1P477

Analysis of transmembrane helical regions of G-protein-coupled receptors (GPCRs) for a novel prediction system

○Toshiyuki TSUJI[1,2], Shigeki MITAKU[1,2]
[1]Dept. of Applied Physics, Graduate School of Engineering Nagoya Univ., [2]21 Century COE Program "Frontiers of Computational Science" Graduate School of Engineering Nagoya Univ.

The G-protein-coupled receptors (GPCRs) are involved in the initial step of the signal transduction pathways, having seven transmembrane helices and extracellular N terminus. In spite of the common features, sequence motifs for the superfamily of GPCRs are not found. The difference in sequences of GPCRs is partly due to many kinds of different ligands that activate the receptors. Another difficulty in the prediction of GPCRs is the existence of the hydrophilic transmembrane helices which can hardly predicted, and the discrimination of GPCRs from other types of membrane proteins by the number of transmembrane regions fail in many cases. Thus, GPCRs can't be detected by ordinary sequence similarity searches nor membrane protein prediction systems. In this study, we analyzed physicochemical properties of several transmembrane regions around the transmembrane helices: the hydrophobicity, amphiphilicity, and the densities of characteristic amino acid residues such as proline. It was found that the transmembrane helices have common physicochemical properties according to the position in the sequences. Using the individuality of those features of seven transmembrane helices, we are developing a novel prediction system for GPCRs. We will report the performances of the system, SOSUIGPCR, in the poster.

1P478

Sequence and structure features of GPCRs, which are effective to determine the GPCR and G-protein binding selectivity

○Makiko Suwa[1], Takahiko Muramatsu[2]
[1]Computational Biology Research Center, National Institute of Advanced Industrial Science and Technology, [2]Nara Institute of Science and Technology

G-Protein-Coupled Receptors (GPCRs) are the major membrane proteins which play a role of signaling to the inner cell. An external ligand stimulus to a GPCR induces the coupling with G-proteins, (Gi/o, Gq/11, Gs and G12/13), followed by different kinds of signal transduction. We assume that whole structural segments of ligand, GPCR and G-protein are effective to determine GPCR and G-protein coupling. Based on this assumption we tried to identify several descriptors relating to G-protein selection, from ligand, GPCRs, G-protein complex. First, we identified several functional residue sites in GPCRs that are related to coupling selectivity, by mapping known GPCR sequences onto the structure of rhodopsin. These residues are located mainly at the intracellular loops, and found that the occurrence of positively/negatively charged amino acids of the characteristic residues varies depending on the G-protein selectivity. It is interesting that some characteristic residues are located at or near the extracellular terminus of transmembrane helices, which is far from the GPCR/G-protein binding interface. We have identified also another parameters effective to G-protein selection, such as, ligand size, length of C-terminal loop and I3 loop. etc. By combining these features as feature vectors, we describe a novel program, that predicts GPCR and G-protein coupling selectivity based on support vector machine (SVM). Applying this system to known GPCR sequences each binding G-protein is predicted with high sensitivity and specificity on average.

1P479

A possible effect of alternative splicing on multiple regions of a single protein

○Masafumi Shionyu[1], Kei Yura[2], Mitiko Go[1,3]
[1]Fac. Bio-Sci., Nagahama Inst. Bio-Sci. Tech., [2]Quantum Bioinfo., JAEA, [3]Ochanomizu Univ.

Alternative splicing (AS) is a mechanism to increase the number of transcripts from a single gene by altering the usage of exons during mRNA splicing process. In the AS process, not only a single exon, but multiple exons are a target of alternative usage for generating a mature mRNA. For example, *Drosophila* Dscam gene has four clusters of exons each of which provides one exon from many to a mature mRNA. Hence, theoretically Dscam gene can generate 38,000 variant transcripts. However there could be a correlation in usage of exons from different clusters and the gene cannot bear such huge number of mature mRNAs. Most of computational studies of AS have used the EST sequences to detect AS events, and EST analyses generally miss correlation of usage in different exons. To detect simultaneous alteration in exon usage by AS, we have developed a pipeline that can systematically detect alternatively spliced exons using genomic and full-length transcript sequences. The pipeline further assigns AS regions, which are defined as protein regions modified by alternatively spliced exon, onto protein three-dimensional (3D) structures. We analyzed alteration of multiple AS regions, a simultaneous alteration of AS regions, using human data and found that about 7,200 isoforms out of 3,923 genes have multiple AS regions. We, then, assessed the spatial relationship between multiple AS regions on protein 3D structure. We will discuss the impact of multiple AS regions on protein function and stability.

1P480

Database analysis of S•••O and S•••N interactions for phospholipase A2

○Michio Iwaoka, Noriyoshi Isozumi
Department of Chemistry, School of Science, Tokai University

The roles of sulfur atoms in proteins include the effects to stabilize the structure thermodynamically by formation of disulfide bonds and to comprise the active sites of several redox enzymes. However, in a view of structural biology the sulfur atoms have been considered to be merely hydrophobic groups in protein structures. We have recently found that many sulfur atoms in proteins specifically form weak nonbonded interactions (i.e., S•••X interactions) with nearby oxygen, nitrogen, and sulfur atoms. In this paper, possible roles of S•••X interactions in protein functions and evolution were analyzed for phospholipase A2 (PLA2) [1], for which a lot of structures have been deposited in Protein Data Bank (PDB). PLA2 is an enzyme present in a wide range of organisms, such as mammals, and is also know as a component of snake venom. The globular enzyme consists of about 130 amino acid residues, including seven disulfide (SS) linkages and one methionine residue (Met). We collected the high-resolution structure data from PDB and searched for close S•••X contacts extensively. For bovine PLA2, four S•••O and one S•••N interactions were characterized. For snake PLA2, additional three S•••O interactions were identified. Most of these interactions were found to be present near the active site and make clusters in the phylogenetic tree. The results suggested that S•••X interactions would have some implications not only for protein structures but also for protein functions and evolution.

[1] M. Iwaoka, N. Isozumi, BIOPHYSICS, 2, 23 (2006).