of all cancers. pH (Low) Insertion Peptide (pHLIP) is a soluble peptide that binds and inserts into cell membranes under acidic conditions. However, developing pHLIP into a novel cancer targeting agent will require informed design of a variant that can distinguish between the pH range of 7.2 (healthy tissue) and 6.8 (most cancers). One of the key steps in pHLIP function is the process of folding into an α-helix before insertion. Partial helical formation of pHLIP under alkaline conditions has been reported in literature, and recent results from our group [1] support this claim. These observations, coupled with biophysical theory of peptide folding [2], leads to our hypothesis that the apparent pKa of insertion of pHLIP is tied to its helix-forming propensity in solution. We have tested this hypothesis through long timescale (~μs) molecular dynamics simulations of pHLIP in implicit solvent. Comparison with the insertion behavior observed in two variants of pHLIP that use non-natural amino acids [3] allows us to tie the unique properties of pHLIP to its primary amino acid sequence. Application of the Lifson-Roig model of helix-coil transition to our results yielded a complete thermodynamic description of state I of pHLIP. Collectively, our results are the first steps towards informed design of pHLIP variants with properties ideally suited to target cancer cells.

[1] Renet. al. *Proteins.*, (Submitted);
[2] Almeida et al., *BBA*, 1818:178 (2012);
[3] Onyango et al., *Angew. Chem. Intl. Ed.*, 54:3658 (2015).

Computational Methods and Bioinformatics II

2217-Pos Board B537

Extending Rule-Based Modeling to the Spatial Domain with Virtual Cell (VCELL)

James C. Schaff, Dan Vasilescu, Ion I. Moraru, Leslie M. Loew,
Michael L. Blinov.
Center for Cell Analysis and Modeling, University of Connecticut School of Medicine, Farmington, CT, USA.

When a kinetic model has to account for all the potential molecular complexes and interactions between multivalent or multistate molecules, the reaction network may be too large and complex to be manually specified. Rule-based modeling solves this problem by expressing molecular interactions in the form of reaction rules that serve as generators of reactions. Thus, the individual species and the reactions connecting them are generated automatically. Recently, we introduced rule-based modeling into the popular Virtual Cell (VCell) modeling framework (Schaff et al., 2016), even permitting manually specified reaction networks to be merged with rule-based model constituents. The same set of rules can be simulated using both network generation (BioNetGen, Faeder et al., 2009) and network-free simulation (NFSim, Emonet et al., 2011). Here we present a compartmental extension of VCell rule-based capabilities. The VCell paradigm in which every species and location has an assigned compartment, was extended to the rule-based description. Both BioNetGen and NFSim engines were modified to account for compartments. Every reactant and product pattern and every rule have their assigned locations. Moreover, to prevent involuntary translocation of species between compartments, a new feature of rule-based modeling was introduced, allowing anchoring a molecule to a given compartment. This way, any species that has this molecule as one of its molecular components, will remain in the anchored compartment. In addition to ODE, stochastic kinetic and NFSim simulators within VCell, this now permits VCell users to apply rule-based models to reaction-diffusion in complex geometries using either the VCell PDE deterministic solvers or the Smoldyn stochastic simulator. The expanded VCell framework was used to simulate several biological systems, such as membrane-bound clustering of Nephrin-Nick-NWasp proteins. (Supported by NIH grant P41 GM103313).

2218-Pos Board B538

Cloud Computing for All-To-All Protein-Protein Docking on Azure HPC

Masahito Ohue[1], Yuki Yamamoto[1], Takanori Hayashi[1], Yuri Matsuzaki[2], Yutaka Akiyama[1].
[1]Computer Science, Tokyo Institute of Technology, Tokyo, Japan, [2]ACLS, Tokyo Institute of Technology, Tokyo, Japan.

Cloud computing environments, such as Amazon AWS, Microsoft Azure, Google Cloud Platform, etc., achieve computational performance improvement remarkably in recent years, and is also useful in parallel computing (high-performance computing, HPC) fields. Cloud enables users to get thousands of CPU cores and GPU accelerators casually, and several software are used very easy by cloud images. We have transplanted original protein-protein interaction prediction (protein-protein docking) software, MEGADOCK, to the Microsoft Azure HPC environment. We have obtained strong scaling value of 84% with virtual machines of Azure A9 instance up to 1,200 CPU cores. In future, MEGADOCK will link to web interface and GUI local client on Azure cloud to be more easy-to-use for non-computer scientists.

