Crystallographic and Functional Studies of Nip7 a Conserved Protein Involved in pre-rRNA Processing

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Ribosome biogenesis requires the coordinate function of a large number of factors including endo- and exonucleases, RNA helicases, rRNA modifying enzymes and snoRNPs. Nip7p function was inferred from studies using *Saccharomyces cerevisiae* conditional strains. These studies revealed that Nip7p is required for 27S pre-rRNA processing and 60S ribosome biogenesis. In addition, Nip7p interacts with Rrp43p, a component of the exosome complex, and with the nucleolar proteins Nop8p and Nop53p. Highly conserved Nip7p homologues are found in all eukaryotes and putative homologues are also found in *Archaea*. The C-terminal half of the protein contains a conserved domain (named PUA, after pseudo-uridine archeosine synthetase). This domain is found in several other RNA modifying enzymes. Functional analysis by means of primer extension revealed that both Nip7p and Rrp43p deficiency leads to similar defects in pre-rRNA processing. For structural studies, we have cloned the *Pyrococcus abyssi* Nip7p homologue (PaNip7) and produced the recombinant protein in *E. coli*. Following induction, PaNip7 was purified and submitted to crystallization trials. X-ray diffraction data were collected using synchrotron radiation from native crystals and an iodide derivative. PaNip7 crystal structure was solved using the SIRAS method and refined at 1.8 Å resolution. 3D structure shows a two alpha-beta domain protein. Structural and functional analysis will be presented.

Keywords: Nip7p, 3D structure, rRNA processing