



Mechanisms of enhanced inorganic phosphorus accumulation by periphyton in paddy fields as affected by calcium and ferrous ions



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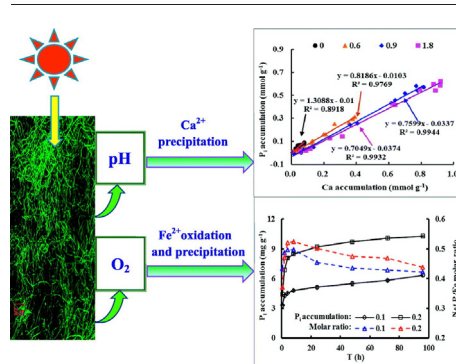
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HIGHLIGHTS

- Calcium and ferrous ions promoted inorganic P (P_i) accumulation on the periphyton.
- P precipitation and coprecipitation with carbonates or $Fe(OH)_3$ were main mechanisms.
- Efficiency of $Fe(II)$ was higher than $Ca(II)$ in enhancing P_i abiotic accumulation.
- Periphyton would help reduce P fixation in soil and P loss from the paddy fields.

GRAPHICAL ABSTRACT



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ABSTRACT

The effect of periphyton propagation in paddy fields on phosphorus biogeochemical cycling has received little attention. In this phytotron study, inorganic phosphorus (P_i) accumulation by periphyton was investigated for varying inputs of calcium [$Ca(II)$] or ferrous iron [$Fe(II)$], and lighting conditions. Results indicated that additions of $Ca(II)$ or $Fe(II)$ enhanced abiotic accumulation of P_i by up to 16 times, and decreased solution P_i concentration by up to 50%, especially under light condition. The enhanced P_i accumulation into periphyton intensified with increasing P_i concentration, and P_i accumulation showed a positive linear relationship with Ca or Fe accumulation. Abiotic accumulation of P_i induced by $Ca(II)$ was mainly through Ca-phosphate precipitation, and co-precipitation of P with carbonates at $pH > 8$. Accumulation with added $Fe(II)$ was mainly considered to be through $Fe(III)$ phosphate precipitation coupled with adsorption of P_i by ferric hydroxides. Moreover, $Fe(II)$ was more effective than $Ca(II)$ in promoting abiotic accumulation of P_i by periphyton. Our results indicate the potential for controlling environmental factors to enhance the role of periphyton in biogeochemical cycling and P-use efficiency in paddy rice fields and to reduce P discharged to neighboring water bodies.

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1. Introduction

Paddy fields comprise the largest anthropogenic wetland ecosystem, and they subjected to intensive human activities (Kögel-Knabner et al., 2010). Rice is the staple food for more than half of the world's population and phosphorus (P) is a limiting nutrient for rice production

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(Fageria et al., 2011; Raghothama, 1999). Farmers commonly apply a large base dose of P fertilizer before planting rice (Ayaga et al., 2006; Lan et al., 2012). However, since P uptake by rice is minimal during the seedling stage, excess fertilizer P is either fixed by soil particles, such as iron (Fe) and aluminum (Al) oxides/hydroxides and calcium (Ca) bounded carbonates, or leached out of paddy fields (Arai and Sparks, 2007; Lgwe et al., 2010; Lan et al., 2012). Then, during the critical flowering and maturation stages of high P demand, plant-available P to rice is low. Consequently, P use efficiency is low in paddy fields, and these fields are an important nonpoint source of P runoff (MacDonald et al., 2011; Yang and Lu, 2014; Zhou and Zhu, 2003). Better management strategies based on a mechanistic understanding of P biogeochemical processes are needed to improve P use efficiency and reduce P discharged from paddy fields.

Periphyton is ubiquitous in paddy fields because light conditions, water, temperature, and nutrients are suitable for periphyton to proliferate in the early stage of rice growth (Hagerthey et al., 2012; Kirk, 2004; Yang et al., 2016). Periphyton is an under-water micro-floral community that attaches to substrates such as submerged plants or plant parts, rocks, and sediments (Larned, 2010). Its components include algae, bacteria, protozoa, metazoans, epiphytes associated with inorganic minerals such as Fe and Al hydroxides, and Ca/Mg carbonates (Wu et al., 2012, 2014). Periphyton can act as a potential P sink in wetlands and is also regarded as a buffer for P precipitation and release between sediments and water (Dodds, 2003; Drake et al., 2012; Lu et al., 2016a; Wu et al., 2011; Zamalloa et al., 2012). Phosphorus is normally in excess for rice during the early stages of growth (Ayaga et al., 2006; Lan et al., 2012). In this stage, the periphyton microorganisms thrive under sufficient light and could potentially accumulate P, which should reduce P fixation by soils and P discharged to the neighboring water bodies. Periphyton would decompose and potentially release P as the canopy cover fills during later stages of rice growth (Drake et al., 2012). Hence, the timing of biogeochemical P cycling through periphyton could be in synchrony with the timing of crop needs, thereby improving P use efficiency and reducing P loss from the paddy fields. However, this potentially beneficial effect of periphyton on P cycling in paddy fields has received little attention.

