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Investigation on organic matter-mineral interaction by confocal multispectral and time-resolved microscopy

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Organic matter makes up less than 5% of soil and sediment and is often linked to the particulate mineral fraction, forming an organo-mineral microsystem that certainly contributes to its stability in the natural environment. However, most of the techniques used to study and quantify organic matter necessarily require the separation of the organic phase from the mineral phase by chemical or physical extraction and, subsequently fractionation methods that destroy and/or degrade the original aggregate morphological structure. The present work demonstrates that Scanning Confocal Microscopy (MC) can be used as a non-destructive fluorescence technique capable of characterizing organic matter (OM) interacting with the surface of the mineral fraction in soils and sediments without prior sample preparation and use of extraction or chemical fractionation of its components. Organic matter (OM) interacts with the mineral surface through molecular stacking in the form of stable molecular aggregates. Besides that, aggregate states also favor energy transfer processes between aggregated molecules which strongly affects the dynamics of excited state producing spectral shifts to the red and changes in life-time that can be correlated to aggregate morphology and to molecular amount deposited on the surface. These features confer a high spectral and intensity contrast of the confocal images. Here, infrared 2-photon (2P) excitation proved to be adequate to selectively excite OM aggregate states in the visible region between 400 and 700 nm, which allows a direct access to the fundamental aspects of the organic matter-mineral interactions.

We will show that the use of confocal methodologies, together with image analysis, provide helpful tools to understand the complex OM interactions at a molecular level. Here, we studied the interaction of OM with sodium bicarbonate and sodium hydroxide surfaces that form fractal crystals. When a drop of water containing both soil and solubilized bicarbonate or hydroxide salts is dried on a glass surface, dendritic-type salt crystals are first formed on the glass surface within the water droplet. In a second step of the droplet drying process, suspended organic molecules deposit on the surface of these fractal crystals. We will show that the morphology and molecular packaging substantially change spectral and life-time properties which strongly depend on the amount of OM on the crystal surface. Special features can be obtained from linear unmixing of

spectral images using 1P and 2P excitation at 375 nm and 750 nm respectively for OM interacting with powder bicarbonate. Therefore, molecular aggregates of interacting fluorescence-emitting species can be used to characterize OM regarding the morphological, molecular structure and interactions with inorganic surfaces. These properties determine the stability of the original OM packing and the limits for the molecular stacking on different active surfaces in nature.