Molecular-genetic analysis of the structural determinants and primary functions of eukaryotic ribosomal proteins L16 and L19 in *Saccharomyces cerevisiae*

Ana Jiménez López, Álvaro Gil Hernández, Rafael Giraldo Suárez, Mercedes Tamame González
IBFG CSIC/USAL, Salamanca, ES, CIB, CSIC, Madrid, ES

The way in which protein and RNA macromolecules perform their roles in the eukaryotic ribosomal complex can be investigated using the genetic amenable organims *Saccharomyces cerevisiae*. The eukaryotic ribosome is larger and more complex than its prokaryotic counterpart, containing additional expansion segments of rRNA (ES), novel r-proteins (Rp) and Rp-extensions that likely correlate with the higher complexity and regulation of eukaryotic translation. The recently resolved crystal structures of the eukaryotic ribosome predict new molecular interactions amongst Rp and ES, the functional significance of which is unknown.

Mutations altering the dosage, structural integrity or on-time assembly of Rps into pre-ribosomal particles impair specific steps of the pre-rRNA processing pathway, r-subunit biogenesis or 80S function, and are cause of ribosome heterogeneity, leading to changes in the patterns of protein synthesis, cell growth and differentiation. Moreover, haploinsufficiency or Rp-mutation is associated with tumour predisposition in many organisms, and with human “ ribosomopathies” .

To unveil the primary roles in ribogenesis and translation of the essential L16 and L19 proteins of the 60S r-subunit, we generated random mutations in one of the two corresponding gene-paralogues in S. cerevisiae and selected partial loss-of-function and lethal mutants. L19 is predicted to map at the polypeptide exit tunnel of the 60S, with its α-helix at the C-terminal domain contacting the ES6 of 18S rRNA as part of the new eukaryotic intersubunit bridge eB12. L16 contacts rRNAs 25S and 5.8S and maps near to the ribosomal stalk, in a region interacting with translation initiation factors. Molecular and cellular analysis of the L16 and L19 mutant phenotypes, along with bioinformatic predictions, allows correlation of defects in vivo with alterations of molecular interactions predicted from the yeast ribosome crystal structures. Data will be presented on the location of a set of L16 and L19 mutations, the pre-rRNA processing and translational defects of the mutants, and the impact of mutations on the GCN4 specific mechanism of translational regulation.