Intrinsically disordered proteins and their interactions

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Cagliari
GREECE
Aegean Sea
TURKEY
Mediterranean Sea
Palermo
Ionian Sea
Sicily
Tunis
Rhodes
TUNISIA
Valletta
MALTA
300 Kilometers
300 Miles

IDPs/IDRs

- Intrinsically disordered proteins/regions (IDPs/IDRs)
- Do not adopt a well-defined structure in isolation under native-like conditions
- Ensemble of very different conformations
- Functional proteins
- Often involved in PPIs

Article No. jmbi.1999.3110 available online at http://www.idealibrary.com on IDEAL® *J. Mol. Biol.* (1999) **293**, 321–331

JMB

Intrinsically Unstructured Proteins: Re-assessing the Protein Structure-Function Paradigm

Peter E. Wright* and H. Jane Dyson*

Experimental detection of disorder

From the literature

- Failed attempts to crystallize
- Lack of NMR signals
- Heat stability
- Protease sensitivity
- Increased molecular volume

NMR

- HSQC
- chemical shifts (CS)
- residual dipolar couplings (RDCs)
- paramagnetic relaxation enhancement (PRE)

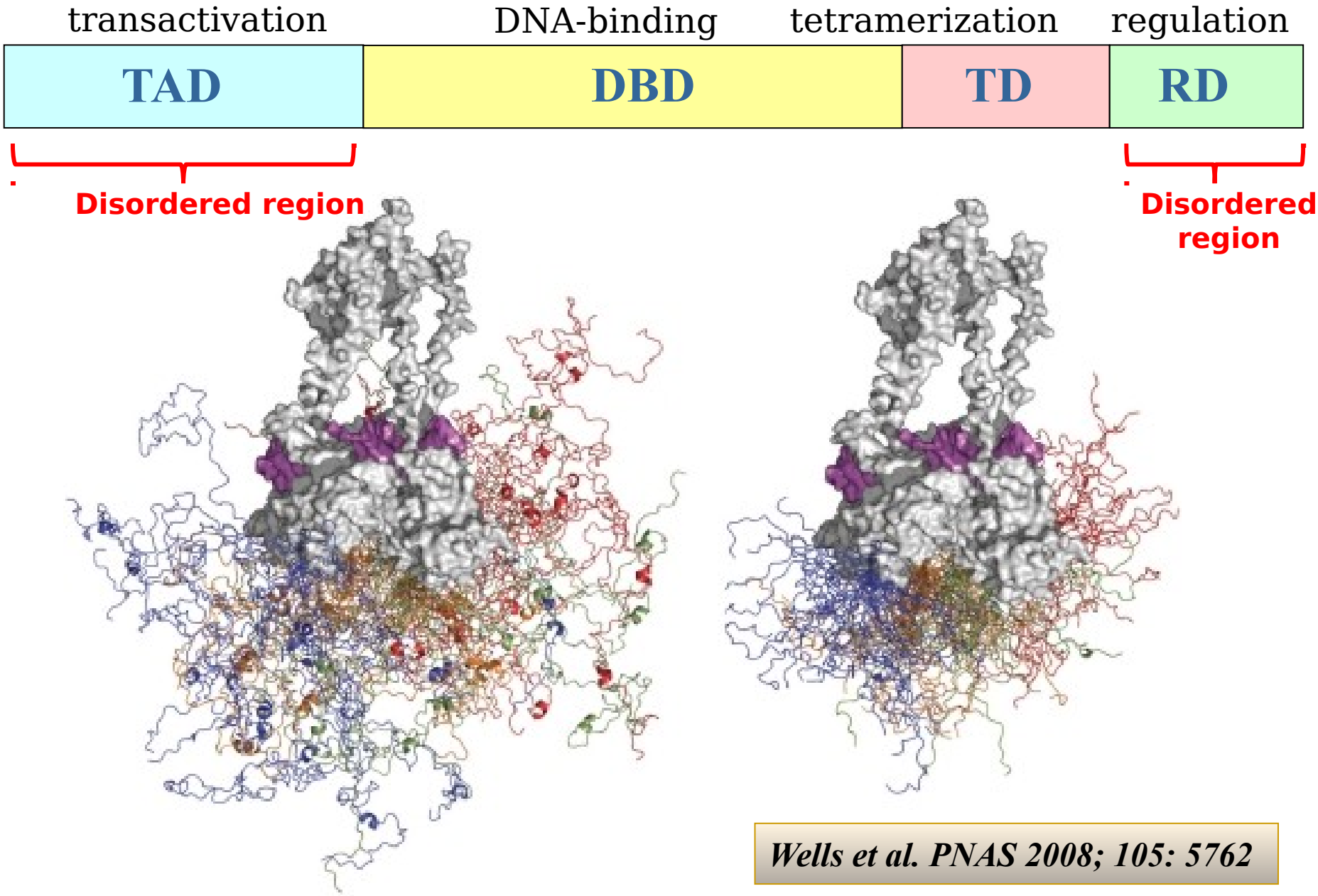
Disordered proteins

In the PDB

Missing electron density regions from the PDB

NMR structures with large structural variations

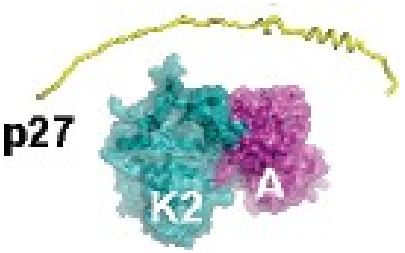
p53 tumor suppressor



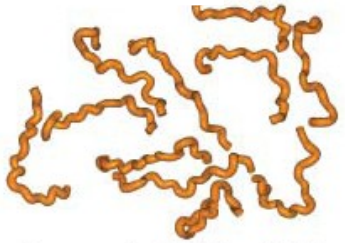
Wells et al. PNAS 2008; 105: 5762

Heterogeneity in protein disorder

Transient structures

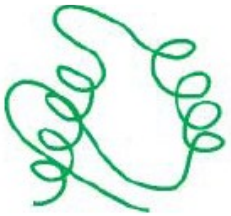


RC-like



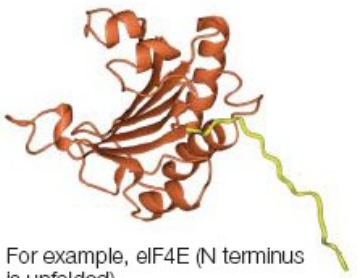
For example, ACTR (no NCBD)

Compact



For example, NCBD (no ACTR)

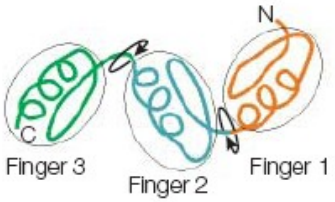
Mostly folded, local disorder



For example, eIF4E (N terminus is unfolded)