2219-Pos Board B539

Acceleration of Cardiac Simulations for Cloud Computing Resources

Delvin Huynh, Yuanfang Xie, Daisuke Sato.
University of California Davis, Davis, CA, USA.

Computer simulation has greatly facilitated our understanding of the cardiovascular system and is boosting the development of personalized medicine for cardiac diseases. The simulation of the heart is computationally intensive due to the complexity and multi-scale essence of the cardiovascular system. Parallel computing has been one of the commonly used solutions for such large-scale simulations. More recently, rapidly growing cloud computing resources, such as Amazon Elastic Compute Cloud and Google Compute Engine, provide a new infrastructure for parallel computing. However, the high latencies of the network limit the application of cloud computing. In this study, we developed a novel algorithm to facilitate the utilization of cloud based computing resources. By exploiting the fact that the time scale of diffusion of ions is much slower than the time scale of reaction in ion channels/pumps, this algorithm improves the simulation speed by more than twice as much; the larger the latencies in networks, the faster our algorithm performs the simulations. Targeted computer models at many system scales will be drastically optimized in terms of efficiency including cardiac tissue models, models of subcellular calcium cycling, and a combination of them. Techniques developed here can be also applied for any number of other complex reaction-diffusion systems in other fields.

2220-Pos Board B540

Re-Docking by Analyzing the Profile of Protein-Protein Interaction

Yosuke Amano[1], Wataru Nemoto[1], Nobuyuki Uchikoga[2].
[1]Dept. Sci. & Eng., Tokyo Denki University, Saitama, Japan, [2]Dept. Phys., Chuo University, Tokyo, Japan.

Various proteins exert molecular functions through forming a protein complex. Elucidation of a complex structure is essential to understand its molecular functions. At present, the number of complex structures in PDB is not enough for us to understand the information coded on the protein-protein interaction network. *In silico* analyses, docking simulation has been applied to augment the lack of complex structure information. In general, software for docking simulation rotates a protein (ligand) around its interaction partner protein (receptor) to generate many complex structure candidates. Hereafter, these candidates are referred to as decoys. Similar complex structures to the native structure (near-native structures) are expected to exist among the decoys. However, in some protein pair cases, there is no near-native structure in a set of decoys because of the shortage of docking space, that is one of the important problems to be solved. This problem was addressed by expanding the docking space based on re-docking strategy [Uchikoga et al. 2013 *PLOS ONE* **8**:e69365]. Re-docking is a second round docking step after the initial docking. Then, analyses of Interaction FingerPrints (IFPs) [Uchikoga and Hirokawa 2010 *BMC Bioinformatics* **11**:236-245] have been proved to obtain near-native structures efficiently by focusing on the docking space corresponding to a decoy. Hence, we developed a software named Pftkool that is a tool for re-docking by calculating and classifying IFPs. Pftkool uses k-means algorithm to search receptor surface for target residues to perform re-docking. In general, the number of k of k-means algorithm should be specified by a user. However, our analyses show that k is automatically determined in the case of re-docking.

2221-Pos Board B541

A Docking Based Approach to Analyze Interaction Surfaces of Virus-Host Protein-Protein Interactions

Yuri Matsuzaki[1], Jaak Simm[2], Nobuyuki Uchikoga[3], Yutaka Akiyama[1].
[1]Tokyo Institute of Technology, Tokyo, Japan, [2]KU Leuven, Leuven, Belgium, [3]Chuo University, Tokyo, Japan.

Core elements of cell regulation are made up of protein-protein interaction (PPI) networks. Predicting relevant interacting partners from their tertiary structure is a challenging topic where computer science methods have potential to contribute to biology. Protein-protein rigid docking has been applied for this purpose by several projects. Docking-based approaches have advantages such that (i) they can suggest binding poses of predicted binding partners that would help understanding the interaction mechanisms, (ii) comparing docking results of both non-binders and binders can lead to understanding the specificity of PPI from structural viewpoints. However, the prediction power is limited mainly because of poor correlation between docking score and actual protein-protein binding affinity. To improve state of the art we propose a machine learning approach to improve docking-based PPI predictions by using residue profiles obtained by rigid docking. A possible application of our method is a problem in which we have one 'receptor' protein and pickup proteins that have potential to interact with it from a pool of candidate 'ligand' proteins. We define an interface fingerprint of a pair of proteins as a collection of residues of a 'receptor'

protein, with information of how often each residue are included in binding sites of high scoring docking models. We applied multiple methods of machine learning to discriminate binders and non-binders using those profiles as input. In this poster we present an evaluation of our method by applying it to proteins of Protein Docking Benchmark ver. 5.0, bacterial chemotaxis, and host-virus protein interactions.We further applied our method to analyze proposed host-virus PPI interaction surfaces by post-docking analysis to compare surfaces of PPIs between the host proteins.

