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Probing potential microbial coupling of carbon and nitrogen cycling during decomposition of maize residue by ¹³C-DNA-SIP

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ABSTRACT

The links between microbial taxa, *in situ* organic matter decomposition, and coupling of carbon (C) and nitrogen (N) cycles remain unresolved. Here, we used stable isotope probing (SIP) technique to investigate bacterial carbon assimilation and C and N coupling during decomposition of ¹³C-labeled maize residue in a black soil from Northeast China. Bacteria assimilating carbon from maize residue (16S rRNA analysis) were primarily distributed in the Phyla Actinobacteria, Firmicutes and Proteobacteria. These include the recognized stubble decomposing lineages of *Arthrobacter*, *Streptomyces*, *Bacillus* and *Rhizobium*, but also lineages not previously reported (*Agromyces*, *Blastococcus*, *Gemmatimonas*, *Glycomyces*, *Heliobacillus*, *Lysobacter*, *Microlunatus*, *Mycoplasma*, *Natronocella*, *Ohtaekwangia*, *Paenibacillus*, *Schlege-lella*, *Sorangium*, *Steroidobacter* and *Thermacetogenium*). Analysis of nitrogen fixation (*nifH*) and denitrification (*nirS*) genes in heavy-fraction DNA was used to link microbial taxa involved in N cycling to C transformation of the maize residue. A cluster of *nifH* genotypes affiliated with Rhizobium and two other 'uncultured' clusters dominated the N-fixing clone library, and genotypes affiliated with *Kocuria varians* and an uncultured cluster dominated the library of nitrite reducing (*nirS*) taxa. The results suggest that plant residue decomposition may stimulate both N-fixation and denitrification through direct C-feeding of related microbes in soil.

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1. Introduction

Globally it is estimated that 3.75 billion tons of crop residues are produced annually (Smil, 1999), and most of these are incorporated into arable soils. The decomposition of crop residues not only affects the soil C balance, but also greatly influences the cycling of other elements such as nitrogen (N), phosphorus (P), and sulfur (S), and thereby impacts overall soil fertility (Smil, 1999; Lu et al., 2009). Efforts have been made to better understand the processes of residue decomposition and coupling of nutrient cycles. For instances, abiotic factors, such as residue chemistry, soil properties (e.g. N availability) and climate, have been identified as the major environmental factors controlling residue decomposition (Melillo et al., 1982; Aerts, 1997). The diversity and succession of soil

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microbes, the major drivers of residue decomposition, have also been frequently monitored during this process (Rui et al., 2009; Marschner et al., 2011). However, the linking of microbial taxa with function is not well understood and remains a key area for investigation.

Recently, stable isotope probing (SIP) has been used to bridge the gap between microbial identity and function during processes such as residue decomposition. Bernard et al. (2007) used this technique to identify bacteria associated with wheat-residue cycling and found that C from ¹³C-labeled wheat was mainly assimilated by the Betaproteobacteria taxa *Janthinobacterium*, *Massilia* and *Variovorax*, and the Gammaproteobacteria taxa *Xanthomonas* and *Pseudomonas*. Subsequently, several studies using ¹³C-labeled fresh potato, alfalfa and rice or ¹⁵N-labeled maize and soybean have demonstrated that active residue-decomposing microbes were unevenly distributed among the phyla Actinobacteria and Proteobacteria, and the taxa assimilating the labeled residues differed with residue chemistry, soil moisture and soil types (España et al., 2011a; Shrestha et al., 2011; Li et al., 2012; Semenov







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