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Processing and statistical analysis of soil-root images

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Importance of the hotspots such as rhizosphere, the small soil volume that surrounds and is influenced by plant roots, calls for spatially explicit methods to visualize distribution of microbial activities in this active site (Kuzyakov and Blagodatskaya, 2015). Zymography technique has previously been adapted to visualize the spatial dynamics of enzyme activities in rhizosphere (Spohn and Kuzyakov, 2014). Following further developing of soil zymography –to obtain a higher resolution of enzyme activities - we aimed to 1) quantify the images, 2) determine whether the pattern (e.g. distribution of hotspots in space) is clumped (aggregated) or regular (dispersed).
To this end, we incubated soil-filled rhizoboxes with maize Zea mays L. and without maize (control box) for two weeks. In situ soil zymography was applied to visualize enzymatic activity of β-glucosidase and phosphatase at soil-root interface. Spatial resolution of fluorescent images was improved by direct application of a substrate saturated membrane to the soil-root system. Furthermore, we applied "spatial point pattern analysis" to determine whether the pattern (e.g. distribution of hotspots in space) is clumped (aggregated) or regular (dispersed).
Our results demonstrated that distribution of hotspots at rhizosphere is clumped (aggregated) compare to control box without plant which showed regular (dispersed) pattern. These patterns were similar in all three replicates and for both enzymes. We conclude that improved zymography is promising in situ technique to identify, analyze, visualize and quantify spatial distribution of enzyme activities in the rhizosphere. Moreover, such different patterns should be considered in assessments and modeling of rhizosphere extension and the corresponding effects on soil properties and functions.

Key words: rhizosphere, spatial point pattern, enzyme activity, zymography, maize.