

s1.m7.p15 **Structural study of a tumor-associated human DEAD-box RNA helicase, rck/p54.** Tsutomu Matsui,^a Keita Hogetsu,^a Yukihiro Akao,^b Takao Sato,^a Takashi Kumasaka^a and Nobuo Tanaka^a, ^aGraduate School of Bioscience and Biotechnology, Tokyo Institute of Technology, 4259 Nagatsuta, Midori-ku, Yokohama, 226-8501 Japan, Japan, and ^bDepartment of Genetic Diagnosis, Gifu International Institute of Biotechnology, 2193-128 Mitake, Kani-gun, Gifu 505-0116, Japan. E-mail: ntanaka@bio.titech.ac.jp

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The structural changes of RNA play a very important role that is required by all living organisms. DEAD-box RNA helicase family is considered to disrupt RNA structures and facilitate their rearrangement by unwinding short stretches of duplex RNA in an ATP-dependent manner. rck/p54 from human consists of 472 amino acid residues and is a member of DEAD-box RNA helicase family. In previous study, rck/p54 was thought to contribute in cell proliferation and/or in carcinogenesis[1].

In the present study, the limited proteolysis experiments of rck/p54 were used to truncate the N-terminal domain (1-288) of rck/p54, thereby succeeding in the crystallization and of Nc-rck/p54, i.e., the N-terminal core domain (70-288) of rck/p54[2]. The structure of Nc-rck/p54 was solved at 2.0 Å and is the first structure in human DEAD-box helicase. To understand the mechanism of rck/p54, the biological, dynamic light scattering and electron-microscopic analyses with their substrates were carried out. These studies have revealed the reaction using conformational change and the substrate recognition. Dynamic light scattering experiment showed that AMP-PNP, nonhydrolysis ATP analog, was enough to have the conformational change from open to close conformation although DEAD-box RNA helicase could bind ATP and RNA between two domains. The ATPase assay of rck/p54 showed that the hydrolysis activity of rck/p54 seemed to be influenced by the amount of duplex regions in RNA. Crystal structure of Nc-rck/p54 have provided further knowledge that Q motif and GG motif of DEAD box RNA helicase could play an important role in substrate recognitions of ATP and RNA, respectively[3].

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- [2] Matsui, T., Hogetsu, K., Y. Akao, Tanaka, M., Sato, T., Kumasaka, T., and Tanaka, N. (2004). Crystallization and X-ray analysis of the N-terminal core domain of a tumor-associated human DEAD-box RNA helicase, rck/p54. *Acta Cryst.* **D60**, 156-159
- [3] Hogetsu, K., Matsui, T., Y. Akao, Tanaka, M., Sato, T., Kumasaka, T., and Tanaka, N. Crystal Structure of rck/p54 N-terminal core domain: A tumor-associated human DEAD-box RNA helicase. *in preparations*.

s1.m7.p16 **Crystal and Solution Structure of the Putative Antiterminator Protein Rv1626 from *Mycobacterium tuberculosis*.** J. P. Morth^a, V. Feng^b, L. J. Perry^b, Dmitri I. Svergun^c and P. A. Tucker^a. ^aEMBL Hamburg Outstation, c/o DESY, Notkestrasse 85, Germany. ^bUCLA-DOE Institute for Genomics and Proteomics, University of California, Los Angeles 90095, USA. ^cInstitute of Crystallography, Russian Academy of Sciences, Leninsky pr. 59, 117333 Moscow, Russia. E-mail: premo@embl-hamburg.de

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The project is a part of the *Mycobacterium tuberculosis* structural genomics project. Rv1626 was initially identified by sequence comparison as a response regulator of a two component system without an apparent histidine kinase associated with it. Just recently a new domain type was discovered by sequence comparison, the domain was found in (A)miR and (N)asR (T)ranscription (A)ntitermination (R)egulators (ANTAR) [1]. Rv1626 contain a C-terminal ANTAR domain in addition to the N-terminal receiver domain, common to response regulators.

The untagged Rv1626 was purified from an *E. coli* expression strain, and crystallised using the microbatch method. Heavy atom derivatation was done by a quick soak using sodium iodide. The structure was solved by single anomalous scattering (SAS) from the anomalous signal of iodide. The structure shows high structural similarity to an antiterminator protein AmiR, except for a kink region in the linker helix between the N- and C-terminal domains of Rv1626, which breaks up the proposed coiled coil found present in the AmiR dimer. To analyse the oligomeric state in solution small angle X-ray scattering (SAXS) was performed. The molecule in solution was found to be monomeric as it is in the crystal, but in solution it undergoes a conformational change which is triggered by changes in ionic strength.

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