BP 18: Focus session: Integrative Structural Modeling

Modern biological questions - and the growing recognition that biomolecular dynamics are central to molecular function - demand increasingly integrated technological strategies to probe the structure and behavior of biomolecules. Yet bringing insights from diverse experimental techniques together in a rigorous, quantitative way remains a formidable challenge, requiring deep expertise across methods as well as sophisticated modeling approaches. This focus session aims to ignite collaboration at this interface. By gathering experimentalists with complementary skill sets and researchers specializing in computational and theoretical modeling, we will create a space for vibrant exchange, cross-disciplinary learning, to lead to the development of well-informed, predictive models of complex biological systems.

Organized by Richard Börner (HS Mittweida) and Sigrid Milles (FMP Berlin)

Time: Wednesday 10:30–12:45 Location: BAR/0106

Invited Talk BP 18.1 Wed 10:30 BAR/0106

Protein complex structure prediction, state-of-the-art and challenges — •Ezgi Karaca — Izmir Biomedicine and Genome Center — Dokuz Eylul University, Izmir, TR

The advent of artificial intelligence has reshaped structural biology, enabling unprecedented accuracy in protein modeling. As the assessor for CASP14 and CASP15, I witnessed remarkable progress facilitated by AlphaFold. While CASP14 participants struggled with intersubunit contacts, CASP15 saw prediction success rates rise from 31-percent to 90-percent by incorporating AlphaFold2 (AF2) into customized pipelines. This improvement was driven by curated multiple sequence alignments (MSAs), though complexes with shallow MSAs remained challenging. Addressing this, we developed MinnieFold, an efficient protocol achieving a 50-fold reduction in GPU usage while maintaining accuracy in antibody-antigen complexes. Further investigating AF's performance, we analyzed 276 human-parasite interactions across 15 species. Comparing AF2 and AlphaFold3 (AF3), we observed striking differences in predicted structures. This presentation highlights these advances, challenges, and our contributions to AI-driven structural biology.

BP 18.2 Wed 11:00 BAR/0106

Extending the Fluorophore Dye Library for FRET-guided Modeling — •Isolde Kirk[1], Felix Erichson[1], Josephine Meitzner[1], Patrick K. Quoika[2], and Richard Börner[1] — [1]Laserinstitut Hochschule Mittweida, Mittweida, Germany — [2]Technische Universität München, Munich, Germany

Integrative structural modeling combines experimental observables with computational methods to obtain realistic descriptions of biomolecular structures and their dynamics. One such approach integrates Förster Resonance Energy Transfer (FRET) and dynamic fluorescence anisotropy with molecular dynamics (MD) simulations. Ensuring comparability between experiment and simulation requires realistic modeling of fluorescent dyes in silico. In practice, existing dye libraries do not cover the full range of available fluorophores, including phosphate-backbone-linked carbocyanine (Cy) dyes. We therefore modeled and parametrized a FRET dye pair of Cy3 and 5 with a backbone-linked Cy3 fragment for MD simulations. We used Gaussian to assign partial charges with the restrained electrostatic potential method, as implemented in Antechamber, yielding several parametrization variants. The dye fragments were then incorporated into a DNA hairpin model and characterized using MD simulations with GROMACS. Depending on the parametrization, the DNA hairpin showed different structural responses, ranging from stable dye intercalation to partial helix destabilization. One parametrization showed agreement with the experimental data and therefore provides a validated extension for future FRET-guided modeling.

BP 18.3 Wed 11:15 BAR/0106

Structural dynamics and long-range interactions controlling timing of the Neurospora circadian clock — •Muriel Hartsch[1], Ida Marie Vedel[1], Kathrin Motzny[1], Michael Brunner[2], and Sigrid Milles[1] — [1]Leibniz-FMP Berlin, Robert-Roessle-Str. 10, 13125 Berlin — [2]Heidelberg University, Biochemistry Center (BZH), Im Neuenheimer Feld 328, 69120 Heidelberg

The function and maintenance of the circadian clock in Neurospora crassa are governed by a feedback loop involving both negative and positive regulatory elements, which together drive the oscillating circadian rhythm with a period of approximately 24 hours. The dimeric, intrinsically disordered protein FREQUENCY (FRQ) is a key component of the negative feedback complex and subject to post-transcriptional hyperphosphorylation by casein kinase 1a (CK1a). Phosphorylation of clock proteins is highly conserved across species, from fungi to mammals, with the human PERIOD (PER) protein being a notable example. However, the precise functions associated with hyperphosphorylation remain poorly understood. We hypothesize that time-dependent hyperphosphorylation of FRQ at multiple sites facilitates a transition from closed to open conformation, regulating interactions with its partners. Using nuclear magnetic resonance (NMR) and single-molecule fluorescence resonance energy transfer (smFRET), we investigate the conformational dynamics going along with FRQ phosphorylation by recombinant CK1a. This will allow, combined with structural modeling of the intrinsically disordered protein, to understand how phosphorylation alters the conformation and triggers a switch in FRQ.