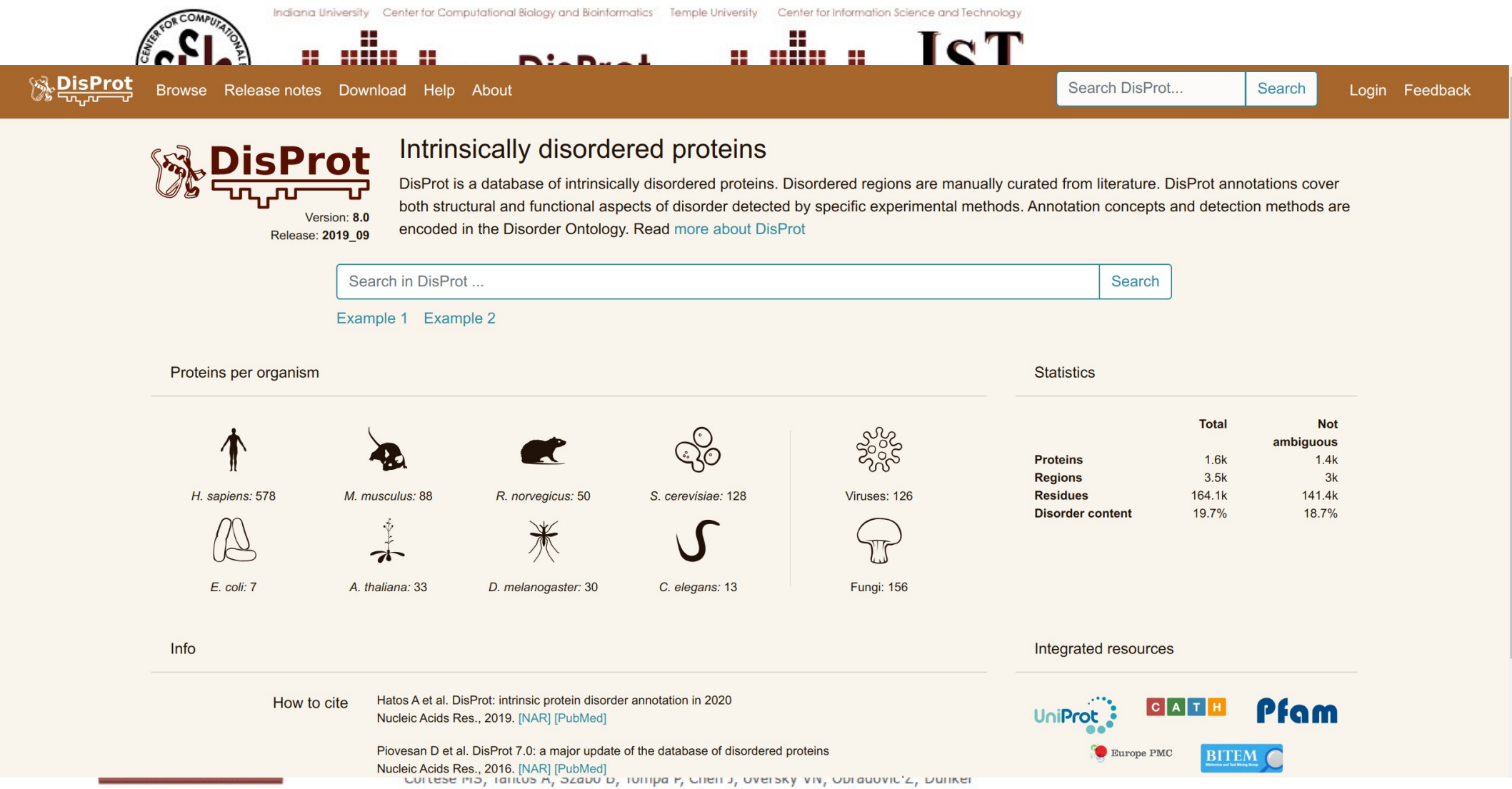
Flexible loop

Linked folded domains
(beads on a string)



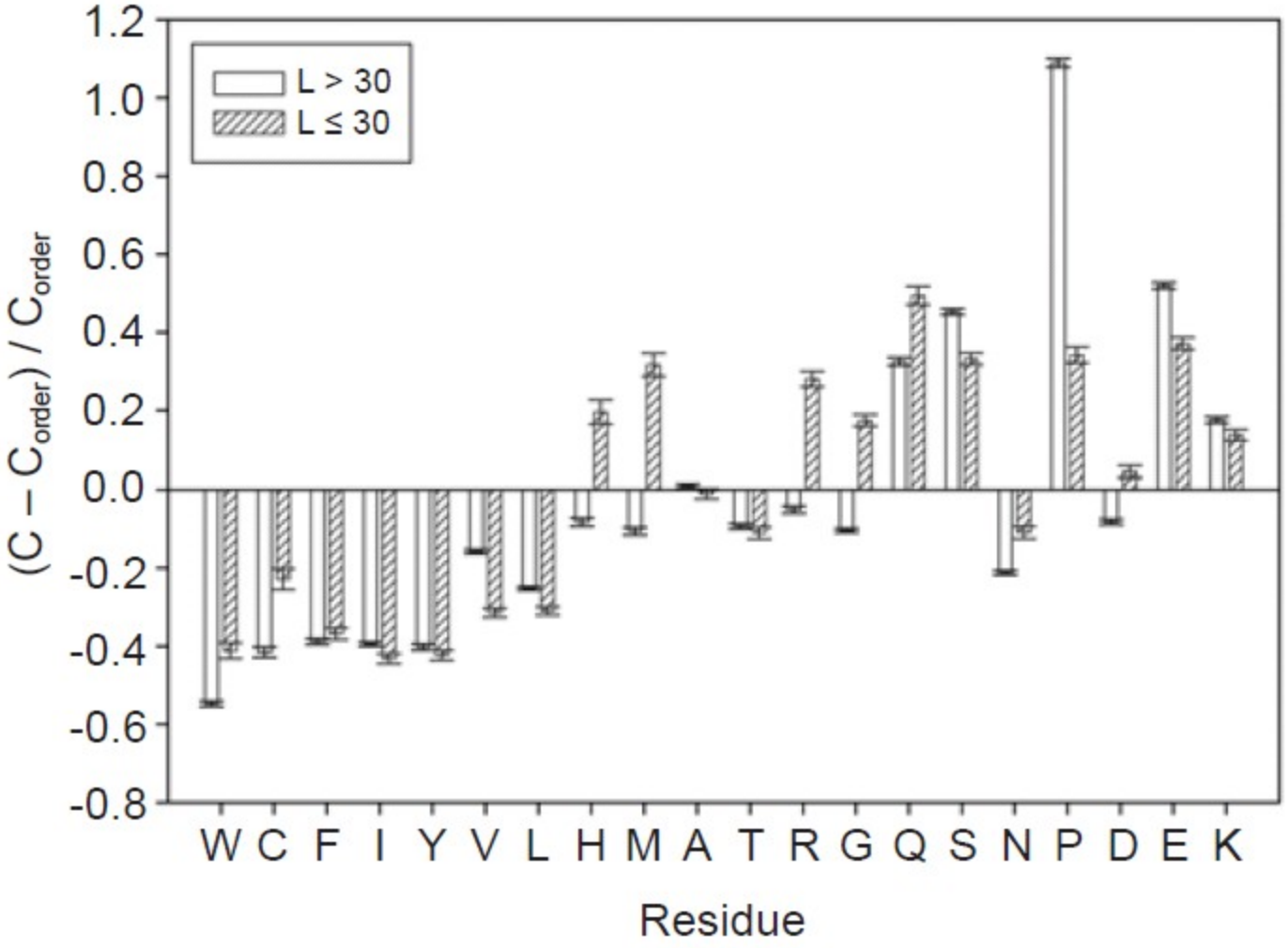
For example, zinc fingers (no DNA)

DisProt



Disprot – old version

Amino Acid Compositions



Protein disorder is encoded in the amino acid sequence

Prediction methods for protein disorder

Over 50 methods …

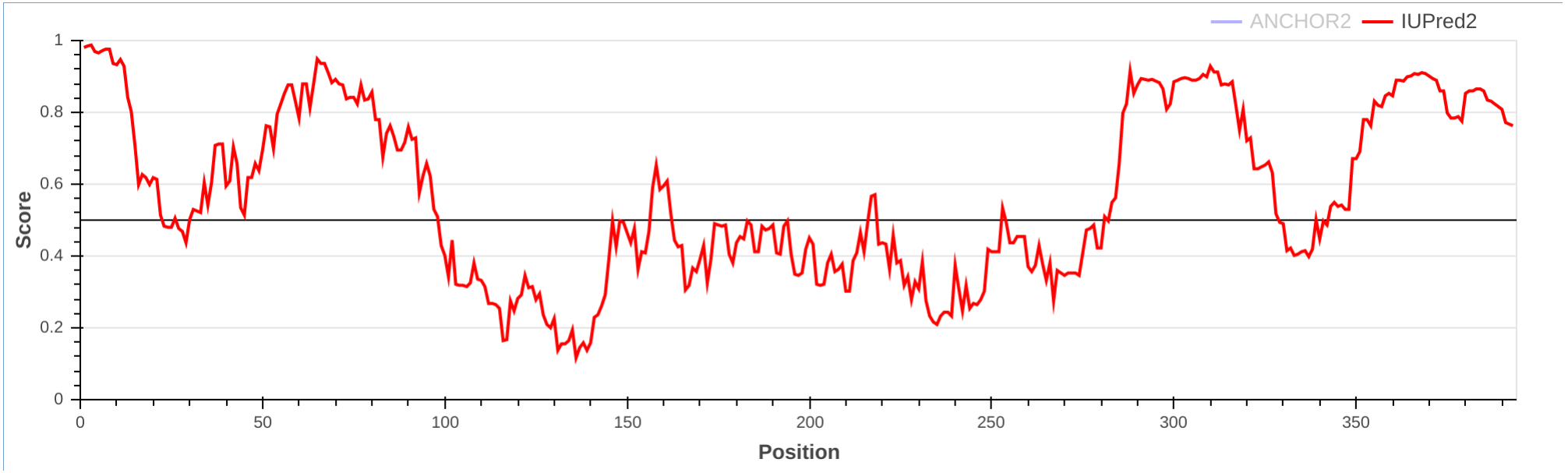
- Based on amino acid propensity scales
  - GlobPlot (secondary structure propensities)
- Simplified biophysical models
  - **IUPred**
- Machine learning approaches
  - Disopred, Espritz
- Meta servers
- Deep learning