2222-Pos Board B542

***In Silico* Screening for Chemical Scaffolds as Suitable Natural Inhibitors of Kinesin EG5 Divulges Morelloflavone, a Biflavonoid, as Potential Anti-cancer Compound**

Tomisin Happy Ogunwa, Takayuki Miyanishi.
School of Fisheries and Environmental Sciences, Nagasaki university, Nagasaki, Japan.

Natural products remain a source of chemical scaffold pool for drug design and development. Kinesin Eg5 has emerged as a clinical target for anticancer agents. In our search for potent natural inhibitors of kinesin Eg5, we employed *in silico* tools to screen selected diverse chemical scaffolds (compounds) that were obtained from medicinal plants. Surprisingly, the molecular interaction analysis for the selected compounds adjudged morelloflavone (a biflavonoid) as a potential ATP-noncompetitive inhibitor of kinesin Eg5 protein which occupied the putative L5/α2/α3 allosteric pocket on the protein. Compared to STLC with binding energy value −10.0 Kcal/mol, morelloflavone displayed binding energy value of −10.2 Kcal/mol and a 90 percent binding site similarity. It is also worth noting that morelloflavone was embedded within the cavity formed by amino acid residues Ile-136, Glu-116, Glu-118, Trp-127, Gly-117, Ala-133, Glu-215, Leu-214, Tyr-211 and hence, displayed a reliable tendency to block the enzymatic catalysis of kinesin Eg5 allosterically. The compound established hydrogen bonds with Glu-118 and Tyr-211 having minimum length of 2.97Å and hydrophobic interactions occurred with alkyl side chain of residues Gly-117, Glu-116, Ala-218, Ile-136, Arg-119 and Asp-130 while π-stacking interaction is observed between the aromatic ring of morelloflavone and Arg-119. These interactions anchored morelloflavone into the binding site. The results obtained in this work indicate the strong affinity and inhibitory potential of this compound on kinesin Eg5, hence lending credence to the yet untapped anticancer capacities of morelloflavone. We therefore suggest *in vitro* and *ex vivo* evaluation of this compound as anticancer agent targeting kinesin Eg5 protein.

2223-Pos Board B543

Development of Postprocessing Method of Protein-Ligand Docking using Interaction Fingerprint

Nobuaki Yasuo[1], Masakazu Sekijima[1,2].
[1]Department of Computer Science, Tokyo Institute of Technology, Tokyo, Japan, [2]Advanced Computational Drug Discovery Unit, Tokyo Institute of Technology, Tokyo, Japan.

Protein-ligand docking is an important method in Structure-based Drug Discovery [1]. Although many programs have been developed for docking [2], the accuracy is still insufficient due to the difficulty in the scoring function [3]. Interaction fingerprint is one of the solutions, which generate fingerprints of ligands using the interactions between the ligand and the protein. Interaction fingerprints use the information of known compounds so that compounds that have similar interaction to the known active ligands are expected to find through the virtual screening. However, existing interaction fingerprints such as SIFt [4] and SPLIF [5] only assess the existence or the distance of the interactions and do not consider the strength correctly. In this study, we made a new scoring function of protein-ligand docking called SIEVE-Score (Similarity of Interaction Energy VEctor-Score), which can consider the strength of each interaction explicitly. SIEVE-Score is calculated based on the similarity of the interaction energy vector, which is the list of interaction energy between the ligand and each residue of the protein. We also evaluate the accuracy of virtual screening using SIEVE-Score after the docking by Glide [6].

