Mass Spectrometry Based Modelling of Macromolecular Assemblies

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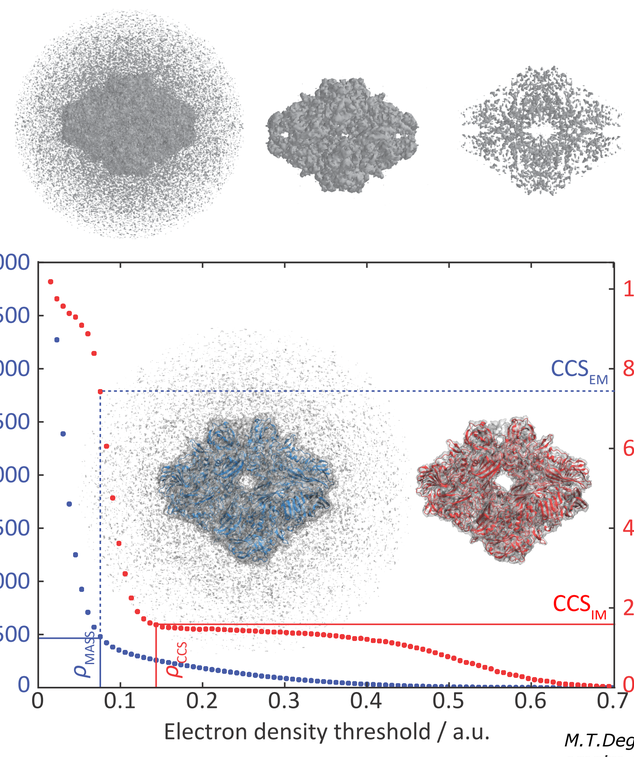
- *We develop computational methods to study structure and dynamics of proteins and protein assemblies*
- *Our software relies on BiobOx, our Python package for structural biology*
- *With our methods we study small Heat Shock Proteins (sHSP) assemblies and their binding to client proteins*

Collision cross section of EM maps



Creating a bead model of an EM map allows calculating its CCS

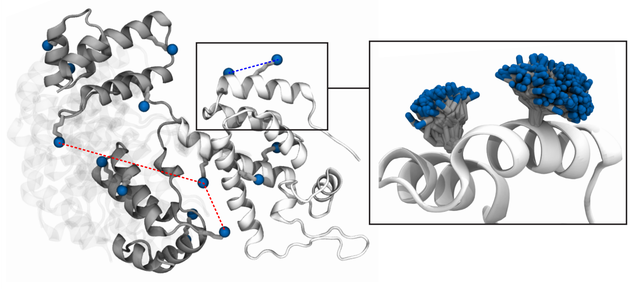
CCS depends on the used electron density cutoff ρ

A criterion to identify the most suitable cutoff: ρ_{CCS} depends on protein mass and EM map resolution

<4% error in estimation of CCS from simulated EM maps, <8% from real ones

M.T.Degiacomi and J.L.P Benesch, Analyst, 2016
emnim.chem.ox.ac.uk

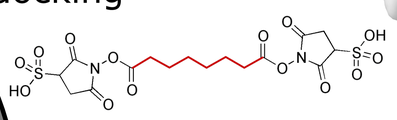
Accurate cross-linking distances



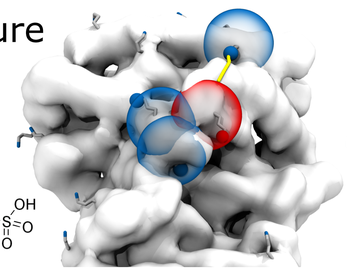
Cross-linking possible depending on cross-linker, side chains and domain dynamics

A new x-linking distance metric: shortest path length between ensembles of alternative side chain orientations and protein conformations

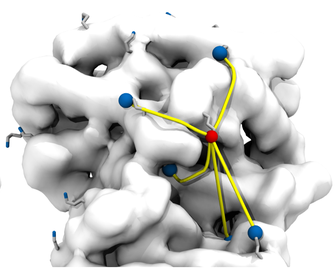
- Explicitly accounts for molecular flexibility
- Improves rationalization of x-linking data vs atomic structure
- Improves performance of restrained protein docking