IUPred

If a residue cannot form enough favorable interactions within its sequential environment, it will not adopt a well defined structure → it will be disordered

- Based on an energy estimation method
- Parameters calculated from statistics of globular proteins
- No training on disordered proteins

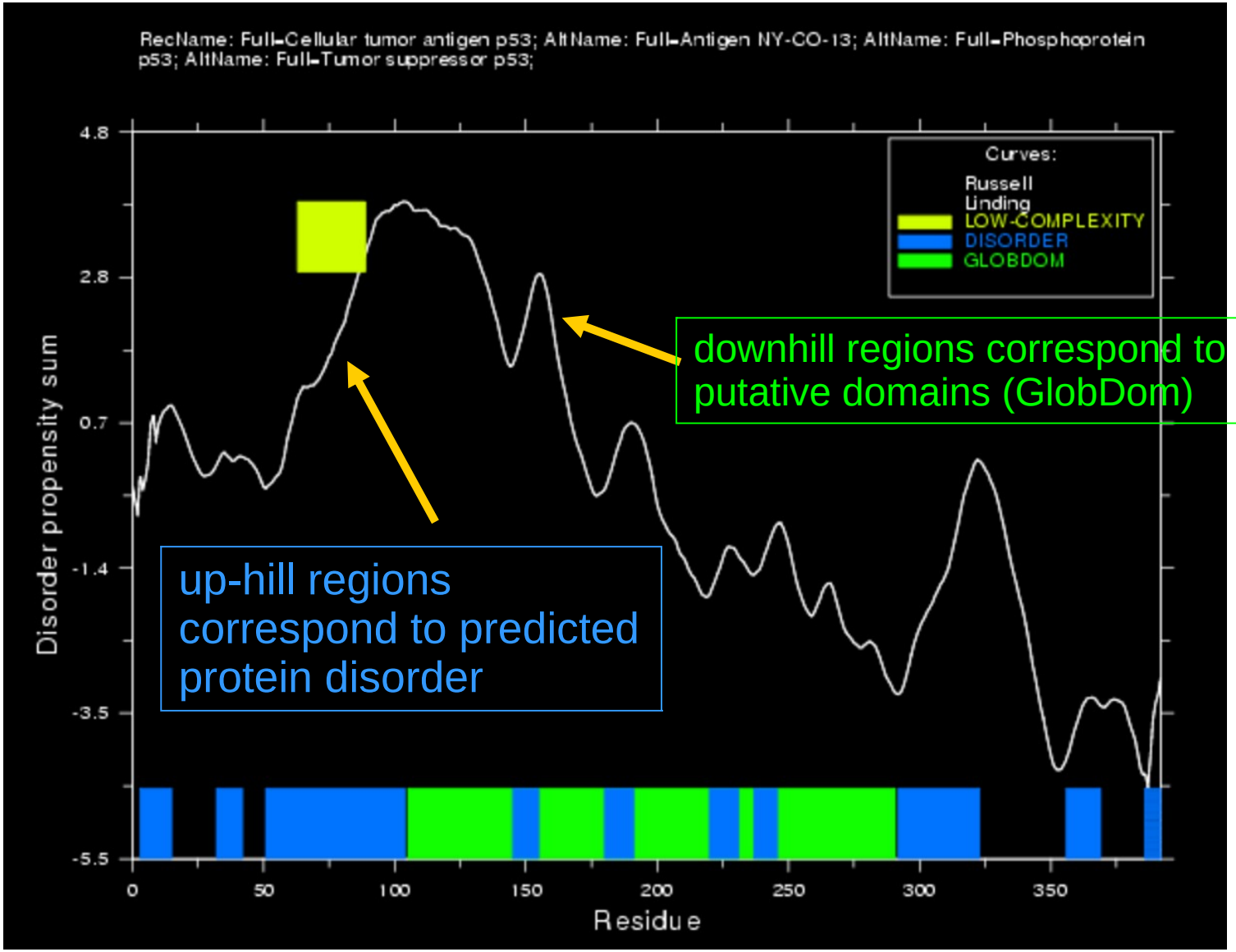
A typical output (IUPred2A)



Predictions are on a per residue basis

Dosztányi et. a. (2005) JMB 347, 827
Erdős et al. (2018) ***NAR 46(W1):W329***

GlobPlot: http://globplot.embl.de/

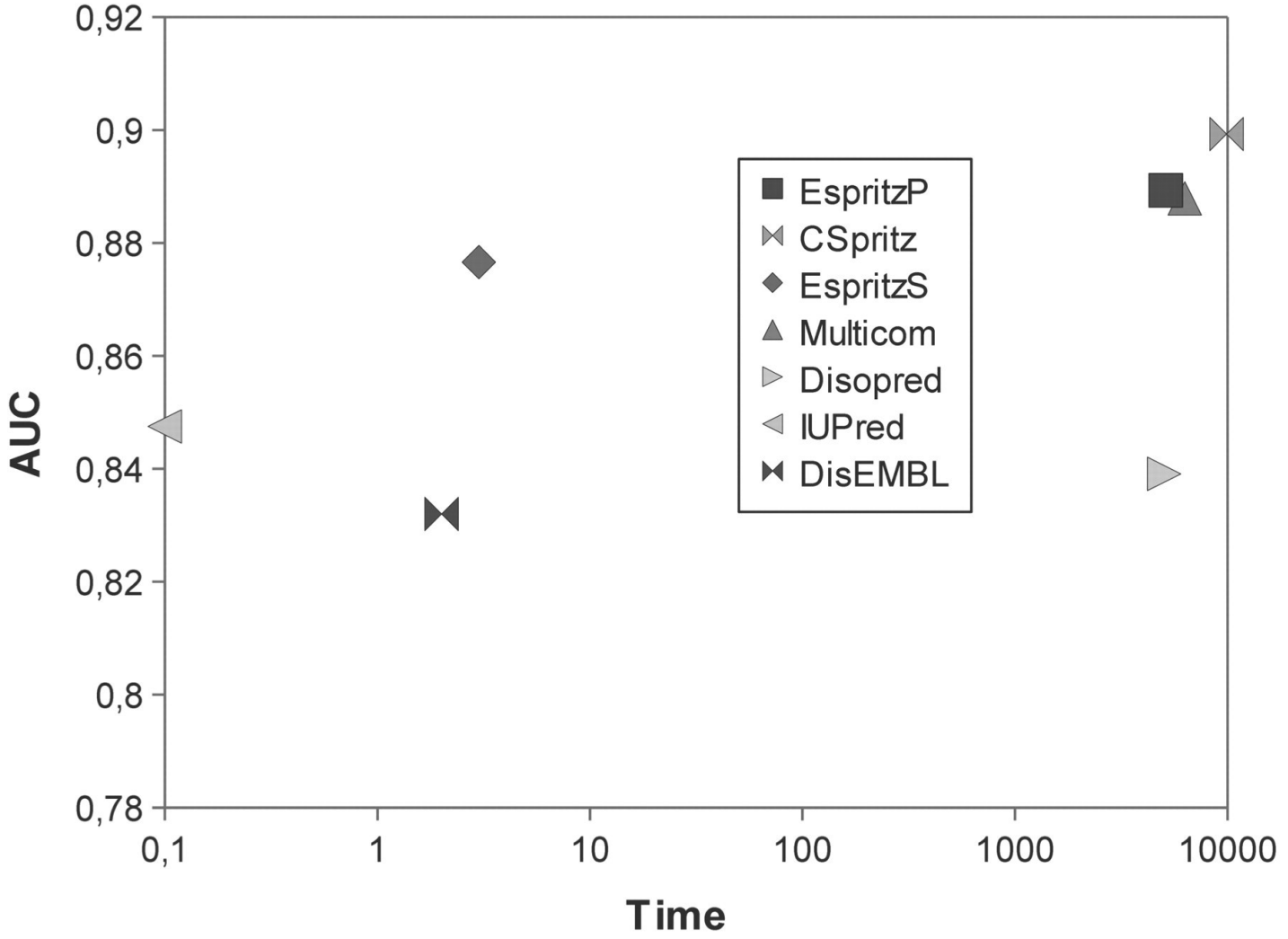


Where are the ordered domains, disordered regions?

