s1.m5.p9 **Roles of the base-stacked XYYX in forming multiple associations of the base-intercalated duplexes.** Jiro Kondo, Wataru Adachi, Shun-ichi Umeda, Kazuhiro Fujita, Tomoko Sunami and Akio Takénaka, *Graduate School of Bioscience and Biotechnology, Tokyo Institute of Technology, 4259 Nagatsuta, Midori-ku, Yokohama, 226-8501, Japan. E-mail: atakenak@bio.titech.ac.jp*

Keywords: DNA; Base-intercalated duplex; DNA multiplex assembly

To understand the biological significance of single-stranded DNAs, it is necessary to reveal structural versatility of DNA containing non-complementary sequences. It has been found that DNA fragments containing the sequence d(gcGAAAgc) prefer to adopt a base-intercalated duplex, and further associated to form hexaplex- and octaplex-assemblies [1,2]. To investigate effects of the central sequence, crystal structures of several mutants with the sequence d(gcGXYAgc) (X = A or G, Y = A, G, T or C) have been determined by X-ray crystallography. In the base-intercalated duplex of d(gcGXYAgc), Watson-Crick G:C base pairs followed by a sheared G:A base pair form the stem regions at both ends of duplexes. To expose the central X and Y residues, stacked alternatively between the two strands, the sheared G:A base pair formations are essential to make the two phosphate backbones closer and to stabilize the central base-stacked XYYX motif. Through these motifs, the base-intercalated duplexes are further associated to form multiple assemblies. In total three kinds of different assemblies have been found [1-4]. In the presence of hexamine cobalt cation, the base-intercalated duplexes prefer to form a hexa-assembly. In the case of calcium ion, similar hexa-assembly is formed. On the other hand, assembly formation is changed in the presence of potassium ion. In relatively lower concentration of potassium ion, four base-intercalated duplexes of d(gcGAGAgc) are associated around the potassium ion to form an octaplex, in which the eight G_5 residues form two G-quartets. In the case of that the central residue is A, the eight A_5 residues are bundled through water-mediated hydrogen bonds. In relatively higher potassium concentration, however, the octaplex is split into just two halves of quadruplexes. A potassium ion is bound on the split surface and separates the split quadruplexes at a distance 11~12 Å. In the central part of each quadruplex, the four G5 residues form two G-duets and bound to the central potassium cation.

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s1.m5.p10 **Towards time-resolved studies with Phycoerythrocyanin (PEC)-crystals from Mastigocladus laminosus.** Angela Krasselt[1], Marius Schmidt[1], Wolfgang Reuter[2], [1]*Physikdepartment E17, TU München, Germany;* [2] *MPI für Biochemie, Martinsried, Germany*. *E-mail: angela@hexa.physik.tu-muenchen.de*

Keywords: Laue crystallography; Phycoerythrocyanin; Phycobilisomes

Introduction: Phycobilisomes (PBS) are peripheral membrane complexes that efficiently harvest light energy and transfer the energy to photosynthetic reaction centers. They are the major photosynthetic antenna complex of cyanobacteria and red algae. All PBS have chromoproteins (phycobiliproteins). Phycobiliprotein colors are a consequence of light absorption by linear tetrapyrrole chromophores that covalently associate with the apoproteins. Phycobiliproteins are composed of α- and β-subunits associated to heterodimers that aggregate into trimers $(\alpha\beta)_3$ and hexamers $(\alpha\beta)_6$. PEC is a protein with bright pink color, which was isolated from the thermophilic cyanobacterium *M. laminosus*. Up to now the photoactivity has not been investigated systematically in the crystal. However, in the isolated α-subunit a Z/E isomerization of the chromophore has been observed in the dimer [1]. Isomerization as well as following re-isomerization to the dark state may be so fast that is it has not been observed with the slow methods employed so far.

Goal: In a first step the crystallization conditions must be improved. In addition a high resolution structure of the PEC dark state must be collected by monochromatic radiation to a resolution better than 2.0 Å at room temperature. The acquired structure will serve as a reference. In the next phase Laue data sets will be collected. Finally time-resolved crystallographic experiments will follow.

Crystallization & x-ray: From PEC well scattering hexagonal prisms of space group $P6_3$ can be grown [2]. We improved the protein purification and crystallization conditions. We obtained PEC-crystals which are sufficiently small (around 300 μm) and stable for measurements at room temperature. Most importantly the mosaicity is in the range of 0.15 deg, which is very suitable for Laue-diffraction. The structure of PEC was initially determined 1990 by Duerring et al. [3]. Our first data were collected at room temperature on a home source. The structure analysis and the refinement has been performed to 3.0 Å resolution. The PEC model was refined by using CNS and O for model building to a crystallographic R-factor of 18 %. At the Synchrotron the resolution most likely can be substantially improved. The possibility exist that the hexagonal symmetry is a pseudo-symmetry and the structure of the linker is unintentionally averaged by the subsequent threefold application of the crystallographic symmetry operation. Structural analysis of the linkerprotein would be a highly wellcome byproduct. Preliminary Laue data and monochromatic data sets were collected at the BioCars 14-ID beamline at the Advanced Photon Source (APS, Argonne, USA).

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