

Determination of glyphosate bioconcentration in lettuce (*Lactuca sativa*) simultaneously exposed to cylindrospermopsin in a soil system

P. Lessa¹, S. Sengupta², E. Pinto^{1,3}, M. Freitas^{1,4}, J. Azevedo⁴, F. Oliveira⁴, A. Campos⁴, V. Vasconcelos^{4,5}

¹School of Health, Polytechnic Institute of Porto, Portugal; ²School of Bio-sciences and Technology, VIT University, India; ³REQUIMTE/ Departamento de Ciências Químicas, Laboratório de Bromatologia e Hidrologia da Faculdade de Farmácia da Universidade do Porto, Portugal; ⁴CIIMAR/CIMAR-Interdisciplinary Centre of Marine and Environmental Research, University of Porto, Portugal; ⁵Faculty of Sciences, Porto University, Portugal

Glyphosate has become the most widely used herbicide worldwide. The mode of action of this herbicide is linked to the inhibition of 5-enolpyruvylshikimate-3-phosphate synthase enzyme, which blocks the plant's biosynthesis of aromatic amino acids.

In humans, recent studies suggest that glyphosate can be a potential endocrine disruptor and the *International Agency for Research on Cancer* (IARC) has classified it as probably carcinogenic (group 2 A). Besides the single effects, the interaction of glyphosate with other emergent contaminants that may occur simultaneously in environment can be highly expectable. Cylindrospermopsin is a secondary metabolite of *cyanoHABS* (*cyanobacterial Harmful Algal Blooms*) increasingly recurrent in freshwater. The presence of this cyanotoxin may have negative impacts in the water quality and ultimately in human health, mainly due to the consumption of contaminated water and food. Furthermore, recent studies have suggested that some cyanotoxins (e.g., microcystins) can change the membrane permeability of roots, resulting in changes in the accumulation rates of contaminants in plants. Since edible plants are exposed to a wide variety of substances through irrigation water, there is increasing concern in the potential adverse effects of the interactions between those substances when present simultaneously in a mixture, especially when this can have potential public health consequences. The aim of this study was to determine the bioconcentration of glyphosate in *Lactuca sativa* simultaneously exposed to cylindrospermopsin. In an experimental assay, lettuce plants were irrigated for 15 days in a soil system culture with 50 µg/kg of cylindrospermopsin-containing crude extract and 0.75 mg/kg of glyphosate. The concentration of glyphosate in lettuce plants (roots and leaves) and soil was determined by LC/MS-MS.

The results show that in soil system culture, at the described conditions, there is no bioconcentration of glyphosate in the edible parts of lettuce (BCF of 0.6, i.e., <1). However, interestingly, when plants were exposed to both toxicants in mixture the concentration of glyphosate assimilated by lettuce plants (roots and leaves) was much higher than in the exposure to isolated glyphosate (from 0.04 to 0.21 µg/g in roots and from below the LOQ to 0.84µg/g in leaves). This finding highlights the potential for the enhancement of glyphosate accumulation in plants due to their co-occurrence with cylindrospermopsin, and it underlines the importance of further research regarding the mechanism involved.

According to the results obtained regarding the concentration of glyphosate in soil, it can be hypothesized that it can be in part associated to the higher persistence of glyphosate in soil in the presence of cylindrospermopsin (0.45 µg/g in single exposure and 1.33 µg/g in the exposure to the mixture). Notwithstanding, it would also be important to develop these studies in other culture systems (e.g., hydroponic system), which can maximize the availability of the toxicants to the roots, once the adsorptive effects of soil particles and other plant substrates are not present to retain the toxicants.

Acknowledgements: Horizon2020. Marie Skłodowska-Curie grant agreement No 823860.