Prediction of protein disorder

- Disordered is encoded in the amino acid sequence
- Can be predicted from the sequence
- ~80% accuracy (>0.8 AUC)
  - *Necci et al. 2018. Bioinformatics 34, 445*
  - *Neilsen&Mulder 2019. Sci Rep, 9, 5137*
  - *CAID*
- Challenges
  - Small, noisy datasets
  - Disorder is heterogeneous
  - “Flavors” of disorder

Which is the best method? Speed

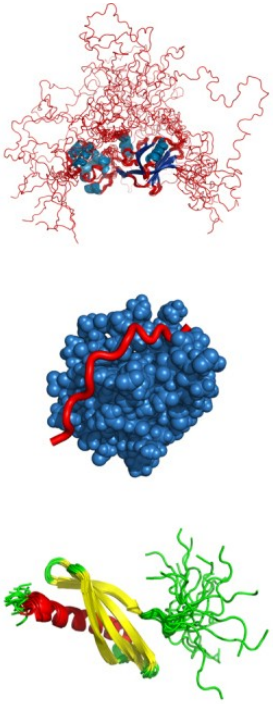


Time versus performance plot for different predictors.

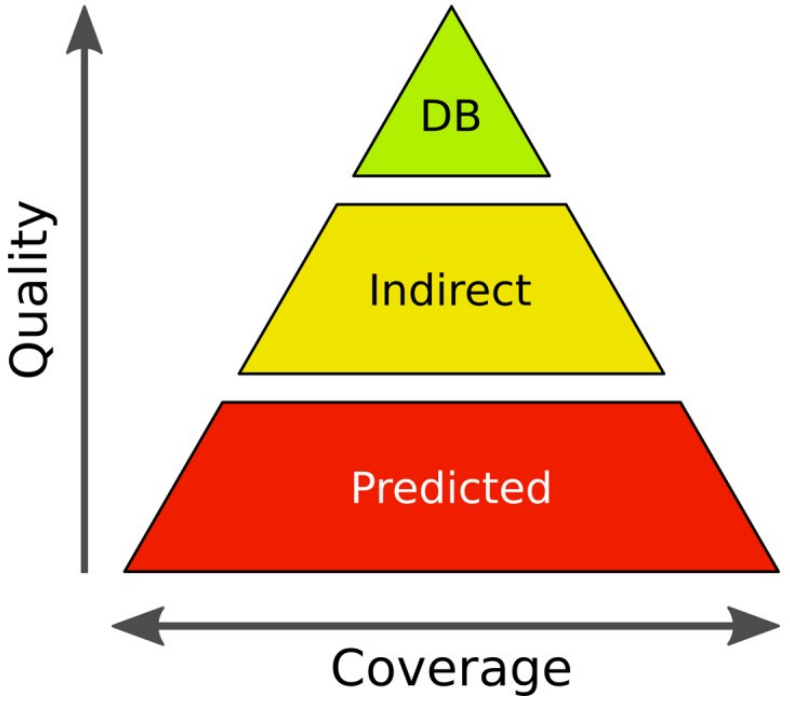
Ian Walsh et al. Bioinformatics 2012;28:503

MobiDB

A

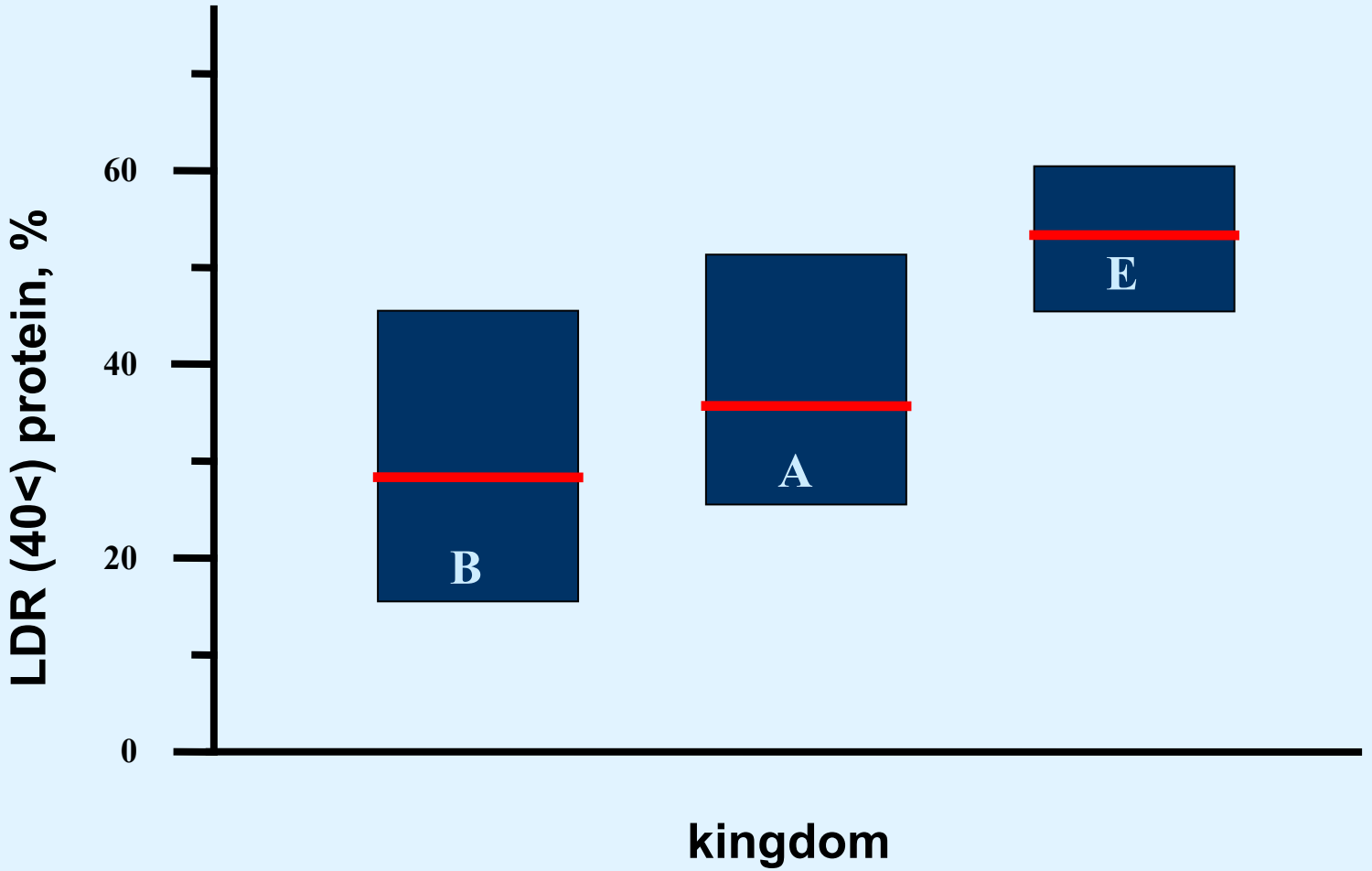


B



Piovesan D. et al. NAR. 2018;46(D1):D471–D476.

