

Long-term mesocosm experiments to investigate microbial degradation of fluorescent tracers

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Uranine (UR) and sulforhodamine B (SRB) are two of the most commonly used fluorescent tracers in hydrology. Their suitability to be used as ideal tracers has been discussed since they might interact with the soil or become degraded. However, these properties have recently served to mimic processes of sorption and degradation of pollutants. The present study attempts to investigate to what extent UR and SRB could be used to imitate such processes in environments where saturation conditions are variable and the presence of plants might play an important role. For this purpose, both tracers were applied to 36 small mesocosms filled with a layer of 10 cm of gravel and 30 cm of sand in which 6 types of treatments, with 3 replicates each, were implemented based on the presence of two species of wetland plants (*Typha latifolia* and *Phalaris arundinacea*) and two types of hydrological conditions (saturated and unsaturated). The entire experiment lasted 10 months, during which two injections of equal concentration of tracers were performed. The first months served to ensure the adaptability of the plants and to achieve stationary conditions in the system. Water and sediment samples were collected weekly after the second injection while plants were measured at the end of the experiment distinguishing between roots and aerial parts. Mass balances of the tracers were combined with excitation emission matrix fluorescence spectroscopy (EEMS) to characterize dissolved organic matter in the water and soil. Degradation was quantified by subtracting the non-degraded tracer fraction (sorption and plant uptake) and the remaining non-degraded mass in the water from the tracer mass injected. Results revealed that most of the SRB accumulated in the sand in agreement with its sorption affinity, while UR was mainly found in the pore water. Both tracers showed more degradation in the treatments with plants than the controls. Overall, UR exhibited higher degradation than SRB. Differences between plant species were not found for UR, whereas SRB showed more degradation with *Phalaris* than with *Typha*. The alternation of oxic and anoxic conditions in the unsaturated treatments increased the degradation of both tracers, especially SRB. Two components were identified by parallel factor analysis of the EEMS in addition to the UR, one humic-like associated with biological activity (C1) and one humic-like of terrestrial origin (C3). A high positive correlation (Spearman's $\rho = 0.81$, $p < 0.0001$) between C1 and UR suggested a probable link, in which the increase of UR would lead to an increase of C1. This hypothesis was supported by the results from the controls, where UR exhibited the highest values of C1 while it was absent with SRB. This suggested a possible association of UR with microbial activity, which is in agreement with the high overall degradation rates found for UR. Further research is needed to confirm these results, but we see the combination of EEMS with tracer mass analysis as a promising way to provide additional information on processes of sorption and degradation of contaminants.