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Short communication

Rhizosphere processes associated with the poor nutrient uptake in no-tillage rice (*Oryza sativa* L.) at tillering stage

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ABSTRACT

No-tillage (NT) can result in poor nutrient uptake and reduce biomass production in rice (*Oryza sativa* L.) at early growth stages, but limited information is available on the factors that contribute to the poor nutrient uptake. This study aimed to determine the rhizosphere processes associated with the poor nutrient uptake in NT rice at early growth stages. In field experiments conducted at Changsha, Hunan Province and Nanning, Guangxi Province, China the N, P and K uptake, rhizosphere soil properties and root morphological traits at the tillering stage were compared for rice grown under NT and conventional tillage (CT). The NT rice had 17–43% less N, P and K uptake than did the CT rice. The rhizosphere soil available N, P and K contents were 9–18% greater for the NT rice than the CT rice. The root biomass, surface and length were 7–48% less for rice under NT than under CT. NT rice accumulated 26–37% more pseudomonas than did CT rice in rhizosphere soils. Bioassays indicated that the pseudomonas treatment led to 43–59% reductions in root length, surface and biomass and 39–52% reductions in N, P and K uptake. These results indicate that reduced root growth induced by accumulation of rhizosphere inhibitory pseudomonas was responsible for the poor nutrient uptake in NT rice at the tillering stage. This highlights the need for developing ways to avoid the effect of deleterious soil organisms on NT rice at the tillering stage.

1. Introduction

No-tillage (NT), a crop production system in which the soil is left undisturbed from harvest to planting, has potential benefits of soil conservation and reducing production costs. NT is now practiced on more than 100 million hectares worldwide and most of this NT land lies in North and South America and Australia and is devoted to upland crops (Huggins and Reganold, 2008; Derpsch et al., 2010). In recent years, NT has become increasingly attractive for rice (*Oryza sativa* L.) production in China because of its social, economical and environmental benefits (Huang et al., 2011a). Accordingly, many studies have been conducted to assess the effect of NT on rice (Huang et al., 2011a), and it is reported that NT can cause poor nutrient uptake and consequently reduced biomass production in rice at early growth stages (Huang et al., 2011b, 2012).

Crop nutrient uptake is determined by nutrient supply and nutrient uptake capacity. Nutrient supply can be reflected in rhizosphere available nutrient contents and affected by rhizosphere pH through its influence on cation mobility (Kirk et al., 1993; Zhang et al., 2010). Chen et al. (2013) conducted pot experiments and observed that rice seedlings grown under NT had equal or higher available nutrient contents and a slightly lower pH in rhizosphere soil than under conventional tillage (CT). This suggests that nutrient supply may not be responsible for the poor nutrient uptake in NT rice seedlings, but field experiments are needed to confirm it. On the other hand, nutrient uptake capacity is closely related with root traits. There have been reports showing that NT can lead to reductions in root length and biomass in rice at early growth stages (Jiang et al., 2005; Huang et al., 2012; Chen et al., 2013). In this regard, in the past it was usually assumed that the reduced root growth in NT crops was attributed to soil

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compaction (Hammel, 1989; Mahboubi et al., 1993). But recently, there is growing evidence that rhizosphere microbial changes, often associated with accumulation of inhibitory pseudomonas, are responsible for the reduced root growth in NT crops (Simpfendorfer et al., 2002; Watt et al., 2003, 2006). However, this evidence is mainly from upland crops, and there is limited information available on the relationships between rhizosphere microbial properties and root growth in NT rice (Huang et al., 2013).

In our present study, we compared N, P and K uptake, rhizosphere soil properties and root morphological traits between NT and CT rice at tillering stage in two field experiments. Our objective was to determine the rhizosphere processes that contributed to the poor nutrient uptake in NT rice at the tillering stage.

2. Material and methods

Field experiments were conducted at the Experimental Farm of Guangxi University in Nanning (22°51′N, 108°17′E, 78 m asl), Guangxi Province and at the Experimental Farm of Hunan Agricultural University in Changsha (28°11′N, 113°04′E, 32 m asl), Hunan Province of China. The experiments in Nanning and Changsha were established in 2011 and 2004, respectively. The soil in Nanning was a Ferralsol (FAO classification) with pH = 6.22, organic matter = 24.2 g kg^{-1}, available N = 142 mg kg^{-1}, available P = 34.8 mg kg^{-1}, and available K = 123 mg kg^{-1}; and in Changsha was a Fluvisol with pH = 6.04, organic matter = 15.0 g kg^{-1}, available N = 137 mg kg^{-1}, available P = 38.4 mg kg^{-1}, and available K = 113 mg kg^{-1}. The soil test was based on samples taken from the upper 20 cm (Ap horizon) of the soil. The cropping systems were continuous rice in Nanning and rice-oilseed rape rotation in Changsha.