Protein disorder is prevalent



Protein disorder complements the functional repertoire of globular proteins

Table 2. Correlation and anticorrelation of structural disorder with Swiss-Prot functional categories

Top functions that correlate with long disorder[a]	Top functions that anticorrelate with long disorder
Differentiation	GMP biosynthesis
Transcription	Amino acid biosynthesis
Transcription regulation	Transport
Spermatogenesis	Electron transport
DNA condensation	Lipid A biosynthesis
Cell cycle	Aromatic hydrocarbons catabolism
mRNA processing	Glycolysis
mRNA splicing	Purine biosynthesis
Mitosis	Pyrimidine biosynthesis
Apoptosis	Carbohydrate metabolism
Protein transport	Branched-chain amino acid biosynthesis
Meiosis	Lipopolysaccharide biosynthesis

Xie H et al. J Proteome Res. 2007, 6, 1882

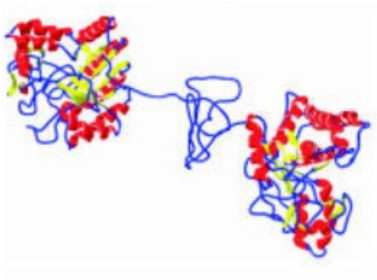
Protein disorder is important

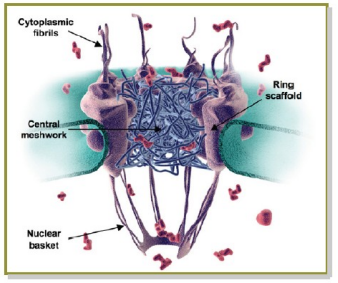
- Prion protein → Prion disease
- CFTR → Cystic fibrosis
- τ → Alzheimer's
- α-synuclein → Parkinson's
- p53, BRCA1 → cancer

How IDPs carry out their functions?

- Entropic chains

  Function directly results from disordered state





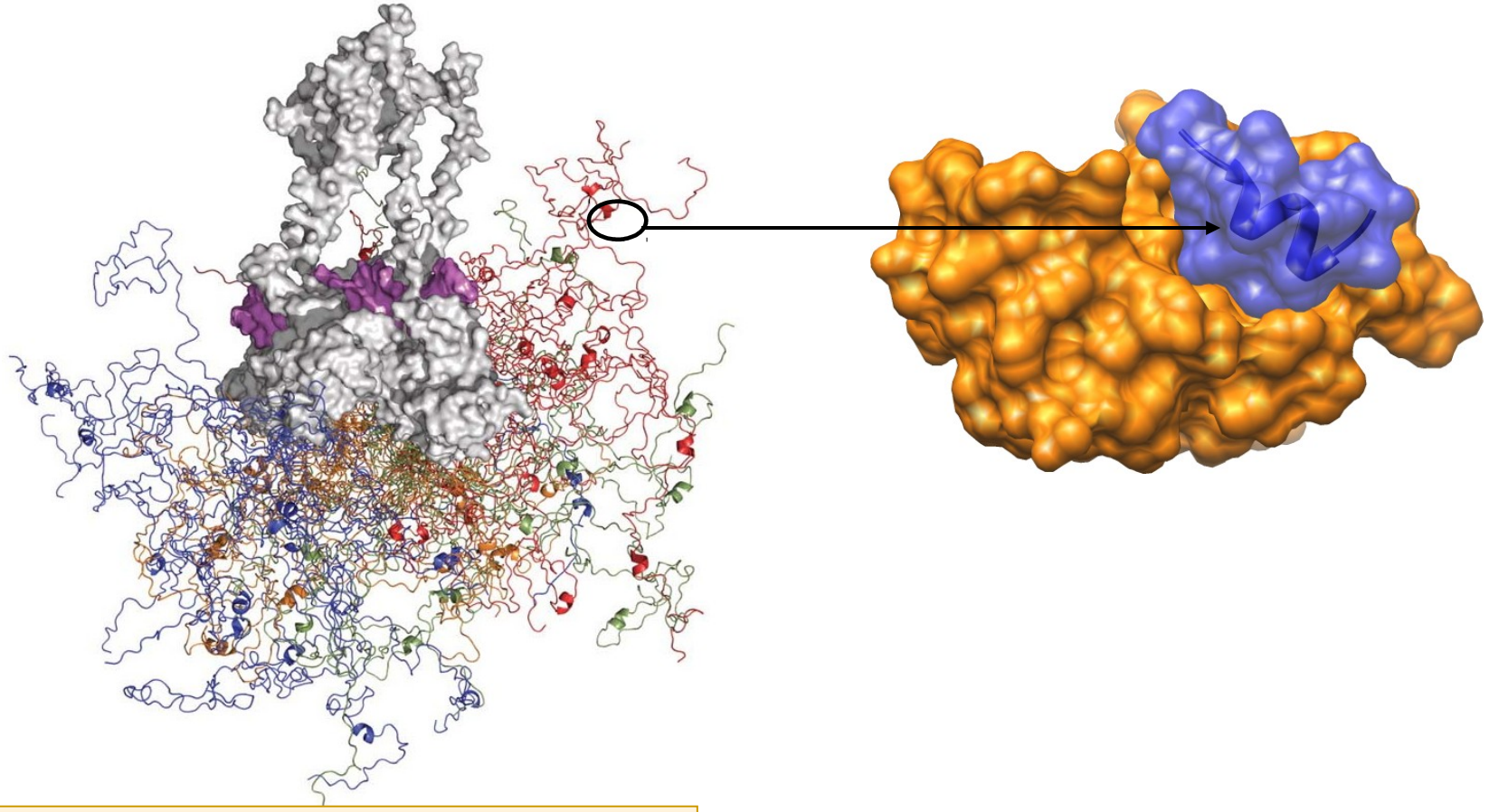
- Molecular recognition

  Coupled folding and binding

- “Assemblages”

  Functional sites formed by phase separation

Interaction of IDPs



Complex between p53 és MDM2

Coupled folding and binding

Binding regions within IDPs

- **SLIMs: Short linear motifs**

  3-11 residues long, average size 6-7 residues

  enriched in IDRs

- **Disordered binding sites, Morfs, LIPs**

  undergo disorder to order transition upon binding

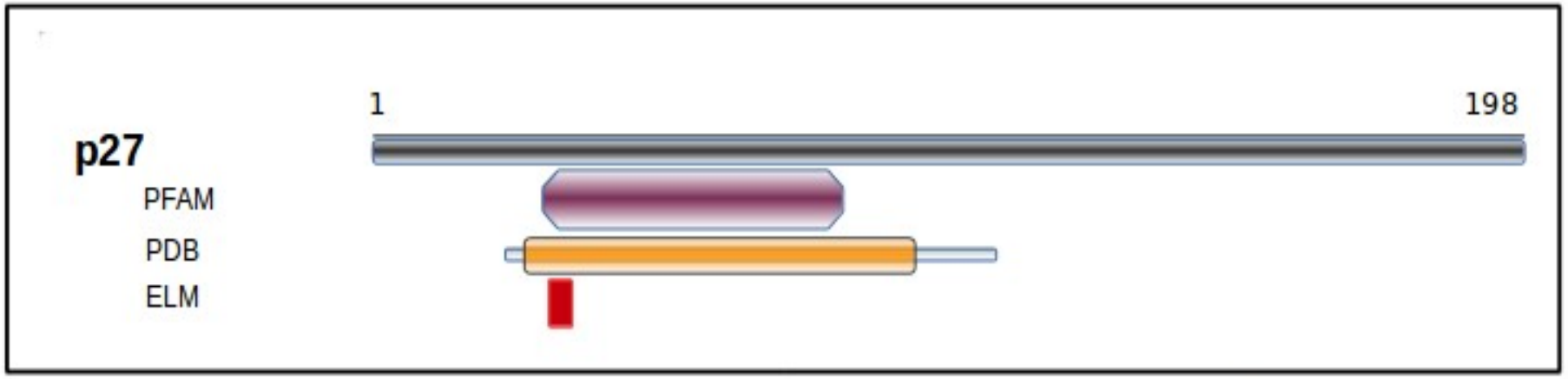
  usually less then 30 residues, can be up to 70

- **Intrinsically disordered domains**

  evolutionary conserved disordered segments

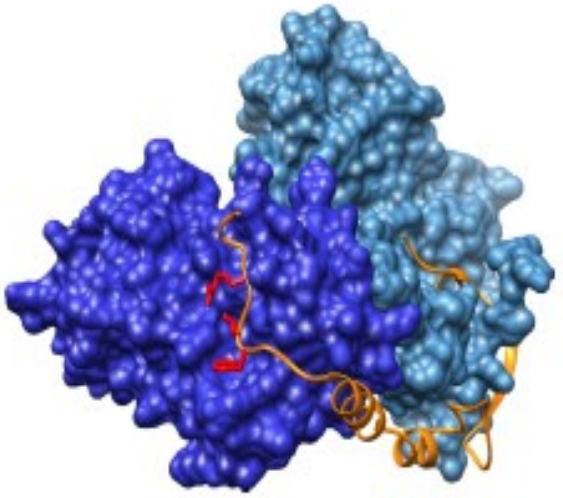
p27

Inhibitor of CDK2-CyclinA complex.



