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*], [a]*Graduate School of Bioscience and Biotechnology, Tokyo Institute of Technology, 4259 Nagatsuta, Midori-ku, Yokohama, 226-8501 Japan, Japan, and* [b]*Department of Genetic Diagnosis, Gifu International Institute of Biotechnology, 2193-128 Mitake, kani-gun, gifu 505-0116,Japan, 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 am 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*. *D***60***, 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.*