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JOURNAL OF
ENVIRONMENTAL
SCIENCES
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Copper pollution decreases the resistance of soil microbial community to subsequent dry–rewetting disturbance

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ARTICLE INFO

Article history:

Received 28 July 2015

Revised 19 October 2015

Accepted 19 October 2015

Available online 19 December 2015

Keywords:

Copper stress

Dry–rewetting disturbance

Bacterial community

Substrate induced respiration

Resistance

ABSTRACT

Dry–rewetting (DW) disturbance frequently occurs in soils due to rainfall and irrigation, and the frequency of DW cycles might exert significant influences on soil microbial communities and their mediated functions. However, how microorganisms respond to DW alternations in soils with a history of heavy metal pollution remains largely unknown. Here, soil laboratory microcosms were constructed to explore the impacts of ten DW cycles on the soil microbial communities in two contrasting soils (fluvo-aquic soil and red soil) under three copper concentrations (zero, medium and high). Results showed that the fluctuations of substrate induced respiration (SIR) decreased with repeated cycles of DW alternation. Furthermore, the resistance values of substrate induced respiration (RS-SIR) were highest in non-copper-stressed (zero) soils. Structural equation model (SEM) analysis ascertained that the shifts of bacterial communities determined the changes of RS-SIR in both soils. The rate of bacterial community variance was significantly lower in non-copper-stressed soil compared to the other two copper-stressed (medium and high) soils, which might lead to the higher RS-SIR in the fluvo-aquic soil. As for the red soil, the substantial increase of the dominant group WPS-2 after DW disturbance might result in the low RS-SIR in the high copper-stressed soil. Moreover, in both soils, the bacterial diversity was highest in non-copper-stressed soils. Our results revealed that initial copper stress could decrease the resistance of soil microbial community structure and function to subsequent DW disturbance.

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Introduction

Terrestrial ecosystems are increasingly influenced by intensive anthropogenic perturbations and climatic changes (Singh et al., 2010), which are predicted to profoundly affect belowground nutrient allocation and lead to increased

fluctuations in precipitation magnitude and frequency at regional and continental scales (Dore, 2005). Soil drying–rewetting (DW) is a common environmental disturbance in agricultural practices exemplified by the frequencies and intensities of irrigation (Shi and Marschner, 2014). Moreover, dry–wet alternations are frequently encountered in paddy

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soils in different seasons, where the field is maintained under flooded conditions in the crop-growing season and under drained conditions in the off-crop season (Kimura and Asakama, 2006). The significant changes in soil conditions due to DW alternations might considerably affect the soil-dwelling microorganisms, which are essential to the sustainability of ecosystem functioning and services. However, our knowledge is lacking regarding the responses of soil microbial communities to the DW alternations, which strongly affects our ability to predict the responses of ecosystem functions to future environmental disturbance.

In recent years, the influence of DW disturbance on soil microbial activity has been frequently investigated (Fierer and Schimel, 2002; Borken and Matzner, 2009). For example, soil nitrification activity was assumed to be inhibited by osmotic stresses due to limited diffusion of substrates in dry soil (Hu et al., 2015a, 2015b). Some microorganisms can withstand the stress and survive by accumulating osmoregulatory solutes under water-limited conditions (Yao et al., 2011). After the rewetting of dry soil, the water potential sharply decreases, which leads to the death of microorganisms by bursting of cells, while microbes with high tolerance quickly release the accumulated solutes to survive (Shi and Marschner, 2014; Hu et al., 2015a, 2015b). It is generally accepted that soil DW events can cause a short-lived flush of carbon, nitrogen and phosphorus nutrients into the soil solution, which increases substrate and nutrient availability to the surviving microbes and stimulates their microbial activity (Shi and Marschner, 2014). Despite these studies on the impacts of DW disturbance on microbial activity, however, few studies had comprehensively investigated the shifts of soil microbial community composition, diversity and function in response to the DW disturbance.