[RK].L.{0,1}[FYLIVMP]

CDN1B_HUMAN	30-33	HPKPSACRNLFGPVDHEEL
MPIP1_HUMAN	11-15	PEPPHRRRLLFACSPPPAS
CDC6_HUMAN	94-98	HSHTLKGRRLVFDNQLTIK
RB_HUMAN	873-877	SNPPKPLKKLRFDIEGSDE
P53_HUMAN	381-385	GQSTSRHKKLMFKTEGPDS
VE1_HPV18	127-130	SGQKKAKRRLFTISDSGYG

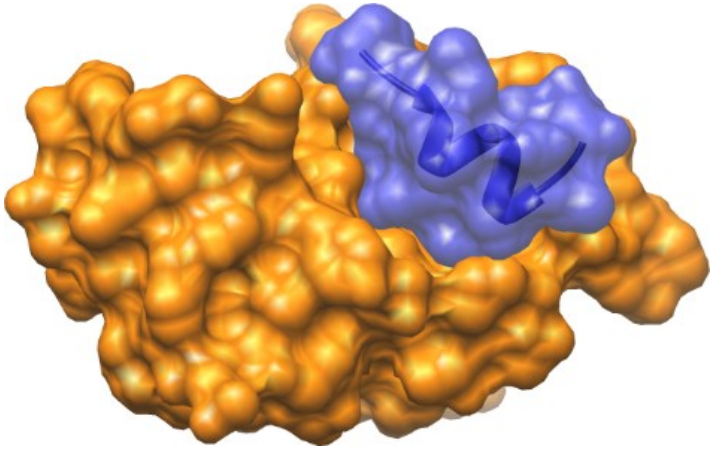


Database of Disordered Binding Sites

DIBS

- Experimentally verified to be disordered
  - Manual, Disprot, ELM, IDEAL, DisBind
- Forms a complex with ordered partners
- **Kd values**

773 proteins
(1577 structures)



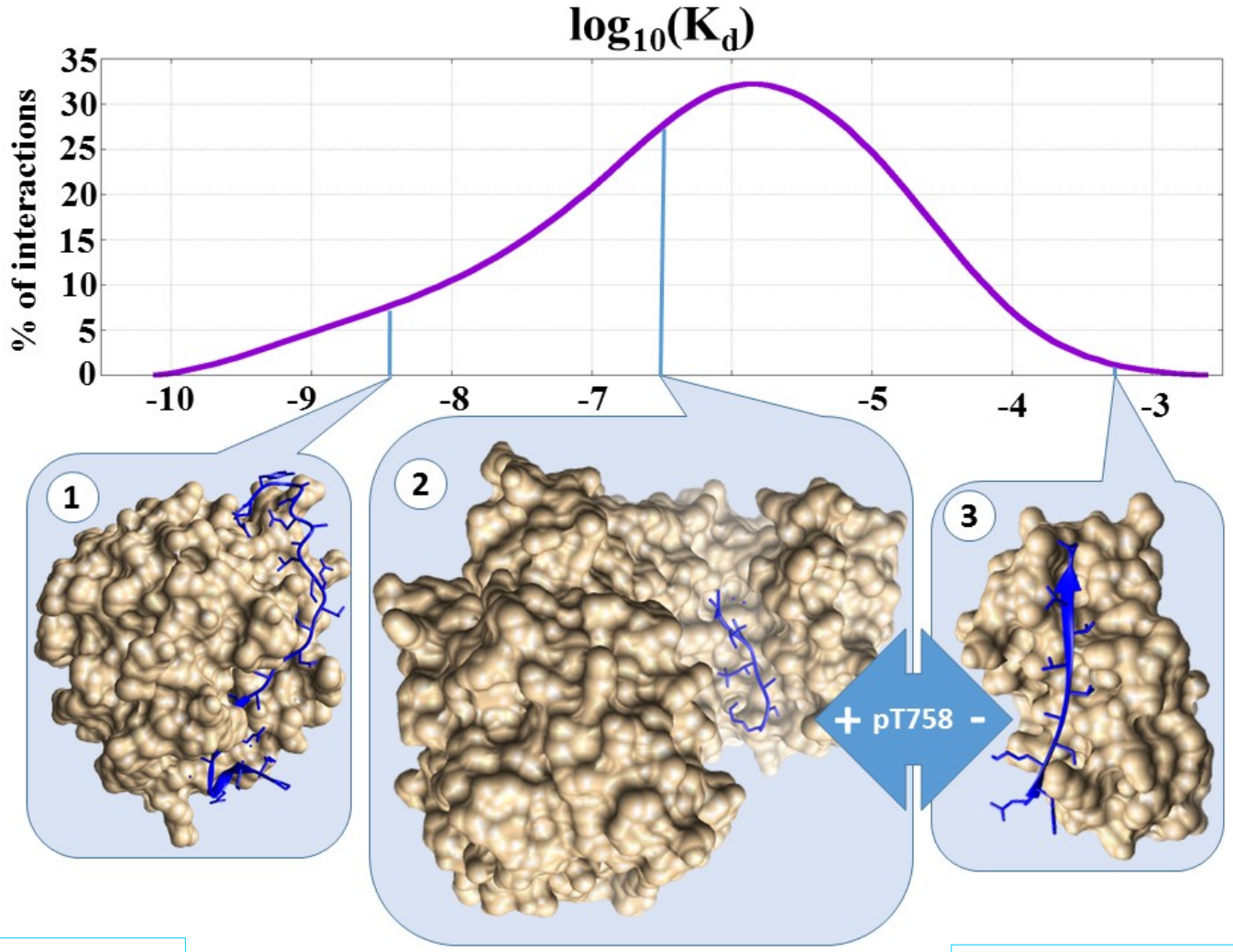
Databases and ontologies

DIBS: a repository of disordered binding sites mediating interactions with ordered proteins

Eva Schad[1], Erzsébet Fichó[1], Rita Pancsa[2], István Simon[1], Zsuzsanna Dosztányi[3,*] and Bálint Mészáros[1,3,*]

Bioinformatics (2017)
doi: 10.1093/bioinformatics/btx640

Kd values



Thrombin-anophelin

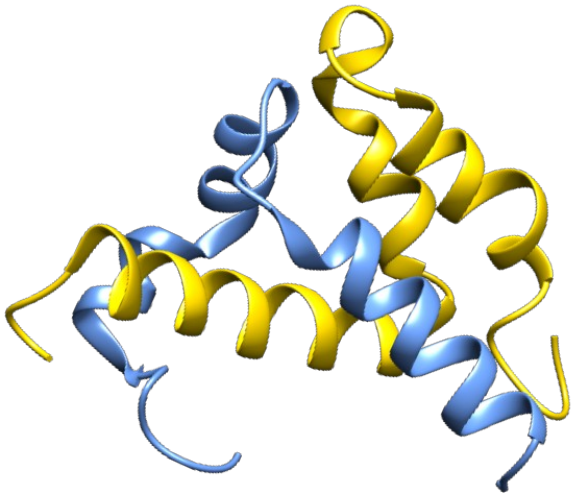
14-3-3ζ-integrin β2

filamin-integrin β2

Mutual Folding Induced by Binding

MFIB

- Both partners are experimentally verified to be disordered
  - Manual, Disprot



205 proteins
(1406 structures)

Databases and ontologies

MFIB: a repository of protein complexes with mutual folding induced by binding

Erzsébet Fichó[1], István Reményi[2], István Simon[1,*] and Bálint Mészáros[1,*]

