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Soil microbial toxicity assessment of a copper-based fungicide in two contrasting soils

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The infestation with the fungus downy mildew (Plasmopara viticola) causes dramatic losses in wine production. Copper (Cu) based fungicides have been used in viticulture since the end of the 19th century, and until today both conventional and organic viticulture strongly rely on Cu to prevent and reduce fungal diseases. Consequently, Cu has built up in many vineyard soils and it is still unclear how this affects soil functioning. The aim of the present study is the evaluation of the soil microbial toxicity of Cu contamination.

Two contrasting agricultural soils, an acidic sandy soil and a calcareous loamy soil, were sampled to conduct an eco-toxicological greenhouse pot experiment. The soils were spiked with a commonly used fungicide based on copper hydroxid in seven concentrations (0, 50, 100, 200, 500, 1500 and 5000 mg Cu kg-1 soil) and Lucerne (Medicago sativa L. cultivar. Plato) was grown in the pots for 3 months.

Sampling was conducted at the beginning and at the end of the study period to test copper's soil microbial toxicity in total microbial biomass and basal respiration, as well as enzyme activities, such as exoglucanase, β-glucosidase, exochitinase, phosphatase, protease, phenol-, peroxidase and urease. Additionally, DOC, TN, Cmic, Nmic, NO_3 and NH4 were determined to provide further insight into the carbon and nitrogen cycle. Microbial community structure was analysed by phospholipid fatty acids (PLFAs), and ergosterol as a fungal biomarker. In addition, molecular tools were applied by extracting soil DNA and performing real time quantitative polymerase chain reaction (qPCR) and a metagenomic approach using 16S and ITS amplification and sequencing with MiSeq platform for the second sampling.

Hydrolytic extracellular enzymes were not clearly affected by rising Cu concentrations, while a trend of increasing activity of oxidative enzymes (phenol- and peroxidase) was observed. Microbial respiration rate as well as the amount of Cmic and Nmic decreased with increasing Cu concentrations. Ergosterol was especially sensitive to Cu and started to decline at even lower concentrations. A shift in the microbial community structure with rising Cu was observed using PLFA. The metagenomics approach enables us to investigate these changes at even finer taxonomic resolution and possible effects on the soil nutrient cycles will be discussed. In summary, our results showed distinct Cu toxicity effects on soil microbial biomass with a larger sensitivity of the loamy soil.