15 min. break

Invited Talk BP 18.4 Wed 11:45 BAR/0106

From Sparse Restraints to All-Atom Models: Integrative Reconstruction of Hidden GPCR Conformations — •Matthias Elgeti — Institute for Drug Discovery, Leipzig University Medical School, Leipzig, Germany

Transmembrane proteins such as G protein coupled receptors (GPCRs) play central roles in cellular signal transduction and constitute major targets for therapeutic intervention. Their intrinsic conformational flexibility, however, leads to the coexistence of multiple interconverting states, which complicates structural characterization by high-resolution methods. Site-directed spin labeling (SDSL) EPR spectroscopy is uniquely suited to probe such flexible systems independent of molecular size, but it provides only sparse structural restraints. To bridge this gap, we integrate SDSL-EPR distance data with deep-learning-based structure prediction and molecular dynamics simulations. Using the angiotensin receptor (AT1R), a key regulator of cardiovascular function, we demonstrate how this integrative approach reveals a ligand-stabilized conformational state with distinct functional properties. We further present a computational pipeline that reconstructs corresponding all-atom models from sparse spectroscopic restraints and enables systematic exploration of hidden conformational states, providing a general framework for structure-guided analysis of signaling mechanisms and future drug discovery.

BP 18.5 Wed 12:15 BAR/0106

RNA 3D Folding Using Diffusion Models and Agentic Tree Search — •Arunodhayan Sampathkumar and Danny Kowerko — Professorship of Media Informatics, Technische Universität Chemnitz

Accurate prediction of RNA three-dimensional structure is essential for understanding RNA function and guiding RNA-based therapeutics. However, RNA folding remains challenging due to complex tertiary interactions, context-dependent base pairing and limited structural data, especially for short sequences with fewer than 80 nucleotides. To address this, we introduce a multi-model RNA structure prediction framework that combines an RNA-adapted Protenix model, a Boltz diffusion sampler and a template-based modeling (TBM) baseline together with GAN-augmented training for underrepresented short RNAs. Protenix offers accurate local geometry, Boltz supplies diverse global conformations and TBM contributes strong constraints when templates exist. All models are evaluated on the Kaggle RNA 3D public and private test sets. Protenix reaches TM-scores of 0.48/0.46, Boltz 0.41/0.40 and TBM 0.61/0.57. We further introduce an agentic tree-search ensemble that selects and refines conformations using con-

sensus scoring and RMSD-based diversity. This ensemble significantly improves performance, achieving TM-scores of 0.68 (public) and 0.63 (private). Our results demonstrate that integrating generative models, template signals and agentic search yields more accurate RNA structures and improves robustness for novel RNA families.

BP 18.6 Wed 12:30 BAR/0106

RNA adsorption in confinement, from electrostatic properties to spatial organization — •Horacio V. Guzman, Willy Menacho, and Ian Addison-Smith — Biophysics & Intelligent Matter Lab, Material Science Institute of Barcelona, CSIC, 08193 Barcelona, Spain

Electrostatic interactions are the main driving force of the RNA-protein association during viral assembly and disassembly processes. However, the properties of confined single stranded ss-RNA inside diverse proteinaceous viral geometries remain largely unexplored due to challenges associated with their high-resolution characterization. Electrostatics and mechanical properties at the RNA-proteins interface are also the origin of ss-RNA stability in confinement. Combining archetypical RNA fragments and diverse confined symmetries, we present multiple-solvent molecular models to explore the most favorable initial spatial organization as a function of the type of confinement. Our method shows that electrostatic interactions drive the 3D RNA structure path towards possible initial conformations. In particular for small RNAs, which are frequently interacting with the proteinaceous subunits during the virus assembly process, also known as packaging signals. We compared our results to X-ray structures of small plant viruses validating the most favorable initial conformations with our method and the virus folding symmetry. Our findings start a quantitative route to elucidate the electrostatic character of RNA-protein interfaces, and a complementary understanding to RNA packaging, assembly and disassembly processes.