Past water management affected GHG production and microbial community pattern in Italian rice paddy soils

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ABSTRACT

The water management system of cultivated paddy rice soils is one of the most important factors affecting the respective magnitudes of CH_4 and N_2O emissions. We hypothesized an effect of past management on soil microbial communities and greenhouse gas (GHG) production potential. The objectives of this study were to i) assess the influence of water management history on GHG production and microbial community structure, ii) relate GHG production to the microbial communities involved in CH_4 and N_2O production inhabiting the different soils. Moreover, the influence of different soil conditioning procedures on GHG production was determined. To reach these aims, we compared four soils with different water management history, using dried and sieved, pre-incubated and fresh soils.

Soil conditioning procedures strongly affected GHG production: drying and sieving induced the highest production rates and the largest differences among soil types, probably through the release of labile substrates. Conversely, soil pre-incubation tended to homogenize and level out the differences among soils.

The water management history strongly affected microbial community structure, which was itself tightly linked to CH_4 and N_2O production. N_2O production was the highest in aerobic soil, which also exhibited the strongest evidence for active nitrifying communities (*NirK*). Drying and rewetting aerobic soil enhanced the production of nitrate, which was further reduced to N_2O through denitrification. As expected, CH_4 production was the lowest in aerobic soil, which showed a less abundant archaeal community. This work supports the hypothesis that microbial communities in paddy soils progressively adapt to water management practices, thereby reinforcing potential differences in GHGs production.

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1. Introduction

Agricultural activities significantly contribute to emissions of key greenhouse gases (GHGs): carbon dioxide (CO_2), methane (CH_4) and nitrous oxide (N_2O) (Smith et al., 2007). Aerobic arable soils mainly emit CO_2 and N_2O, the latter being released in direct proportion of N fertilizer input (van Groenigen et al., 2010). By contrast, irrigated rice cropping systems under mostly anaerobic conditions are responsible for 5—20 per cent of CH_4 emission from all anthropogenic sources (Tilman et al., 2001), and CH_4 emissions contribute the majority of global warming potential (GWP) in rice systems (Linquist et al., 2012).

The CH_4 emission from irrigated rice fields is controlled by production, oxidation and transport processes (Krüger et al., 2001). Following flooding, strictly anaerobic methanogenesis (either acetoclastic or hydrogenotrophic) produces CH_4 as terminal product of anaerobic mineralization of soil organic matter (SOM) degradation, in the absence of alternative electron acceptors (O_2, NO_3^-, Fe(III), and SO_4^{2-}) or microbes capable of using those. The CH_4 produced in anoxic deep soil layers can be oxidized to CO_2 by methanotrophic bacteria while diffusing upwards through oxic soil/water layers.

By contrast, N_2O is produced by soil microbes under both aerobic and anaerobic conditions, through multiple chemical and biological processes, including nitrification (autotrophic and heterotrophic), denitrification, nitrifier—denitrification, co-

denitrification, nitrate ammonification, chemodenitrification of soil nitrite and abiotic decomposition of ammonium nitrate, as reviewed by Butterbach-Bahl et al. (2013). Of these processes, nitrification has a dual role in direct N_2O production (side-product in oxic conditions; end product in nitrifier denitrification in anoxic conditions) and in indirect production by supplying NO_3^- to denitrification. Both nitrifying and denitrifying bacteria have genes coding for each of the enzymes participating in the oxidative and reductive steps. For example, nirK is pivotal for N cycle processes, as it plays a role in ammonia oxidization to nitrite in nitrification (Nitrosomonas europaea), and regulates dissimilatory nitrate reduction in denitrification. Hence, nirK abundance by itself cannot be used to apportion in situ N_2O emissions from denitrification vs. nitrification sources, but it is a valuable indicator of the potential for one or both of these processes to occur. In depth analysis of the site dependent differences in gas kinetics and in gene abundance, as we attempted in this study, can provide more information on the actual functional differences of the studied sites.

However, in general N_2O emissions are very low in permanently flooded rice paddies due to inhibited nitrification and usage of ammonium rather than nitrate based fertilizers (Smith, 1982; Zou et al., 2005), whereas peaks occur during alternate wetting and drying or midseason drainage (Cai et al., 1997; Zheng et al., 2000; Zou et al., 2005). The water management system under which rice is grown is therefore one of the most important factors affecting the respective magnitudes of CH_4 and N_2O emissions. Field drainage, while significantly reducing CH_4, may actually increase N_2O emissions under conditions promoting nitrification and denitrification (Kudo et al., 2014). Therefore, effects of water management techniques on emissions need to be better understood for developing climate-friendly management techniques for rice production. For example, water management may strongly affect microbial communities present in the soil and thereby C and N cycling processes and their impact on field-scale GHG emissions.