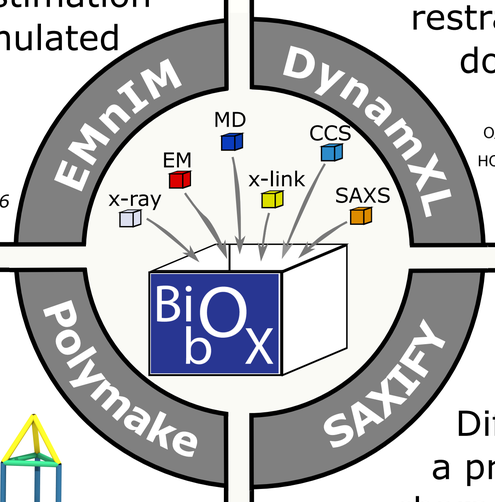
M.T.Degiacomi et al., Structure, in press
dynamxl.chem.ox.ac.uk



shortest path + side chain ensemble



shortest path alone



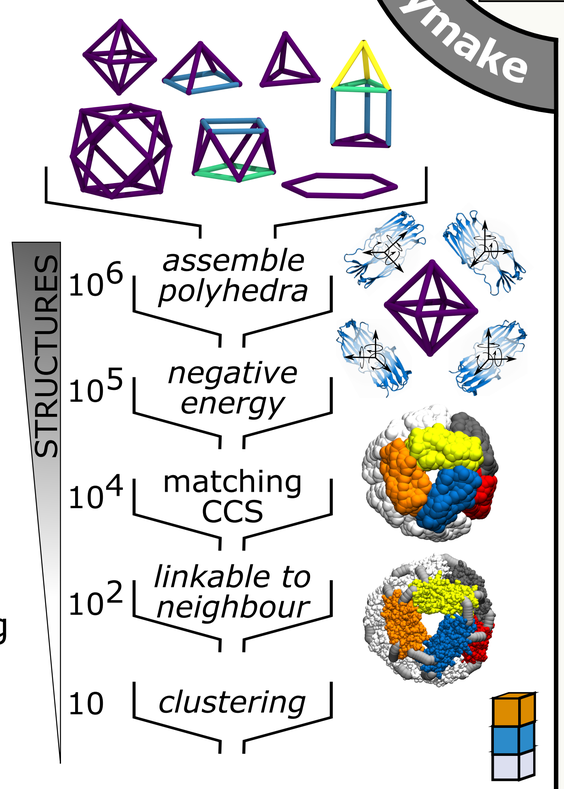
Multimers via flexible scaffolds

Method to assemble protein multimers according to 136 different, deformable topologies

Subunits can be oriented alone, in subgroups, or all together

Can fully explore multimer's conformational space, filtering models consistent with experimental data

Enables the creation of custom filtering pipelines



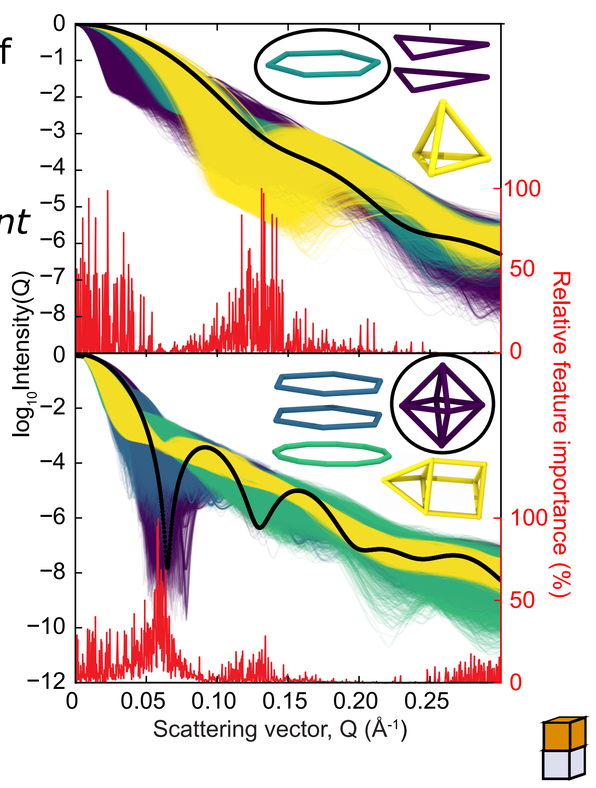
Classifying SAXS signals

Different conformations of a protein usually feature characteristic SAXS profiles

