

## New insights on Cu homeostasis in vine plants through the use of Cu isotopes

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In this study we examine Cu content and isotopic ratios in soils, soil solutions and organs (i.e., roots and leaves) of grapevine plants (i.e., *Vitis vinifera* – cv. Tanat) grown in pot experiments in greenhouse. Six different soils from three wine-growing areas (i.e., Pessac-Leognan, France; Saint Mont, France and Soave, Italy) were selected for their distinct Cu pesticide input history and their pedological characteristics. We calculated Cu-isotope fractionation between soil and soil solution, and during Cu uptake and transfer within the plants (i.e., from soil solution to roots and from roots to leaves) in order to better understand the effect of plant homeostasis on Cu behavior and isotope signature in the soil-plant system.

Soil solutions with low Cu concentrations (i.e., 20 to 100  $\mu\text{g.L}^{-1}$ ) of the Soave and Saint Mont soils are isotopically heavy compared to the respective solid phase ( $\Delta^{65}\text{Cu}$  solution-soil  $\sim 0.4\%$ ), implying a major role of organic ligands in Cu mobility in the soil solution. On the contrary, no significant Cu isotope fractionation was measured between soils and soil solutions from the Pessac-Leognan region, where Cu concentration in soil solution was high (i.e., above 400 until 4000  $\mu\text{g.L}^{-1}$ ). Cu isotope fractionation from soil solution to roots and in a lesser extend during Cu transfer from roots to leaves appear to depend on Cu content in the soil solution and the amount of Cu incorporated into the roots. Our results are in line with the existence of passive and active Cu incorporation pathways, controlled by Cu availability in the soil solution. In the case of low Cu bioavailability, active Cu incorporation in roots is associated with the incorporation of light Cu isotopes (negative  $\Delta^{65}\text{Cu}$  root-solution from -0.21 to -0.39%). By contrast, in the case of high Cu bioavailability, more heavy Cu isotopes passively enter the apoplasm ( $\Delta^{65}\text{Cu}$  root-solution from 0.05 to 0.23%) where it can probably be stored to prevent Cu toxicity. In contrast to data previously published, our study shows that  $\Delta^{65}\text{Cu}$  leaf-root fractionation favors light isotopes, independently of the soil modality.