

**P04.07.249***Acta Cryst.* (2008). A64, C309**Crystal structure of RNA aptamer in complex with human immunoglobulin G**

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Aptamers are single-stranded nucleic acids that bind to molecular targets, including proteins, with high affinity and specificity. RNA aptamer can be discovered in vitro selection process on the basis on their high affinity for the targets. Recently, various applications of RNA aptamers have been introduced, not only to determine a drug target involved in disease pathology, but also to develop therapeutic agents. We have demonstrated the ability of RNA aptamer to bind to human immunoglobulin G (IgG) with high affinity and specificity. The aptamer was shown to bind to the Fc domain of human IgG, but not to other IgG's, with high affinity. The aptamer was observed to compete with protein A, but not with the Fc receptor, for IgG binding. Here, we have determined the crystal structure of the aptamer in complex with human IgG. The electron density has clearly proved two aptamers bound to dimeric IgG. The tertiary structures of the recognition sites on the Fc domain differ significantly between human IgG and other species of IgGs. The structure also explains high specificity and affinity of the selected aptamer to human IgG

Keywords: crystallography of protein RNA, immunoglobulins, RNA-protein interactions

**P04.07.250***Acta Cryst.* (2008). A64, C309**Structural studies of human RIG-I in complex with double-stranded RNA**

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Retinoic acid inducible gene-I (RIG-I) is a critical component of innate immune system that is responsible for the detection and elimination of invading viruses. RIG-I like Toll-like receptor 3 detects viral RNAs inside the cell and initiates downstream signaling to activate IRF-3 and NF-kB genes resulting in the production of Type-I interferon. RIG-I and related viral RNA recognition proteins such as MDA5 and LGP2 are located in the cytoplasm, while Toll-like receptor 3 resides in the endosome. RIG-I is composed of N-terminal CARD domain for signaling via the adaptor molecule IPS-1 and C-terminal helicase domain for RNA recognition. The C-terminal helicase domain recognizes viral double-stranded RNA or single-stranded RNA with 5'-triphosphate group. The helicase activity of RIG-I is required to promote conformational changes of the CARD domain so that it can interact with IPS-1. To study the mechanism of viral dsRNA recognition by RIG-I, the structure of RIG-I C-terminal domain in complex with dsRNA is being studied.

Selenomethionine incorporated RIG-I was expressed using E.coli and purified for crystallization. RIG-I:dsRNA binding was studied by EMSA method to investigate the RIG-I:dsRNA binding property in vitro. The X-ray data was collected from RIG-I:dsRNA complex crystals to 2.6 Å using synchrotron radiation. Structural studies of RIG-I(helicase domain):dsRNA complex will reveal how RIG-I can recognize viral RNA and discriminate it from self RNA and show the mechanism of RNA-induced conformational changes.

Keywords: RIGI, virus, immune system

**P04.07.251***Acta Cryst.* (2008). A64, C309**Structural requirements for recognition and nuclear import of spliceosomal UsnRNPs**

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The biogenesis of nuclear UsnRNPs requires a cytoplasmic maturation step. In the cytoplasm UsnRNAs and sm-proteins form a core UsnRNP, which is a prerequisite for dimethylation of the m7G-cap of the UsnRNA to a m3G-cap by tri-methyl-guanosine-synthase (TGS1). The nuclear import of the UsnRNPs is mediated by importin $\beta$  (Imp $\beta$ ) and snurportin1 (SPN1), that specifically recognises m3G-cap. In contrast to any other characterized active nuclear import the Imp $\beta$ /SPN1/UsnRNP complex does not require RanGTP for the terminal release from the nuclear pore complex (NPC). We have determined the crystal structure of the human TGS1- methyltransferase domain, which consists of the canonical catalytic domain and an additional N-terminal subdomain that is strictly required for enzymatic activity. A m7GTP molecule, representing the minimal substrate, and S-adenosyl-homocysteine are bound in deep clefts of the TGS1 surface an are held in close proximity to each other. We also have solved the crystal structures of the C-terminal domain of SPN1 [1], revealing the structural basis for the specific recognition of the m3G cap, and of the N-terminal domain of SPN1 bound to the nuclear import receptor Imp $\beta$  [2]. The crystal structure of Imp $\beta$  in complex with the N-terminal domain of SPN1 reveals that Imp $\beta$  adopts an open conformation, which is unique for a functional Imp $\beta$ /cargo complex and it resembles the conformation of the Imp $\beta$ /RanGTP complex. As binding of RanGTP to Imp $\beta$  usually triggers the release of import complexes from the NPC, we propose that by mimicking a conformation similar to Imp $\beta$ /RanGTP the independent dissociation of Imp $\beta$ /SPN1 from the NPC is energetically aided.

[1] Strasser et al., EMBOJ 24, 2235-2243, 2005.

[2] Wohlwend et al., JMB 374, 1129-1138, 2007.

Keywords: transport, RNA-protein interaction, methylase

**P04.07.252***Acta Cryst.* (2008). A64, C309-310**Crystallographic studies on molecular motors and switches of the spliceosome**

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