

A CONNECTION BETWEEN TRANSLATION AND SECRETION IS REVEALED BY *TIF51A* AND *YPT1* GENETIC INTERACTIONS

Frigieri, M.C.; João Luiz, M.V.S.; Zanelli, C.F. and Valentini, S.R.

Departamento de Ciências Biológicas, Faculdade de Ciências Farmacêuticas,
UNESP, Araraquara, Brazil

The translation initiation factor 5A (eIF5A) is essential and the only cellular protein that contains the amino acid residue hypusine. eIF5A has been implicated in many cellular processes, but its precise biological function is not clear. Recently, eIF5A was shown to be directly involved with the translational machinery. A screen for synthetic lethal mutations was carried out with the *tif51A-3* mutant and revealed a mutation (G80D) in the essential gene *YPT1*. This gene encodes a small GTPase involved with vesicular trafficking between the endoplasmic reticulum and the Golgi. In order to assess the functionality of the secretory pathway, we investigated the maturation of the vacuolar glycoprotein carboxypeptidase Y (CPY) and verified that *TIF51A* mutants do not accumulate precursor forms of CPY at the nonpermissive temperature (37°C). Further, to verify the Ypt1 and eIF5A localization pattern, yeast extracts were separated into soluble and microsomal fractions. We revealed that Ypt1 and eIF5A are both associated with microsomal membranes and also present in the soluble fraction. Microsomal membranes were then treated with EDTA or RNase to dissociate ribosomes and we demonstrate that eIF5A association with microsomal membranes is dependent on ribosome binding. Finally, we revealed that *YPT1* mutants are sensitive to paromomycin, a protein synthesis inhibitor. Future studies may clarify the connection between translation and secretion.

Supported by FAPESP