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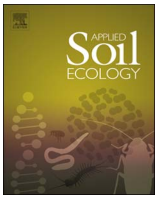

Application of mathematical models to describe rice growth and nutrients uptake in the presence of plant growth promoting microorganisms

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ABSTRACT

Two plant growth promoting microorganisms (PGPM) including *Pantoea ananatis* (KM977993) and *Piriformospora indica* were tested for their ability to improve rice (cv. 'Tarom Mahalli') growth and yield under field condition, which was arranged in a split-plot based on a randomized complete block design with three replications. The experimental soil was classified as Haplic Luvisols and Gleyic Cambisols according to WRB. Four levels of potassium sulfate fertilizer (PSF: zero, 60, 120 and 180 kg ha^{-1}) were used as the main plot and four levels of inoculation (single inoculations with *P. ananatis, P. indica*, co-inoculation and control) served as the sub-plots. Based on our knowledge, this is the first study that use a range of mathematical models to describe the effect of PGPM on rice growth and yield. The results indicated that the tiller number hill^{-1} (TNH), leaf area index (LAI), biomass dry weight (BDW), grain yield (GY), harvest index (HI), uptake of potassium (UK) in the straw and grain, HI of K, uptake of nitrogen (UN) in the grain and protein content increased by 9.0–27.2%, 11.7–45.4%, 11.1–24.7%, 13.6–30.6%, 1.25–17.6%, 1.81–27.4%, 10.8–39.9%, 7.04–12.5%, 12.3–36.8% and 3.24–5.56%, respectively, compared with the control (depending on the PGPM and PSF). The co-inoculation with *P. ananatis* and *P. indica* as the best treatment, declining the use of PSF (~40.5%) and enhancing GY (~22.6%) compared to the control. Consequently, an increase in BDW, UN and UK can be related to increasing TNH, LAI, radiation interception capacity and photosynthesis efficiency. Our findings showed, for the first time, physiological aspects of the effects both PGPM and PSF on rice growth and also recommended that PGPM can be used as biofertilizer in sustainable rice production systems.

1. Introduction

Rice (*Oryza sativa* L.) is one of the most important tropical cereals in Asia, South America and Africa. It is a staple food for about 50% of the population around the world, including Iran. Rice is grown in some areas of northern and southern Iran with an annual grain production of 2.5 million tonnes from an area of 0.59 million ha^{-1} (FAO, 2015). It is also eaten every day in some parts of Iran. Estimates indicate that rice yield should be enhanced about 65% in the world by the year 2020, especially in developing countries where it is the main food crop (Fageria, 2015).

Soil is known as an important source of nutrients and energy for all living organisms. Generally, there are a high diversity of microorganisms in rice paddies soil which play a key role in the availability of macro- and micro-nutrients that are needed for the plant (Bakhshandeh et al., 2014). Plant growth promoting microorganisms (PGPMs), as a part of microbial population in the soil, are vital components in sustainable rice production systems and also are safe for the environment. They are usually living immediately around the surface of rice root where influenced by root exudates (rhizosphere area). PGPMs can regulate the nutrient flow in the soil and influence plant growth and productivity by various mechanisms that are fully described in Ahemad and Kibret (2014) and Meena et al. (2017).

The third macro-nutrient next to nitrogen (N) and phosphorus (P) is known as potassium (K). K is absorbed by roots equal to N and or second after N in some plants like rice, cotton and banana (Fageria, 2015). However, the ability of the uptake of potassium (UK) and or K requirement by plants is varied and depending on plant species (Nieves-Cordones et al., 2014). It is available to plants about 1–2% of total K in the soil (K^+, soluble forms) while 90–98% of this is unavailable for plant uptake, as a result of the strong binding force between K and other minerals such as mica and feldspar (Bakhshandeh et al., 2017c). K is involved in many plant metabolisms such as cell division and growth, photosynthetic process, regulates the activity of stomata cells and

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production of carbohydrate, protein and oil (Fageria, 2015). It can also regulate many metabolic pathways inside the plant through enzymes activity and improve plant resistance to abiotic and biotic stresses (Bakhshandeh et al., 2017c; Meena et al., 2016). Generally, adequate values of K are necessary for a plant to reach its maximum yield potential. Furthermore, K deficiency in annual crops such as rice resulted in a slow grow and lower yields production (Bakhshandeh et al., 2017c).

