

Affects N fertilization intensity and composition of root exudation from two plant species differing in their exploitation strategy?

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The rhizosphere represents one of the most important hotspots of microbial activity in soil. As such, it controls soil element cycling and significantly contributes to important ecosystem processes like C and N sequestration. The close plant-microbe-soil interactions in the rhizosphere are mediated by the input of labile exudates into the surroundings of plant roots. Thus microbial performance is constrained by the intensity and composition of root exudation. However, it is poorly understood how closely root exudation corresponds with the plant metabolome and how it is related to plant traits and changing environmental conditions. To fill this gap, we determined the composition of the root metabolic pool and root exudates in two plant species differing in their exploitation type (conservative *Carex acuta* versus competitive *Glyceria maxima*) grown for two months in controlled conditions and treated weekly by two levels of foliar N fertilization. Based on previous studies, we knew that *Glyceria* has, compared to *Carex*, a lower tissue C:N ratio, higher photosynthetic rate, higher allocation belowground and also larger investment to exudation. Prior to extraction, the roots were cleaned by water and immediately frozen in liquid N₂. The root exudates were collected from carefully cleaned roots of living plants encased in glass vials with water and subsequently lyophilised. Both sample types were silylated and analysed for their metabolic profiles using GC-MS/MS.

Our results revealed that the metabolite content in root tissue (DW basis) of *Glyceria* was on average lower compared to *Carex*, but increased with fertilization, while the root tissue of *Carex* was characterized by significantly higher metabolite content in the low intensity fertilization treatment compared to both the control and high N fertilization intensity. In contrast, the amount of exuded compounds was much higher in *Glyceria* compared to *Carex* in the control plants, but decreased for *Glyceria* and increased for *Carex* in fertilized plants, resulting in comparable exudate flow from the most fertilized plants of both species. The exudation intensity decreased from 24% of the tissue metabolic content during 1h in non-fertilized *Glyceria* individuals to 7% in most fertilized plants, while *Carex* released between 3% and 5% of the root metabolite content. The *Glyceria* exudates contained significantly higher amounts of sugars and organic acids compared to their root metabolic pool, and significantly higher proportion of sugars compared to exudates from *Carex*. Considering the metabolic profiles, the composition of exudates from *Glyceria* was significantly distinct between the fertilized and unfertilized individuals, while the fertilized *Carex* plants closely corresponded to the controls. Our results have shown that *Glyceria*, representing competitive plant species, invest high proportion of assimilates into exudation under N limiting conditions, but strongly reduces these expenses when N is available. It also actively controls the composition of root exudates released into the soil environment, while exudation from *Carex* roots result rather from a passive diffusion of low molecular compounds from the root tissue.