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Influence of pyrite and farmyard manure on population dynamics of soil methanotroph and rice yield in saline rain-fed paddy field

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ABSTRACT

Influence of farmyard manure (FYM) and pyrite application on methanotroph population and rice yield was examined during July 2008 to October 2008 in rain-fed saline paddy field using a rice variety namely HUR-3010. Four treatment plots with three replicates were established in completely randomized block design. The experimental design consisted of (a) control, (b) FYM, (c) pyrite, and (d) FYM + pyrite. Average methanotroph population was highest in FYM + pyrite treated plot (79.0×10^5 cells g^{-1} dry soil), and lowest in control plot (23.0×10^5 cells g^{-1} dry soil). Regression analysis exhibited negative relationship of methanotroph population with EC ($R^2 = -0.937$) and NH_4^+–N ($R^2 = -0.892$). Rice yield was highest in FYM + pyrite applied plots, while lowest in the control plot. Results suggested that the application of FYM and pyrite singly or in combination significantly enhance the number of soil methanotroph as well as rice yield in the saline paddy field.

1. Introduction

A considerable increase in atmospheric methane (CH_4) during the last century is believed to be responsible for major climate disturbances (IPCC, 2001). CH_4 is the second most important greenhouse gas produced from many sources including paddy fields (Zheng et al., 2008). Before releasing into the atmosphere, the produced CH_4 is subject to oxidation by methane-oxidizing bacteria (methanotroph) in the surface soil layer and the rhizosphere. Measurement of CH_4 in paddy fields indicated that about 60–80% of the CH_4 produced during a rice growing season may be oxidized by methanotroph before it reaches to the atmosphere (Sass et al., 1991; Conrad and Rothfus, 1991). The population size of methanotrophic bacteria (MB) may be one of the important factors that governs the extent of CH_4 consumption in saline or alkaline environments, particularly, the saline rain-fed paddy fields (Khmelenina et al., 2000; Carini et al., 2005). The rain-fed saline paddy fields may assume importance in quantification of the methanotrophs, if substantial amount of CH_4 is consumed by MB in such soils. The metahnotrophic communities in aerated soils are the largest biological sink for atmospheric CH_4 (Dalal and Allen, 2008; Dorr et al., 2010). Saline paddy fields are important as salinity is known to affect most of the microbial activities. However, the information about the impact of soil salinity on population size of methanotrophs from saline paddy fields is lacking. Therefore, there are strong reasons to determine the population size of methanotrophs in the rain-fed saline paddy field.

Salinity is known to affect almost half of the world's dry lands, especially paddy fields (Srivastava et al., 2009). In India, the rice growing area is comprised of 42.4 m ha, of which about 25% is infested by salinity rising problems (Tilak et al., 2005). There is general decline in the microbial activities, soil nutrition status and productivity under the influence of soil salinity. Thus, a high salt concentration in paddy soils may lead to a decline in paddy yield and soil microbial flora (Tilak et al., 2005). Population of methanotrophic bacteria constitutes significant component of various types of environments such as saline, hyper saline, wetland paddy soils, soda lakes, tropical rice soils and lake sediments (Khmelenina et al., 2000; Eller and Frenzel, 2001; Kaluzhnaya et al., 2001; Rahalkar et al., 2009; Vishwakarma et al., 2009). However, information regarding methanotroph population in saline paddy fields is unknown despite their prominent role in mitigation of CH_4 load of the atmosphere.

Pyrite application is one of the common amendments used in reclamation of saline soils. Organic manures are considered another important management practice to maintain soil fertility and restoration of saline waste land paddy fields (Saenjan and Sributta, 2002; Yunchen et al., 2009). Watanabe et al. (2009) very recently reported that the continuous application of rice straw compost has positive effects on rice yield as well as on soil physical properties. Since the pyrite application significantly reduces the salinity strength and improves the soil properties of saline paddy fields of upper Gangetic plains (Pandey et al., 2005) and organic amendments are traditional practices for

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Table 1
The average monthly rainfall and temperature of the experimental area during the study period (July 2008 to October 2008). The meteorological data was obtained from Remote Sensing Application Centre, Uttar Pradesh (RSAC-UP), Lucknow, situated about 10 km away from the experimental sites.

Months	Mean total rainfall (mm)	Mean number of rainy days	Mean temperature (°C)	
			Daily minimum	Daily maximum
July	296.5	11.6	26.2	33.5
August	317.3	13.5	25.8	32.3
September	185.7	8.2	24.1	33.0
October	44.8	3.0	18.0	32.5

rehabilitating such disturbed soils (Supparattanapan et al., 2009), we conducted an experiment to study the effect of pyrite and farmyard manure (FYM) amendment on population size of methanotroph and rice yield in rain-fed paddy field. In present study, it is expected that pyrite and FYM application in the salt affected paddy soil may influence the number of methanotroph and paddy crop yield.