To date, many researchers reported that farmers usually use large amounts of chemical fertilizers that are required for plants which can cause harmful effects on human health and environment (Bakhshandeh et al., 2015; Meena et al., 2016). In addition, increasing the cost of chemical fertilizers along with lower nutrient use efficiency (NUE) has not stopped every year which resulted in a decline farmer's income (Meena et al., 2017). Among PGPMs, several fungal and bacteria species called potassium solubilizing microorganism (KSM) that improve plant growth using solubilization of insoluble forms of K by various mechanisms like organic acid production (Meena et al., 2016). A range of most important KSMs were belonging to the *Bacillus*, *Burkholderia*, *Enterobacter*, *Paenibacillus*, *Pantoea*, *Pseudomonas* for bacteria and *Aspergillus* and *Glomus mosseae* for fungal genera (Bakhshandeh et al., 2017; Meena et al., 2016). In general, interaction between KSMs and plants (e.g., rice) determines the plant health and soil fertility. Based on previous studies, seed and seedling root inoculations with KSMs significantly increased the growth and productivity of rice through both direct (the ability of solubilizing insoluble minerals like P and K, increasing the availability of nutrients and plant hormone production) (Bakhshandeh et al., 2015; Ebrahimi-Chamani et al., 2015) and indirect mechanisms (biocontrol substances) (Agrawal et al., 2017; Shrestha et al., 2016).

Recently, the beneficial effects of PGPM/KSM on plant growth and development are well documented. For example, rice seedling root inoculation with *Piriformospora indica* enhanced biomass dry weight (BDW) and root dry weight (RDW) by 47 and 63% relative to the control (Ghabooli et al., 2015). Similarly, in rice Ashraf-Abdolahi and Zarea (2015) reported that *P. indica* increased tillers number hill^{-1} (TNH), stem dry weight (SDW) and the number of grain in panicle by 10.9, 19.1 and 7.2% in comparison with the control, respectively. *P. indica* could also improve crop yield and product quality in various crops that are well documented in Dolatabadi et al. (2011) and Andrade-Linares et al. (2013). Single inoculations of rice seed with *Pantoea ananatis*, *Rahnella aquatilis* and *Enterobacter* sp. significantly increased plant height, BDW and UK in the leaves, stem and root ranged from 10.8–15.1%, 27.4–65.3%, 35.5–76.9%, 17.6–52.9% and 25.0–75.0% (depending on the bacteria strain), respectively, as compared to the control (Bakhshandeh et al., 2017c). In other study, *Azosprilium amazonense* inoculation with rice enhanced grain yield (GY), panicles number and UN in the grain by 7–11%, 3–19% and 3.5–18.5% relative to the control, respectively (Rodrigues et al., 2008). Duarah et al. (2011) showed that application of chemical fertilizers (e.g., N, P and K) along with phosphate solubilizing bacteria (PSB) reduce the nutrient runoff or leaching and enhance NUE of the applied fertilizers in rice. Consequently, application of PGPMs alone and or in combination with chemical fertilizers can reduce the amount of chemical fertilizers consumption (up to 50%) and support eco-friendly crop production (Bakhshandeh et al., 2015; Meena et al., 2016). Furthermore, the percent changes of yields in the presence of PGPMs was reported to be ranged from 25 to 65% (depending on plant species, soil type, soil moisture content, soil pH and environmental conditions) (Meena et al., 2016) and also PGPMs are able to increase nutrient bioavailability or NUE by ~20–40% for various nutrients (Meena et al., 2017). However, application of local PGPMs could be better than others because they are competent, adapted and dominant in a particular geographical area (Bakhshandeh et al., 2014). Therefore, this study aimed to (i) determine the effects of potassium sulfate fertilizer (PSF) on rice growth and also to estimate an adequate value of PSF when GY reached its maximum using modeling approaches, (ii) determine N and K concentration and uptake of them by rice plant, (iii) evaluate the efficiency of two PGPMs namely *P. ananatis* and *P. indica* on rice growth and yield, separately and as co-inoculation, then select the best inoculation treatment. Based on our knowledge, this is the first study that use a range of mathematical models to describe the effect of PGPMs on the growth and yield of rice. These models are easier to understand and its parameters are easier to interpret physiologically.