Moreover, in the case of rice paddies, previous land use may strongly affect GHG fluxes and the relative importance of CH_4 with respect to other gases. Several authors reported delayed or minor CH_4 emissions when paddy fields were previously managed under aerobic conditions (Hatala et al., 2012; Brzezińska et al., 2014; Pittelkow et al., 2014). Even if methanogen communities can persist in soil during dry periods (Angel et al., 2011), a full development of methanogen communities appears to require a certain amount of time (Brzezińska et al., 2014). However, as indicated by Watanabe et al. (2011) the ecology of methanogens in soil under non-flooded condition is still not fully understood. Even less is known about nitrifier/denitrifier communities in soils under different water regimes and crops cultivation. The present manuscript therefore intends to cover this knowledge gap by specifically focusing on the impact of past management on GHG production and soil microbial communities in anoxic environments. To our knowledge no other articles specifically approached this topic on rice paddies, even if legacy effects of past management have been regarded as important for mitigating GHG emissions in croplands (Ogle et al., 2014) and microbial community composition (Jangid et al., 2011).

We hypothesized that soil history in terms of i) water management, ii) type of cultivation and iii) duration of flooding affects the microbial community composition, which requires an adaptation period in the response to new management practices.

Laboratory approaches are useful to understand soil processes because the controlled conditions allow us to analyze responses that can otherwise be masked by the high level of heterogeneity and variability encountered in the field (Schaufler et al., 2010). However, soil storage and conditioning procedures may strongly affect results, through physico-chemical and microbiological

changes. In order to reduce possible artifacts, soil microbial activity is usually determined in fresh samples (Trasar-Cepeda et al., 2000). Nevertheless, air-drying is preferred for practical reasons allowing soil to be stable during storage with minimum cost and may significantly reduce variability among soil samples collected at different moisture content (Haney et al., 2004). Air drying and sieving can cause a temporary increase of mineralization of organic matter released from broken soil aggregates (Hassink, 1992; Degens, 1998). Moreover, rewetting of air-dried soil causes slaking, i.e. increased breakdown of soil aggregates, which leads to a release of mineralizable organic matter. Thus, the comparison of different preparation methods for soil samples can provide specific information on processes affecting GHG emissions.

To assess the influence of soil management on N_2O and CH_4 potential production, four soils with different history of water management were compared, using three different soil conditioning procedures.

The objectives of this study were to i) assess the influence of water management history on GHG production potential and microbial community structure, ii) relate the microbial communities inhabiting the different soils to CH_4 and N_2O production and iii) determine the influence of different soil conditioning procedures on GHG production measurement. By means of ad hoc laboratory approach, the work aimed to present a comprehensive view of the interactions among GHG production, microorganisms involved and past management. The effect of past water management systems in rice paddies on the CH_4 and N_2O producers adaptation and consequent GHG production has rarely been addressed, as present literature mainly focuses on seasonal dynamics (Jiao et al., 2006; Ferrè et al., 2012; Breidenbach and Conrad, 2014 this last on CH_4 only).

2. Materials and methods

2.1. Experimental design

Four fields were selected at the Cantaglia experimental farm (Società Italiana Sementi-SIS, Bologna, Italy), characterized by different history of water management: PF-2 (2 years old rice paddy, permanently flooded), PF-1 (1 year old rice paddy, permanently flooded), AF-1 (1 year old rice paddy, alternately flooded and dried), NEVER (aerobic field, never flooded). From each field three replicates were collected at 0–15 cm depth.

Soils from the four fields are mesic Thapto-Histic Fluvaquent (Soil Survey Staff, 2010) and show similar texture (14% sand, 42% clay, 44% silt) and pH 8.3. Other chemical characteristics are reported in Table 1.

Three different soil conditioning procedures were compared: DRY (dried soils sieved at 2 mm), PREINCUBATED (dried soils sieved at 2 mm with 1 week preincubation 1:1.5 soil:water ratio), FRESH (undisturbed fresh soils).

2.2. GHG analysis

For each experiment a set of twelve 120 mL serum flasks were prepared with 1:1.5 soil:water ratio (v:v). The headspace air was replaced with 99% He + 1% O_2. The incubation system is a thermostated water bath at 25 °C with positions for crimp-sealed serum flasks (120 mL) with magnetic stirring. Headspace gas was sampled every 4 h by a CTC GC-PAL autosampler and a peristaltic pump and measured by a GC (Model 7890A, Agilent, Santa Clara, CA, US) equipped with 20 m wide-bore (0.53 mm diameter) Poraplot Q column, 30 m 5 Å mol sieve (0.53 mm diameter), 2 HayeSep columns for backflushing water, a thermal conductivity detector (TCD) for analysing CO_2, O_2 and N_2, an electron capture detector