Bioinformatics (2017)
doi: 10.1093/bioinformatics/btx486

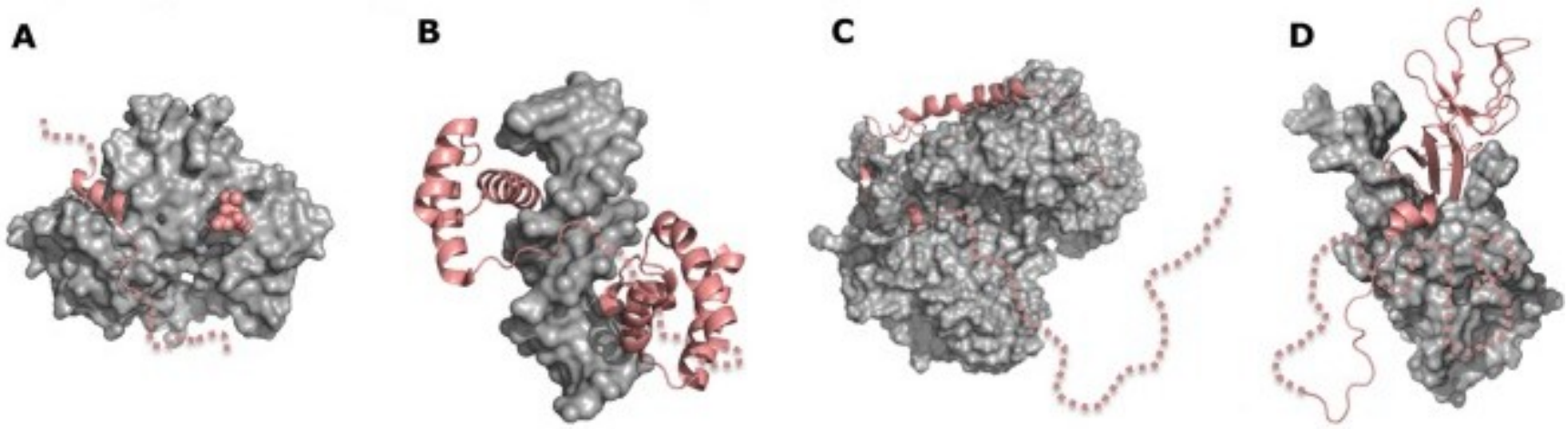
FuzDB

FUZZY COMPLEXES DATABASE

Home | Browse | Search | Methods | Classes | Upload | Analysis | Help | FAQs

Database of fuzzy protein complexes
FuzDB v3.3

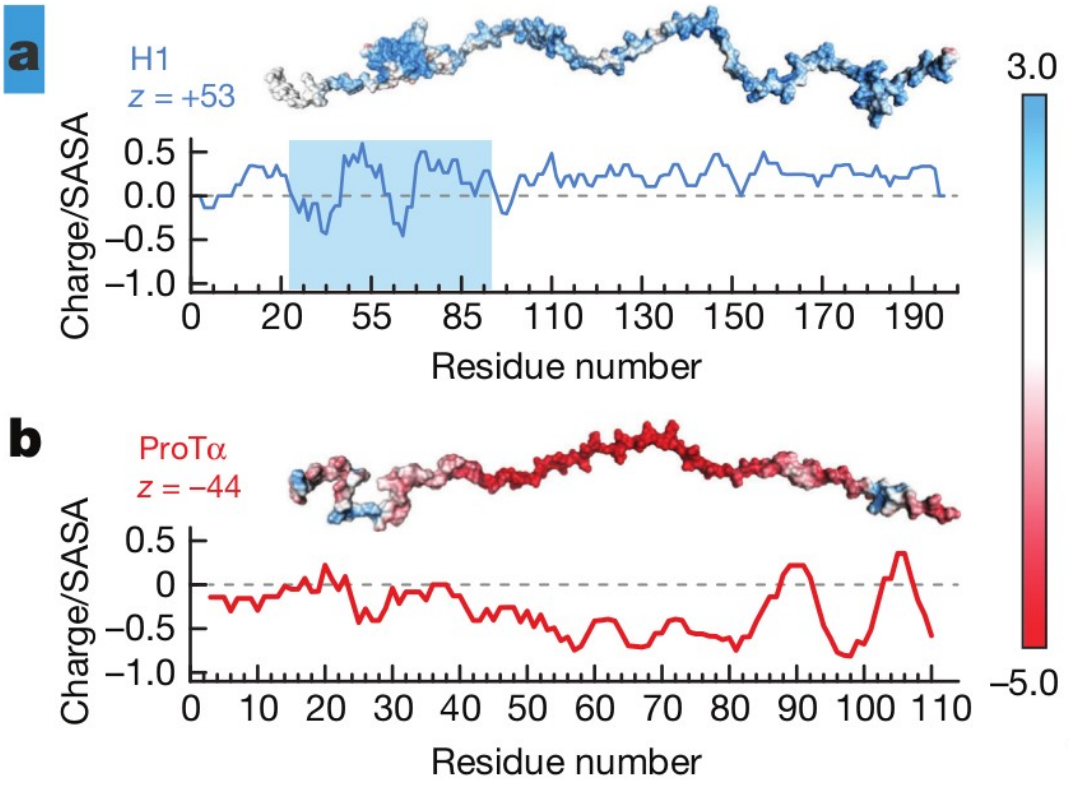
Citation: Nucleic Acids Research (2017) Jan 4;45(D1):D228-D235. abstract full [PDF]

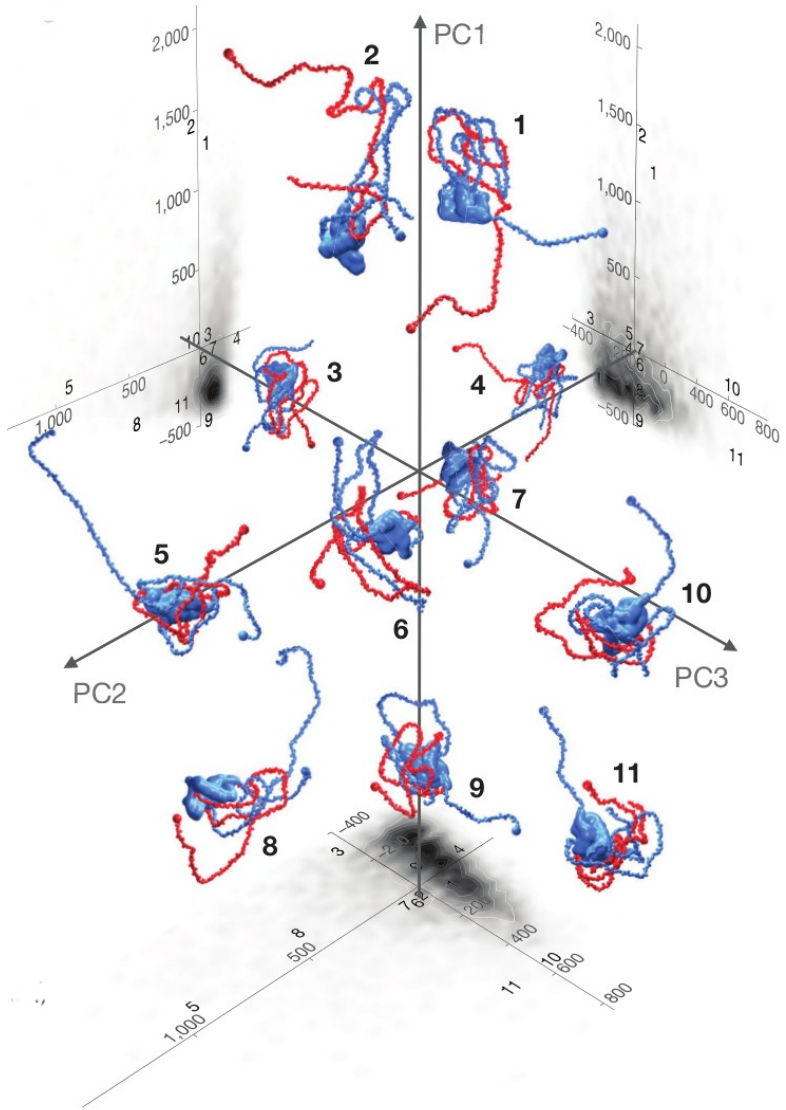


polymorphic clamp flanking random

Fuzzy complexes

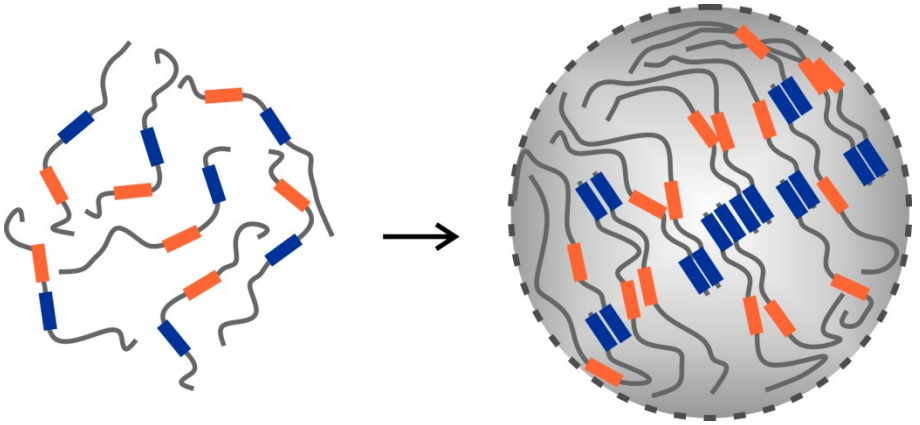
Extreme disorder in complex





Borgia et al. Nature 2018, 555, 61.