[1] Chiba, S., *et al*. Identification of potential inhibitors based on compound proposal contest: Tyrosine-protein kinase Yes as a target. *Scientific reports* 5:17209, 2015.
[2] Elizabeth Y., Jessica H., and Paul A. R., Improvements, trends, and new ideas in molecular docking: 2012–2013 in review. *Journal of Molecular Recognition*, 28(10):581–604, 2015.
[3] Yan L., Li H., Zhihai L., and Renxiao W., Comparative assessment of scoring functions on an updated benchmark: 2. evaluation methods and general results. *Journal of Chemical Information and Modeling*, 54(6):1717–1736, 2014.
[4] Zhan D., Claudio C., and Juswinder S., Structural interaction fingerprint (SIFt): a novel method for analyzing three-dimensional protein-ligand binding interactions. *Journal of Medicinal Chemistry*, 47(2):337–344, 2004.
[5] Da C., and Kireev D., Structural protein–ligand interaction fingerprints (SPLIF) for structure- based virtual screening: Method and benchmark study. *Journal of Chemical Information and Modeling*, 54(9):2555–2561, 2014.
[6] Richard A. F., *et al*., Glide: a new approach for rapid, accurate docking and scoring. 1. method and assessment of docking accuracy. *Journal of Medicinal Chemistry*, 47(7):1739–1749, 2004.

Optical Spectroscopy: CD, UV-VIS, Vibrational, Fluorescence I

2224-Pos Board B544

New Thiol-Reactive Eu-Complex for Distance Measurements by LRET

Felix Faschinger[1], Mirjam Zimmermann[1], Guenther Knoer[2], Hermann Gruber[1].
[1]Institute of Biophysics, Johannes Kepler University, Linz, Austria, [2]Institute of Inorganic Chemistry, Johannes Kepler University, Linz, Austria.

Crystallography and NMR spectroscopy are ideally suited to resolve the 3D structures of biomolecules but the material and time demand for each structure is high. Fluorescence resonance energy transfer (FRET) provides less structural information but is better suited to study conformational changes and structure-function relationships by screening a large number of mutants or experimental conditions. Moreover, FRET allows for real time monitoring of conformational changes induced by specific ligands. Usually, FRET yields only a crude estimate of the donor-acceptor distance, due to the fact that the relative orientation of donor and acceptor are rarely known. Luminescence resonance energy transfer (LRET) is much better suited for distance measurements because the orientation factor (and thus the Forster distance) are known, and because energy transfer is measured by a change of lifetime, rather than of signal intensity. In LRET-experiments the ideal donors are highly stable Eu/Tb-complexes, with a single lifetime that is not influenced by attaching the complex to a biomolecule. Several terpyridine-based Eu-complexes described in literature have promising properties concerning uniform lifetimes after protein labeling but all described complexes have rather long linkers which prevent accurate distance measurements. In this study, a new terpyridine-based Eu-complex with maleimide very close to the metal ion center was synthesized and found to have ideal properties for distance measurement by LRET: After linking to the single cysteine of BSA, the complex showed a quantum yield of 30%, a single lifetime of 1.2 ms, comparable to the best known Eu-complexes, and the lifetime was unaffected by phosphate or EDTA. In conclusion, this new Eu complex appears ideally suited for reliable measurement of intra- or intermolecular distances.

The work was supported by grant W0125 of the Austrian science fund (FWF)

2225-Pos Board B545

A Novel FRET Technique to Characterize the Oligomerization State of Protein-Protein Interactions

Philipp J. Heckmeier, Mark G. Teese, Dieter Langosch.
Lehrstuhl für Chemie der Biopolymere, Technische Universität München, Freising, Germany.

Protein-protein interactions are the fundamental driving force of numerous cellular processes and cell signaling pathways. Characterizing whether proteins interact as dimers, trimers, or higher oligomers is essential to understanding these interactions. Several microscopy and advanced imaging techniques relying on Förster resonance energy transfer (FRET) between identical fluorophores (homo-FRET) have been developed to estimate protein stoichiometry. The increased FRET in oligomers is detected by measuring depolarization or emission time. Homo-FRET methods have a strong advantage in requiring only a single fluorophore, greatly simplifying sample preparation in comparison to conventional hetero-FRET methods. However, most homo-FRET methods require sophisticated imaging equipment, and both theoretical models and applications have been restricted to the study of membrane-bound proteins. Using a simple bulk homo-FRET and laser photobleaching approach, we demonstrate the feasibility of characterizing the oligomerization state of an interacting protein in-vitro. To simulate oligomers in a proof of concept, we constructed an extensive repertoire of fusion proteins with 1-6 consecutive green fluorescent protein (GFP) domains. We show how the resulting homo-FRET (measurable via steady-state anisotropy or fluorescence polarization) is proportional to the oligomerization state of proximal GFP domains. For the first time, this is demonstrated with soluble proteins. In both membrane and soluble proteins, oligomerization increases FRET and therefore anisotropy. However for soluble proteins oligomerization also slows fluorophore rotation, leading to a size-dependent decrease in anisotropy. Through gradual photobleaching of