STRUCTURAL BIOLOGY REVEALS A NEW PROTEIN FAMILY FROM *S.CEREVISIAE* WITH A NOVEL FOLD AND IMPLICATED IN THE METABOLISM CONTROL AND DRUG RESISTANCE

Domitrovic, T.[1]; Freire, J.G[1]; Masuda, C.A[2], Almeida, M. S.[2]; Kozlov, G.[3]; Gehring, K. [3]; Kurtenbach, E.[1]

[1]Instituto de Biofísica CCF, [2]Instituto de Bioquímica Médica, UFRJ, Rio de Janeiro, Brazil, [3]McGill University, Montreal, Canada

We have undertaken a small-scale structural genome project focusing on *S. cerevisiae* ORFs without characterized functional motifs or known primary sequence homologs. The cloning, expression and purification screening of 9 targets sequences, led to the determination of the crystal structure of Yer067w by Multiple Anomalous Diffraction at 1.7 Å resolution. This 20 KDa protein present an alpha-beta fold where the 7-stranded beta-sheet is backed by 4 alpha-helices on one side. Interestingly, a structure-based search using the servers Secondary Structure Matching and DALI retrieved only proteins with low or insignificant superposition scores, indicating that Yer067w represents a novel fold superfamily. The phylogenetic analysis of Yer067w primary sequence homologs showed that this protein belongs to a well-conserved family exclusive to Ascomycetes. To further understand Yer067w role we have searched for functional hints using yeast strains deleted for this gene and its paralog *YIL057C*. Microarray analysis of Δyer067w revealed important modifications in expression of genes related to oxidative phosphorylation, amino acids and lipid metabolism. In a screening for phenotypes, we verified that all mutants presented growth deficiencies in non fermentative carbon sources and Western bolt analysis showed that the presence of both proteins are tightly linked to growth on respiratory substrates or low nutrient conditions, suggesting that both proteins are important to the metabolism in glucose free media. Furthermore, Yer067w mutants revealed an antifungal drug resistance phenotype, presenting an increment of 2 times in the MIC for Nystatin and anphotericin B. This work highlights the importance of functional characterization of unknown ORFs for the comprehension of yeast cells metabolism and for uncover new regulatory elements. Key words: structural biology, crystal structure, new fold. Support: FAPERJ, CNPq, CIHR