Liquid-liquid phase separation



formation of liquid/gel-like condensates of protein, RNA, and other biomolecules

driven by multivalent weak interactions

delimited by a phase boundary (no membrane)

Nucleolus, stress granules, P-bodies, Germ granules, heterochromatin, post-synaptic densities,

PhaSePro: https://phasepro.elte.hu/

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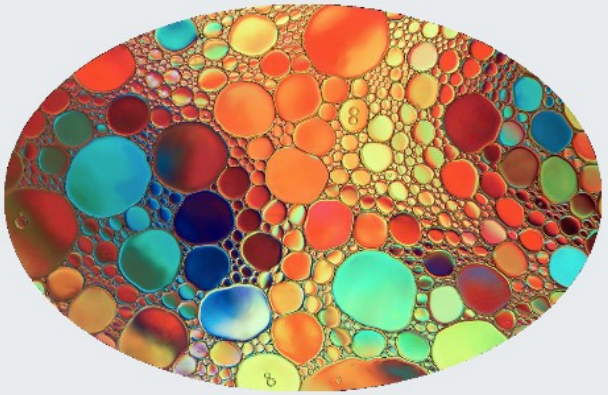
Welcome to PhaSePro!

PhaSePro is the comprehensive database of proteins driving liquid-liquid phase separation (LLPS) in living cells. LLPS is a molecular process employed by all living organisms to form membraneless organelles, mediating crucial cellular functions. PhaSePro is manually curated, it is solely based on experimentally verified cases of LLPS, integrating a wide range of information on the biophysical driving forces, biological function and regulation of these molecular systems.

Learn more »



Search for gene names, full or partial common/UniProt protein names, or UniProt accessions. Example 1 Example 2

Getting started

To get an introduction to the structure and use of PhaSePro, you can visit the selected examples (FUS and TDP-43) by clicking the buttons above, or read the About/Help pages by clicking below.

View details »

Explore

You can start searching the database by entering keywords in the above field, or by browsing the available entries by clicking below.

Browse/Search entries »

Annotate

Help us expand the knowledge about proteins involved in liquid-liquid phase separation by submitting new entries into PhaSePro.

View details »

Partners

TYPE OF PARTNER
Protein-protein binding
Protein-DNA binding
Protein-RNA binding
Protein-lipid binding
Protein-metal binding
Protein-inorganic salt binding

IDR – functional ontology (CV)

I) molecular function of disorder (MFUN)
- type of functional readout of function (such as MF in GO)

II) type of molecular transition (TRAN)
- necessary for function (such as disorder-to-order transition)

III) molecular partner (PART)
- type of partner recognized (protein, RNA, DNA, metabolite)

Predicting function for IDRs

Prediction of binding sites

- Interaction sites are usually linear (consist of only 1 part)
- Enrichment of interaction prone amino acids
- Can be predicted from sequence without predicting the structure
- ~ 10 Methods
  - Biophysical methods (*ANCHOR*)
  - Machine learning (*MorfPred, Morf$_{chibi}$, DISOPRED3)*
  - Evolutionary approaches (*SlimPrints, PhyloHMM*)

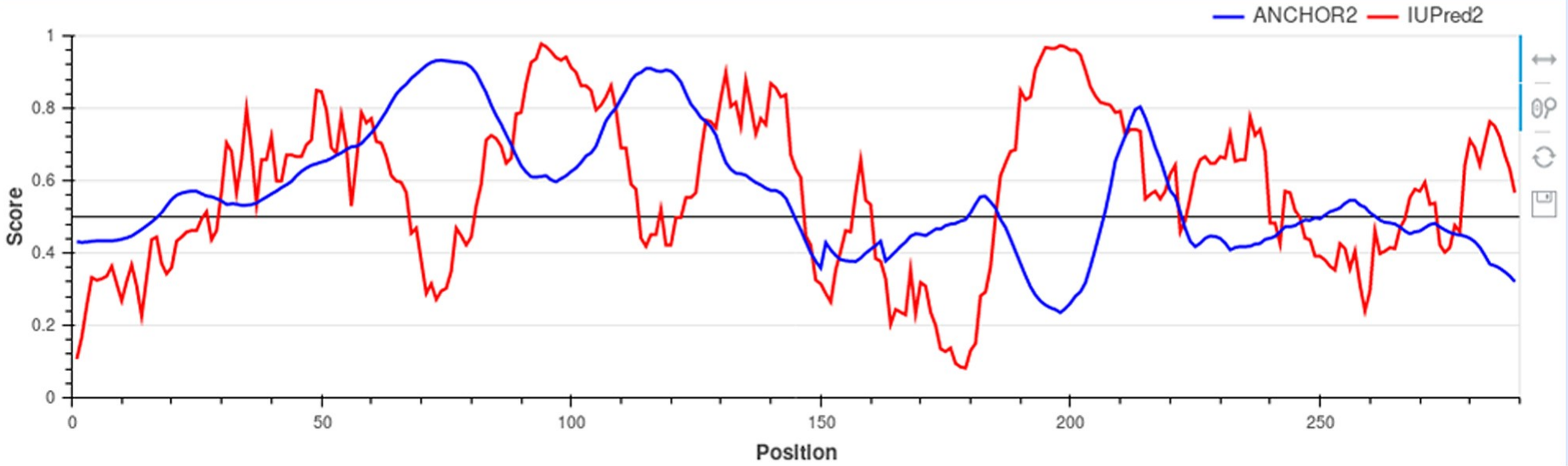
Prediction of disordered binding sites – ANCHOR

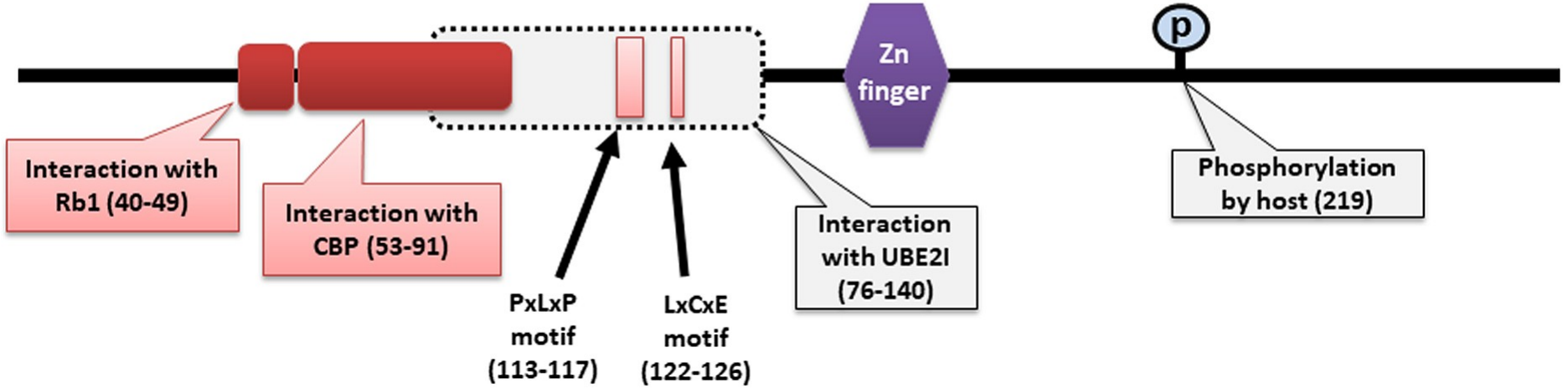
- What discriminates disordered binding regions?
  - A cannot form enough favorable interactions with their sequential environment
  - It is favorable for them to interact with a globular protein

- Based on simplified physical model
  - Based on an energy estimation method using statistical potentials

ANCHOR2

>sp|P03255|E1A_ADE05 Early E1A protein OS=Human adenovirus C serotype 5 PE=1 SV=1





Human adenovirus C early E1A protein