Rice cultivars Guiliangyou 2 and Liangyoupeijiu were used in Nanning and Changsha, respectively. These two cultivars have been widely grown by rice farmers in China. In each site, two tillage methods were set as experimental treatments: no-tillage (NT) and conventional tillage (CT). CT operations were performed by one plowing and two harrowing using a two-wheel power-tiller in Nanning and a water buffalo in Changsha, and the tillage depth was about 20 cm in both sites. Plots were arranged in a randomized complete block design. The experiments were replicated 3 times in Nanning with a plot size of 36 m^2 and 4 times in Changsha with a plot size of 30 m^2. In Nanning, fifteen-day-old seedlings were manually thrown on the soil surface at a density of 29 hills per m^2 with 2 seedlings per hill. Plants received 210 kg N ha^{-1}, 72 kg P_2O_5 ha^{-1} and 180 kg K_2O ha^{-1}. N and K were applied in three splits: 50% as basal, 30% at mid-tillering and 20% at panicle initiation. P was applied as basal. In Changsha, twenty five-day-old seedlings were manually transplanted at a depth of about 2 cm and a hill spacing of 20 cm × 20 cm with 1 seedling per hill. Plants received 150 kg N ha^{-1}, 90 kg P_2O_5 ha^{-1} and 180 kg K_2O ha^{-1}. N was applied in three splits: 50% as basal, 30% at mid-tillering and 20% at panicle initiation. P was applied as basal. K was split equally at basal and panicle initiation. In CT plots, the basal fertilizers were broadcast after the first harrowing and incorporated with the second harrowing. In NT plots, the basal fertilizers were broadcast on the soil surface with no incorporation. The strategy for water management was in the sequence of flooding (about 1.5 cm for seedling throwing; about 3 cm for transplanting), midseason drainage (a period of 10 days just before panicle initiation), reflooding (about 3 cm) and moist intermittent irrigation (grain filling period). Weeds were controlled by herbicides. Paraquat was applied at 7 days before planting for NT plots and bensulfuron-methyl and butachlor were applied at 5 days after planting for both NT and CT plots. Insects and diseases were intensively controlled using chemicals.

Plant and soil samples were taken at 20 days after seedling throwing in Nanning and at 25 days after transplanting in Changsha in 2014. In each site, 20 representative hills (excluding two border hills) of rice plants were selected in each plot. After uprooting the plants, rhizosphere soil was collected from the soil portion tightly adhering to the roots by shaking off the loosely adhering soil (Chen et al., 2013). The rhizosphere soil from each plot was mixed and separated into two sub-samples. One sub-sample was air-dried at room temperature to measure pH and available N, P and K contents. The pH was determined in a soil-water suspension (10 g air-dried soil in 25 ml water) using a digital pH meter (Model 868, Thermo Orion, Massachusetts, USA). The available N content was measured by transforming N into NH_3 (NaOH hydrolyzation diffusion), absorbed by H_3BO_3 solution (2%) at 40 °C for 24 h and then titrated with H_2SO_4 solution (0.005 mol L^{-1}). The available P content was extracted with double acid solution (0.05 mol L^{-1} HCl and 0.0125 mol L^{-1} H_2SO_4) and then measured using ammonium molybdate colorimetric method. The available K content was measured by a flame photometer (FP640, Shanghai Precision & Scientific Instrument Inc., Shanghai, China) after extracting with NH_4OAc solution (1 mol L^{-1}, pH 7.0). Another sub-sample was placed in a sterile zip-lock bag, kept cool on ice during transport to the laboratory, and stored at 4 °C in the laboratory. This sub-sample was used to determine pseudomonas (*Pseudomonas aeruginosa*) number. Two separate 1 g soil from each sub-sample were suspended individually in 10 ml sterile distilled water. Soil suspensions were vortexed for 60 s. Dilutions of the vortexed solution were plated onto pseudomonas isolation agar (Difico, Becton Dickinson, Sparks, MD, USA), incubated for 48 h at 35 °C, and then colony forming units (CFUs) counted using standard spread plate technique (Seeley Jr. et al., 1991). The roots of the uprooted plants were cut at the stem base. The remaining roots in soil were collected by handpicking. All roots from each plot were combined, washed with distilled water, and then scanned using a scanner (Epson Expression 1680 Scanner, Seiko Espon Corp.

Table 1
Aboveground biomass and nutrient uptake in rice seedlings grown under conventional tillage (CT) and no-tillage (NT) in Nanning (cv. Guiliangyou 2), Guangxi Province and Changsha (cv. Liangyoupeijiu), Hunan Province of China in 2014.

Tillage method	Aboveground biomass (g m^{-2})	Nutrient uptake (g m^{-2})		
		N	P	K
Nanning				
CT	96 (18)	3.48 (0.55)	0.42 (0.07)	1.22 (0.19)
NT	59 (8)*	2.57 (0.34)*	0.30 (0.05)*	0.70 (0.10)*
Changsha				
CT	128 (19)	3.88 (0.62)	0.58 (0.07)	1.54 (0.22)
NT	104 (7)*	3.00 (0.15)*	0.47 (0.03)*	1.28 (0.12)*

Data are means (SD) (n = 3 and 4 in Nanning and Changsha, respectively).
* Indicates significant difference between the two tillage methods according to LSD at P = 0.05.