

Plant phosphoproteomics

Jan Fila^{1,2}, Andrea Matros³, Sonja Radau⁴, René Peiman Zahedi⁴, Vera Capkova¹, Hans-Peter Mock³ and David Honys¹

¹Institute of Experimental Botany, Czech Republic

²Charles University in Prague, Czech Republic

³Leibniz Institute of Plant Genetic and Crop Plant Research, Germany

⁴Leibniz-Institut für Analytische Wissenschaften-ISA-e.V., Germany

Protein phosphorylation is one of the most dynamic posttranslational modifications. It often bears the regulatory function, for instance transcription and translation regulation, signal transduction, regulation of cytoskeleton dynamics, cell cycle regulation etc. Despite the importance of phosphorylation, the phosphorylated species are often of lower abundance and co-exist with their native forms in a cell. Due to these reasons together with ion suppression effects, the applying of various enrichment techniques is mostly inevitable.

The enrichment techniques rely on three basic principles – immunoprecipitation, affinity of negatively charged phosphate groups to the positively charged chromatography matrix, and chemical modification of the phosphate groups. The enrichment could be performed either at the level of intact phosphoproteins or alternatively at the level of peptides originating from protein fragmentation done mostly by specific proteases.

In our studies, we concentrate on male gametophyte of tobacco (*Nicotiana tabacum*). Mature pollen is a quiescent, extremely resistant stage with a tough cell wall and rehydrated cytoplasm. Upon pollination, it rehydrates and subsequently the rapid pollen tube growth starts. Such rehydration is accompanied by increased level of metabolism together with translation of the stored transcripts, and such changes are very likely to be regulated via protein phosphorylation, amongst other mechanisms. In order to identify the present phosphoproteins, we applied the combination of phosphoprotein-enriching MOAC with aluminium hydroxide matrix and phosphopeptide-enriching titanium dioxide enrichment. The identification of 139 phosphoprotein candidates was achieved.

Biography

Jan Fila has completed his M.Sc. at the age of 24 years from Charles University in Prague, subject Plant anatomy and physiology. Since autumn 2012, he continues with his Ph.D. at the same university. He has been working for 5 years in the Laboratory of Pollen Biology at the Institute of Experimental Botany. During his studies, he visited IPK in Gatersleben. He has published 3 papers in reputed journals.

fila@ueb.cas.cz