The Crystal Structures of the Pseudouridine Synthases RluC and RluD $% \mathcal{A} = \mathcal{A} = \mathcal{A} + \mathcal{A}$

Kenji Mizutani^a, Yoshitaka Machida^a, Kanako Sugiyama^a, Satoru Unzai^a, Sam-Yong Park^a, Jeremy R. H. Tame^a, ^aProtein Design Laboratory, Yokohama City University, Suehiro 1-7-29, Tsurumi, Yokohama 230-0045, Japan. E-mail: mizutani@tsurumi.yokohama-cu.ac.jp

The most frequent modification of RNA, the conversion of uridine bases to pseudouridines, is found in all living organisms and often in highly conserved locations in ribosomal and transfer RNA.

RluC and RluD are homologous enzymes which each convert three specific uridine bases in *Escherichia coli* ribosomal 23S RNA to pseudouridine: bases 955, 2504, and 2580 in the case of RluC and 1911, 1915, and 1917 in the case of RluD. Both have an N-terminal S4 RNA binding domain. While the loss of RluC has little phenotypic effect, loss of RluD results in a much reduced growth rate. We have determined the crystal structures of the catalytic domain of RluC, and full-length RluD. The S4 domain of RluD appears to be highly flexible or unfolded and is completely invisible in the electron density map. Despite the conserved topology shared by the two proteins, the surface shape and charge distribution are very different. The models suggest significant differences in substrate binding by different pseudouridine synthases. [1]

[1] Mizutani K., Machida Y., Unzai S., Park S.-Y., Tame J.R.H., *Biochemistry*, 2004, **43**, 4454-4463.

Keywords: RNA-binding proteins, structure, protein crystallography