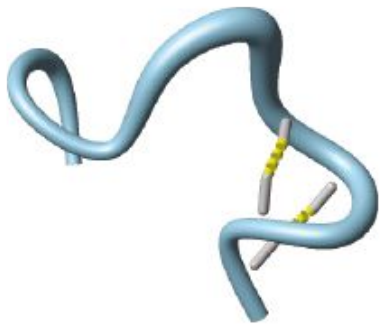
1996 –



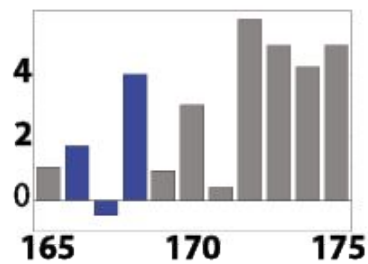
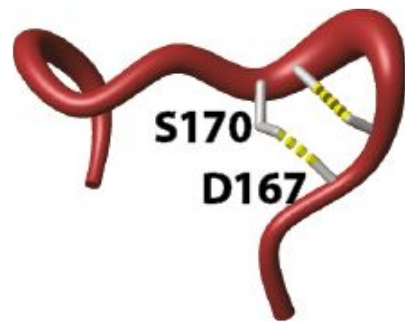


$\beta 2$ - $\alpha 2$ loop in variants of mPrP(121–231)

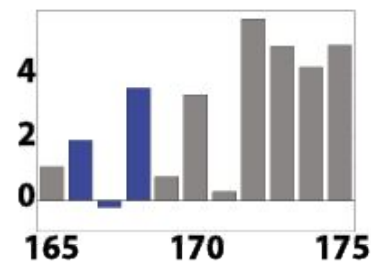
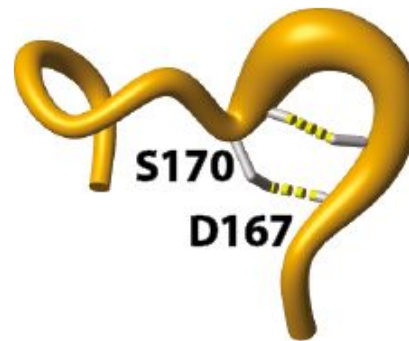
F175A



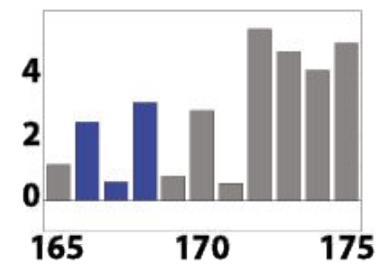
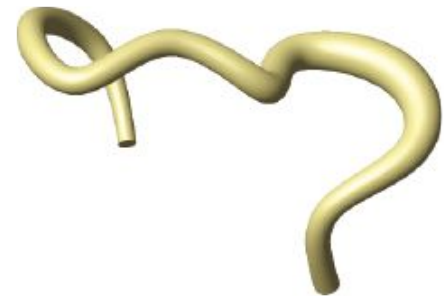
Y169G



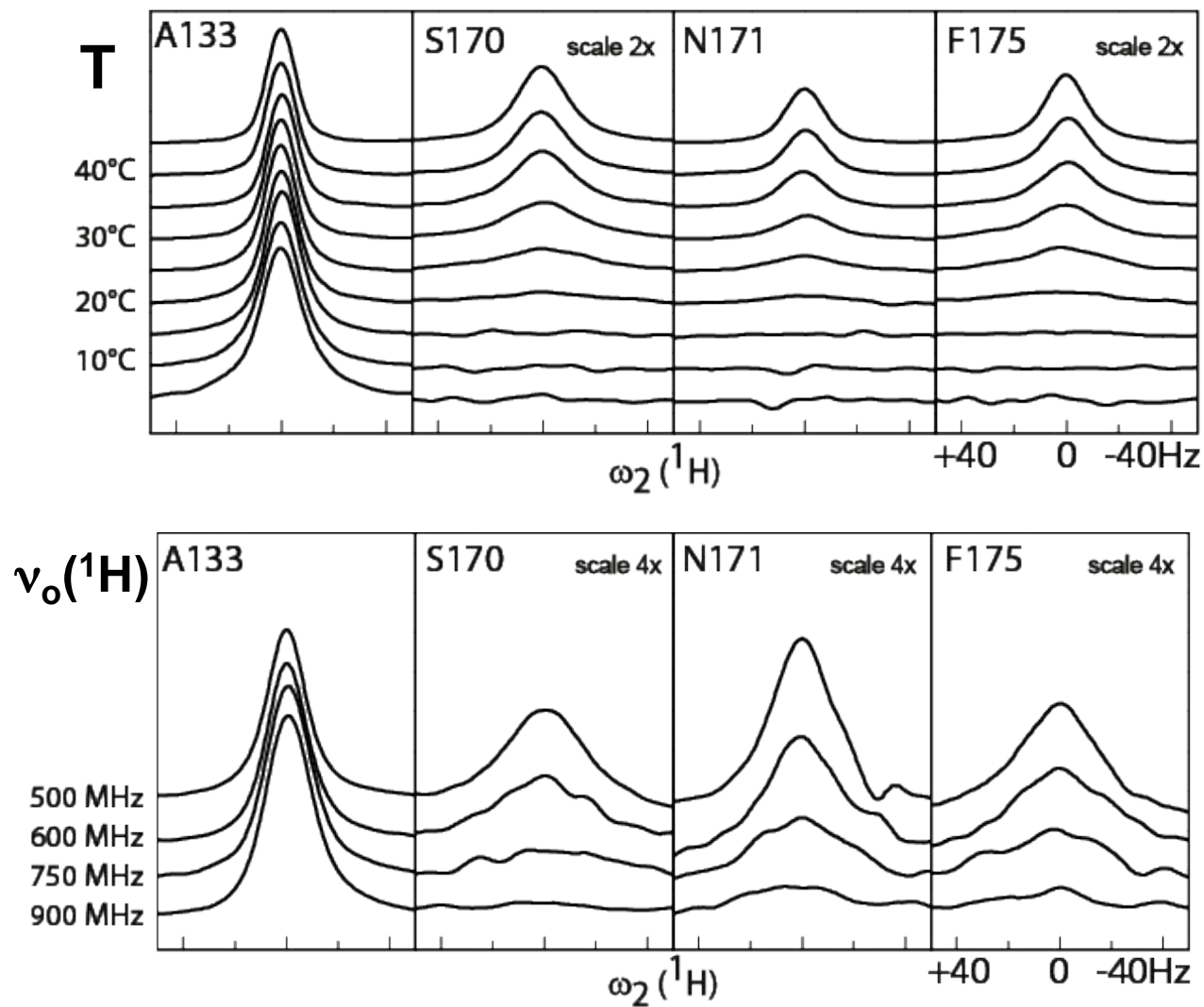
Y169A



Y169A/Y225A/Y226A



mPrP[Y225A, Y226A](121–231)



mPrP

Models: mPrP[Y169A \rightarrow Y]

$\chi^1 = +60$

$\chi^1 = -60$