Given structural examples of different conformations, a learning algorithm can assign an experimental SAXS profile to a specific class

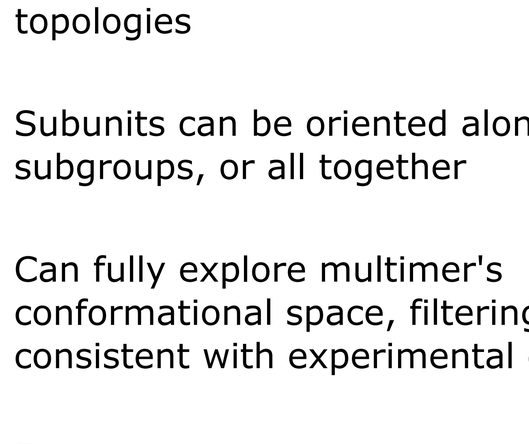
Can select the correct topology for a multimer from a list of candidates (random examples from *Polymake*)

Can discriminate between two different conformations of synaptotagmin simulated with MD



M.T.Degiacomi and J.L.P Benesch, submitted

integrative modelling of sHSP multimers



Dimeric sHSP 16.9 and 18.1 form 6mers linked by their subunits' C-ter

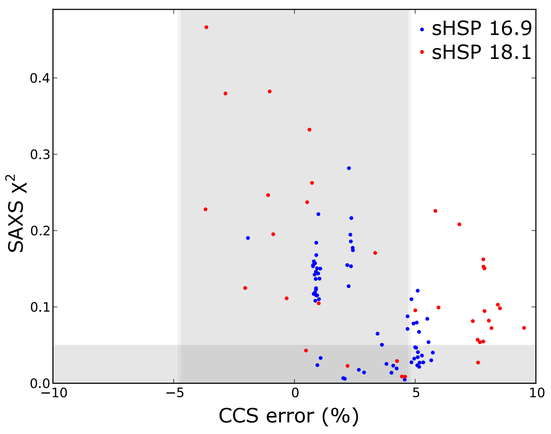
SAXIFY indicates assemblies are tetrahedral

We used *Polymake*, CCS and SAXS data to model 12mers

Some models match SAXS but not CCS and viceversa: the two techniques are complementary

11 sHSP 16.9 and 5 18.1 models match all experimental data

N-terminal region can fit in tetrahedron's centre

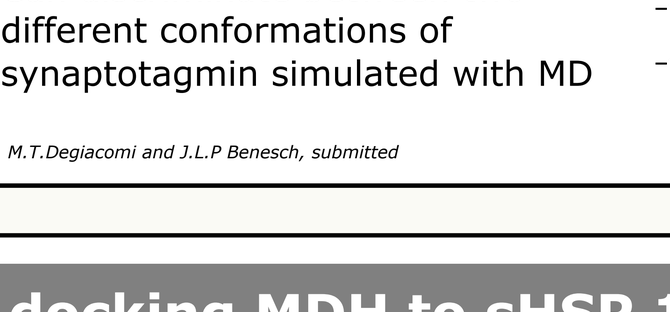


docking MDH to sHSP 18.1

sHSP 18.1 binds monomeric and dimeric Malate dehydrogenase (MDH) at heat shock temperatures

We exploit 25 cross-links to dock MDH to an ensemble of sHSP 18.1 (see left)

Docking using POW optimization engine, distances measured with *DynamXL*



MDH monomer: 10 solutions *MDH dimer: 12 solutions*
color: minimal distance of any sHSP model from any docked MDH. <3 Å in red, >10 Å in blue.

Each face of sHSP 18.1 can bind MDH. Multiple binding consistent with mass spectrometry data

Postdoc position available! Please contact Matteo Degiacomi for inquiries: matteo.t.degiacomi@durham.ac.uk