2. Materials and methods

2.1. Experimental site

This experiment was conducted on the Research Farm of the Department of Environmental Science, Babasaheb Bhimrao Ambedkar (Central) University, Lucknow (lat. 26°52′21″N, long. 80°57′20″E, 110 m msl), and lies in low land area of subtropical tract of central Uttar Pradesh. The study region has a hot subtropical climate with warm summers and cool dry winters. Summers (April–May) are quite hot with a rise in temperature up to a level of 45 °C. Winters (December–February) are cool with a maximum temperature of 21 °C and minimum limit goes as low as 4 °C or less. The normal period of onset of monsoon in this region is the 3rd to 4th week of June, which lasts up to end of September and receives a medium rainfall. About 90% of the annual rainfall is received during monsoon season, but it is highly erratic and unpredictable, at times causing drought spells of varying degrees and durations.

2.2. Experimental design, cultivation of paddy crop and agronomic variables measurements

After removing the plant debris from surface the land preparation was done manually on 28 June 2008 up to 25 cm depth. The experimental plots (3 m × 2 m dimensions) were arranged in a completely randomized block design with three replicates. A strip of 0.25 m was left to separate each plot as well as to check the exchange of solutes, soil nutrients and microorganisms between the plots. The treatments were (a) control, (b) farmyard manure (FYM) (100 t ha^{-1}), (c) pyrite (50 t ha^{-1}), and (d) FYM + pyrite [amount equivalent as in (b) and (c)]. The FYM amendment had N equivalent to 60 kg N ha^{-1}. A basal dose of NPK fertilizer at 100:70:40 kg ha^{-1} was also applied. Half of the recommended dose of nitrogen was added at the time of seed sowing and the remaining nitrogen was applied at the time of panicle initiation (75 DAS) and these treatments were applied on 25 July 2008. The FYM was incorporated superficially in the soil as manual careful homogeneous mixing by hands, while pyrite was applied superficially by broadcasting method. The rain-fed rice variety, selected for present experiment was provided by Department of Genetics and Plant Breeding, Institute of Agriculture Sciences, Banaras Hindu University, Varanasi. Seeds (3–5 per hill) were sown on 31 July 2008 by dibbling method maintaining a distance of 20 cm × 15 cm (Singh et al., 1999) and mature crop was harvested on 31 October 2008. The weeds emerging in experimental plots during the cropping period were removed manually. No irrigation was provided during the crop cycle and the only source of water for the rain-fed paddy crop was natural rainfall. For measurement of all plant growth variables (shoot height, grain yield and dry straw weight), five rice hills were removed from each experimental plot at the time of the crop harvest at maturity. The sampled plants having longest leaves were selected for the measurement of rice shoot height with a meter scale. The seed grains were removed, oven dried at 60 °C till the constant weight was achieved. For measurement of various growth variables (shoot height, grain yield and dry straw weight), rice plants were sampled from each experimental plot in triplicate and data were computed at a rate of per hectare (ha^{-1}).

2.3. Soil sampling, analysis of relevant physical soil properties and enumeration of soil methanotroph

Soil monoliths (10 × 10 × 10 cm^3) were removed randomly between the rows in triplicate from each experimental plot. The soil samples were collected at 25 days regular intervals (except last sampling) during the cropping period, and stored in polyethylene bags. The soil sampling was extended from 25 to 123 DAS. The soil samples were immediately transported to laboratory, spread over the paper sheet for air drying before the analysis. Subsequently, the air dried soil samples were passed through the sieve (mesh size 2 mm) and stored in polyethylene bags at 4 °C for subsequent analysis. Determination of moisture content or numbers of methanotroph, was carried out using freshly collected soil samples. Moisture status of soil was estimated according to Buresh (1991). The EC and pH were measured using EC and pH meters (Systronics, India) respectively as described by Srivastava et al. (2009). The Na^+ concentration of soil from different experimental plots was analyzed using a Varian Atomic Absorption Spectrometer (AA240FS) according to Srivastava et al. (2009). Extractable NH_4^+–N in the soil was estimated colorometrically in 2 M KCl extracts by Phenate method (APHA, 1985).

The number of methanotrophic bacteria (MB) was enumerated by the new Most Probable Number (MPN) technique as described by Saitoh et al. (2002). In present study, instead of using microtiter plates, culture tubes were used. This modified MPN technique gives not only the more precise estimates of small population densities of methanotroph in a paddy soil with no possibility of overestimation of methanotroph population, but also it requires less equipments, labour and is superior to conventional MPN methods (Saitoh et al., 2002). Probably a more precise and accurate method to enumerate culturable MB would be MPN method in tubes with Gas-chromatograph measurement of CH_4 consumption in the headspace gas in each tube.

2.4. Statistical procedures

Results on the effects of the treatments and duration on the population size of soil methanotrophs, soil attributes and plant growth variables were assessed by ANOVA and regression analysis using the SPSS (Version 14.0).