**Employing the redoxomic shotgun strategy OcSILAC to study a highly oxidized yeast model**

Shakir Shakir1, Giovanni Chiappetta1, Alise Ponsero2, Iman Haddad1, Michel Toledano2, Joëlle Vinh1

*1.* *Biological Mass Spectrometry and Proteomics, ESPCI-ParisTech, PSL Research University, 10 rue Vauquelin 75231 Paris cedex05, France*

*CNRS USR 3149, Paris, France*

*2. Laboratoire Stress Oxydants et Cancer, DSV, IBITECS, CEA-Saclay, Gif-sur-Yvette, France*

Contact: shakir.shakir@espci.fr

Reactive oxygen species (ROS) play a crucial role in the regulation of a large array of biological processes ranging from signal transduction and response to stimuli on a cellular level to the process of tissue repair. On the other hand, the accumulation of ROS induces oxidative stress, a phenomenon tied to a number of pathological conditions such as cancer and neurodegenerative diseases. At the proteome level, the presence of an increased amount of ROS leads to a number of posttranslational modifications (PTMs); notably the oxidation of cysteine residues leads to the formation of sulfenic acid (-S-OH), S-nitro groups (S-NO) and disulfides bridges, the latter known as oxidative folding. The need of a better understanding of all these processes has led to the development of specific analytical strategies aiming to characterize and quantify PTMs on cysteine residues. However, redox proteomics remain a technical challenge due to the labile nature of thiol-redox reactions. Furthermore, compared to other PTMs, the number of modified residues per protein can be high. The low abundance of oxidized proteins, combined to the intrinsic heterogeneity of the oxidized forms is another source of complexity.

Our group has developed a robust shotgun strategy called OcSILAC to quantify the change in cysteine oxidation while taking protein profile expression into account. The concept of this approach was presented at the last EUPA meeting in Saint Malo. OcSILAC was tested on a highly oxidized yeast model (∆TRR1) and confirmed the presence of an important oxidative stress condition. We here explore the affected pathways and present the biological significance of our results: from simple oxidized cysteine upregulation that is relevant to oxidative stress to more complicated cases of reduced cysteine upregulation that can only be explained by studying said pathways.