Previous studies suggested that the resistance of a stressed microbial community to a subsequent disturbance depends on the nature of the disturbance (Tobor-Kaplon et al., 2005). Margesin et al. (2011) argued that if the primary and subsequent disturbances were similar, the resistance and resilience of soil microbial community would be stronger. The similarity of the primary and subsequent disturbances might contribute to the development of co-tolerance of microbial communities (Philippot et al., 2008). However, how the resistance of a stressed microbial community will respond to a new disturbance remains largely unknown. Copper is released into the environment by anthropogenic activities, such as sewage irrigation, mining activities, and intensive use of pesticides and herbicides (Smith, 2009). Approximately 3.4 million tons of copper is released to the terrestrial surface every year (Zhou et al., 2000), thus it is necessary to study how the soil microbial community is affected by copper pollution in response to DW disturbance, especially repeated DW alternations.

The main objective of this study was to explore the responses of the bacterial community and activity in copper-contaminated soils to subsequent DW disturbance. As an essential component of soil communities, soil bacteria play pivotal roles in nutrient cycling processes, and they are largely responsible for the functioning of farmland ecosystems (Hu et al., 2015a, 2015b). The activities of soil microorganisms mostly contribute to soil respiration, yet most microorganisms in the

soil are dormant, which leads to low soil respiration (Lin and Brookes, 1999). Soil respiration can be stimulated by adding easily decomposable substrates. We therefore measured substrate induced respiration (SIR) instead of soil basal respiration to characterize the microbial activity. The changes in the bacterial diversity and community composition were also investigated in response to repeated DW alternations. Soils collected from field sites with contrasting soil properties were incubated in a microcosm experiment with ten cycles of DW alternation. We hypothesized that: (1) the resistance of SIR to disturbance might fluctuate in response to repeated DW transitions and be higher in the copper-stressed soils compared to non-copper-stressed soils; (2) the soil bacterial community might significantly change during a period of repeated DW alternations, and the shifts in the bacterial community might determine the resistance of SIR to the disturbance.

1. Materials and methods

1.1. Field description and soil sampling

Soil samples were collected from two long-term experimental sites located in Dezhou (37.33°N, 116.63°E) of Shandong province, and in Qiyang (26.75°N, 111.88°E) of Hunan province, China. The soil in Dezhou is classified as fluvo-aquic soil while the soil in Qiyang as red soil, and the two soils have distinct soil properties, as described previously (Li et al., 2015). Soil pH in Dezhou and Qiyang was measured as 7.9 and 4.3, respectively. The fluvo-aquic soil is a sandy soil with a ratio of clay: silt: sand of 18:18:64, while the red soil is clayey, with a ratio of clay: silt: sand of 46:35:19. In both sites, different amounts of copper chloride powders were mixed thoroughly with surface soil and then applied back into the plots in July 2007. There were eight different copper treatments with the copper levels of 0, 50, 100, 200, 400, 800, 1600, and 3200 mg/kg soil in the fluvo-aquic soil, and 0, 12.5, 25, 50, 100, 200, 400, and 800 mg/kg soil in the red soil. The plots had been planted with maize-wheat rotations under conventional agricultural management practices since 2007. Soil samples were collected from the top 10 cm by mixing five soil cores for each plot in July 2012 and stored at 4°C before the microcosm experiment.

1.2. Soil microcosm incubation

According to a previous study on the effects of copper addition on bacterial communities in the same two long-term experimental sites located in Dezhou (37.33°N, 116.63°E) and Qiyang (26.75°N, 111.88°E) (Li et al., 2015), the added copper concentrations could be classified into three categories including zero, medium and high concentrations based on the significant changes of bacterial community composition, that is, 0, 800 and 3200 mg/kg added copper in the fluvo-aquic soil compared with 0, 200 and 800 mg/kg added copper in the red soil. The treatment without copper addition (Cu0) was regarded as the control in both soils, and DW alternations were performed in soils with three different copper levels. In total, soil microcosms were constructed with eight treatments including Cu0,

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