2. Materials and methods

2.1. Field experiment

A field experiment was conducted in Mazandaran province (located at 36°33′N, 53°E and 25.7 m above sea level) for evaluating the efficiency of two PGPMs on the growth and yield of rice (cv. 'Tahrom Mahalli', known for its high grain quality which was cultivated in northern Iran) under different levels of PSF (K_2SO_4, contained at least 44% soluble K). Two PGPMs were tested: *P. ananatis* with an accession number of KM977993 as proposed by Bakhshandeh et al. (2014) and endophyte fungus *P. indica*, which were provided by Genetics and Agricultural Biotechnology Institute of Tabarestan (GABIT), Sari, Iran. Based on previous studies, *P. ananatis* was a multiple plant growth promoting rhizobacteria because it was able to solubilize insoluble P ($Ca_3(PO_4)_2$, 172 μg ml^{-1} after 5 days at 28 °C), K (mica, 38.9 μg ml^{-1} after 25 days at 28 °C) and produce indole-3-acetic acid (IAA, 8 μg ml^{-1} after 3 days at 28 °C) (Bakhshandeh et al., 2014, 2017c). *P. indica* was also known as an effective plant growth promoting mycorrhizal fungus that its characteristics are fully described in Varma et al. (2012). In this study, *P. ananatis* was grown in nutrient broth (NB; Scharlau, Spain; 8 g l^{-1}) medium as followed Bakhshandeh et al. (2015) to a final density of 10^7 CFU ml^{-1} and also *P. indica* was grown in Kafer medium to a final density of 10^9 CFU ml^{-1} as in Sherameti et al. (2005). The experiment was carried out as a split plot arrangement based on a randomized complete block design with three replications. Four levels of PSF (zero, 60, 120 and 180 kg ha^{-1}) were used as the main plot and four PGPM inoculations, namely, single inoculations with *P. ananatis*, *P. indica*, co-inoculation (*P. ananatis* + *P. indica*) and non-inoculated as a control, served as the sub plots. On the other hand, each main plot size was 10 m long and 6 m width (60 m^2) related to PSF levels and then different subplots (each subplot was 6 m long × 2.5 m width = 15 m^2) related to PGPM inoculations were defined within each of the main plots. Rice seedlings were planted with a row spacing of 20 cm, included 12 rows with four seedling hill^{-1} in each subplot.

2.2. Inoculation treatments and transplanting condition

Rice seeds were dipped in the water for 24 h at the beginning of the experiment and then drained off the water and kept in warm condition for 5 days. Pre-germinated seeds were grown in the nursery for 35 days by keeping under optimal agronomic conditions based on local recommendations. Roots of rice seedling (25 cm in height with 4–5 fully leaves) were inoculated with each PGPM suspension, which was diluted in water to a final density of 10^6 CFU ml^{-1} for *P. ananatis* and 10^8 CFU ml^{-1} for *P. indica*, and then applied by dipping the seedling roots into the suspension, for 12 h at room temperature before transplanting in the paddy field on 6 May 2016. Roots of rice seedling were also treated in the same manner with non-inoculated mediums as a control. The paddy field was fallow last year.

2.3. Soil characteristics

Some physiochemical properties of the soil (at a depth of 0–30 cm, sampled by a soil auger with 10 cm diameter), measured by the Joibar Soil and Water laboratory were: 1.02% organic carbon; pH 7.75; electrical conductivity (EC) of 3.15 dS m^{-1}; 14.2 and 92 mg kg^{-1} available