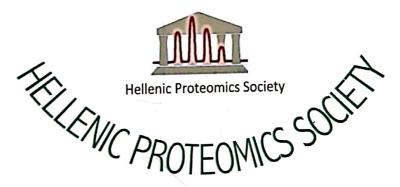
Proteomics and Metabolomics on the Verge of One Health Approach

MEETING

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Unrestricted and quantitative method of post-translational modifications profiling: Timothy grass pollen proteome in relation to increased oxidative stress caused by environmental pollution

Katarina Smiljanic^{a*}, Ivana Prodic^b, Danijela Apostolovic^c, Jelena Mutic^{a,d}, Marianne van Hage^c, Lidija Burazer^c, and Tanja Cirkovic Velickovic^{a,d,f,g}

- a) University of Belgrade–Faculty of Chemistry, CoE for Molecular Food Sciences, Belgrade, Serbia
 - b) Innovation Center Ltd, University of Belgrade-Faculty of Chemistry, Belgrade, Serbia
 - c) Karolinska Institute, Department of Medicine, Solna, Stockholm, Sweden
 d) Ghent University Global Campus, Incheon, South Korea
- e) Institute of Immunology, Virology and Sera Production, "Torlak Institut", Belgrade, Serbia
 - f) Ghent University, Faculty of Bioscience Engineering, Ghent, Belgium
 - g) Serbian Academy of Sciences and Arts, Belgrade, Serbia

The significance of in-depth post-translational modification (PTM) studies of pollen proteomes, when compared with studies on other aspects of pollution and altered pollen allergenicity, has not yet been determined; hence, little progress has been made within this field.

Therefore, we created a comprehensive approach for the comparison of pollen from polluted and environmentally preserved areas. To examine the effects of long-term, in vivo pollen exposure to multiple source pollutants, *Phleum pratense* (Timothy grass) pollen samples were collected along a regional road in Kruševac, central Serbia and were compared with pollen samples from rural, environmentally preserved area over two consecutive pollination seasons. We combined the quantitative comparison of proteome expression profiles from in-solution and 2D-gels with unrestrictive in-depth quantitative PTM profiling using high resolution tandem mass spectrometry and the PEAKS 8.5 Suite platform.

An increased phenolic content and release of sub-pollen particles was found in pollen samples from the polluted area, and significantly higher content of mercury, cadmium, and manganese. Antioxidative defense-related enzymes were significantly upregulated. Seven oxidative PTMs were significantly increased (methionine, histidine, lysine, and proline oxidation; tyrosine glycosylation, lysine 4-hydroxy-2-nonenal adduct, and lysine carbamylation) in pollen exposed to the chemical plant and road traffic pollution sources. Oxidative modifications affected pollen allergens, especially Phl p 6, with several different oxidative modifications.

Quantitative, unrestricted, and detailed PTM searches using an enrichment-free approach was used for the first time to map extensive modifications in the pollen proteome, reflecting increased environmental oxidative stress, primarily caused by increased content of heavy metals in pollen.

*Corresponding author: Katarina Smiljanic Senior Research Associate, PhD University of Belgrade–Faculty of Chemistry, Belgrade, Serbia Studentski trg 12-16, 11000 Belgrade, Serbia Email: katarinas@chem.bg.ac.rs, Tel: +381113336676