Periphyton can modify the local chemical environment and retain P by biotic as well as abiotic accumulation (Scinto and Reddy, 2003). The localized pH in the zone of actively photosynthesizing periphyton increases by up to 1 unit, which favors Ca-phosphate precipitation, and concurrent deposition of carbonate-phosphate complexes (Woodruff et al., 1999). Moreover, periphyton can cause super-saturated O₂ concentrations near the soil surface through photosynthesis, which encourages deposition of metal phosphates (Dodds, 2003). Pei et al. (2015) indicated that abiotic accumulation of Ca-P and Fe/Al-P constituted approximately 75% of the total periphyton P in different lakes. Lu et al. (2016b) also reported that >90% of P removal by the periphyton from wastewater was contributed by Ca-phosphate precipitation and adsorption of phosphate onto CaCO₃. Thus the adsorption or co-precipitation of P with metal salts (especially with CaCO₃) was the main P removal mechanism from wastewaters (Dodds, 2003; Lu et al., 2014; Scinto and Reddy, 2003).

Inorganic ions of Ca and Fe are very important for abiotic P accumulation. In paddy fields, Ca(II) is one of the main cations in soil solution because human activities, such as applying calcium magnesium phosphates and liming agents (e.g., CaCO₃), add Ca(II) to the paddy fields. Co-precipitation of phosphate during calcite formation is generally considered as an important “self-cleaning” mechanism of freshwaters by algal biofilm (House, 1990; Jarvie et al., 2002). However, high inorganic phosphate can also inhibit calcite precipitation, which depends on the phosphate concentration and the extent of supersaturation of the solution with respect to calcite (House, 1987; Jarvie et al., 2002; Reddy, 1977). Although much research has focused on freshwaters with low P concentrations or wastewaters with high P concentrations (Jarvie et al., 2002; Lu et al., 2014; Posadas et al., 2013; Scinto and Reddy,

2003), little information is available on P biogeochemical cycling affected by periphyton in paddy fields containing moderate P concentration. Moreover, water management usually results in drying and wetting cycles and dramatic redox variations in paddy fields. Oxidation and reduction of Fe is particularly dynamic, with Fe(II) widely occurring in reduced paddy fields when submerged with water (Kirk, 2004; Kögel-Knabner et al., 2010). The effect of Fe(II) on P biogeochemical cycling in the presence of the periphyton is not clear. Moreover, periphyton photosynthesis during day-light hours and respiration mainly at night would modify the local soil chemical environment differently.

Given these many chemical variables that could affect periphyton accumulation and release of P, the objectives of this study were to: (i) determine the effect of Ca(II) and Fe(II) additions on P accumulation by periphyton under light and dark conditions, (ii) compare the efficiency of Ca(II) and Fe(II) in promoting P accumulation, and (iii) infer underlying mechanisms responsible for P accumulation by periphyton induced by Ca(II) and Fe(II) in paddy fields. The results obtained in this study should improve our understanding of the role of periphyton in P biogeochemical cycling in the paddy fields, and consequently provide insights to management practices that could increase P-fertilizer use efficiency and reduce the impacts of P to the environment.

2. Materials and methods

2.1. Periphyton culture

Periphyton was cultured from a water sample collected at Pullen Park Lake, a eutrophic lake in Raleigh of North Carolina, USA. The water was incubated in two glass fish tanks of 60 × 25 × 30 cm (l × w × h) under controlled temperature and light conditions in a phytotron. Polypropylene fiber carriers (FCs, Jineng Environmental Protection Company of Yixing, China) were used as a substrate for periphyton growth and enrichment. The FCs were disinfected using 95% alcohol solution and rinsed with deionized water. The FCs were submerged into the 45 L fish tanks filled with 30 L of lake water, and modified BG-II medium composed of the following minerals was added: 16 mg L⁻¹ Na₂CO₃, 85.1 mg L⁻¹ NaNO₃, 8.71 mg L⁻¹ K₂HPO₄, 37 mg L⁻¹ MgSO₄·7H₂O, 36 mg L⁻¹ CaCl₂·2H₂O, 2.86 mg L⁻¹ H₃BO₃, 0.18 mg L⁻¹ MnCl₂·4H₂O, 4.36 mg L⁻¹ Na₂EDTA·2H₂O, 3.15 mg L⁻¹ FeCl₃·6H₂O; trace elements of 0.22 mg L⁻¹ ZnSO₄, 0.39 mg L⁻¹ Na₂MoO₄, 0.079 mg L⁻¹ CuSO₄·5H₂O, 0.012 mg L⁻¹ Co(NO₃)₂·6H₂O; and vitamins of 0.13 mg L⁻¹ Vitamin B12, 0.33 mg L⁻¹ Thiamin, 0.02 mg L⁻¹ Biotin. The fish tanks with FCs were incubated for 2 months in a manually controlled climatic (phytotron) box and exposed to a light/dark cycle of 14 h (28 °C)/10 h (20 °C), a light intensity of 12,000 lx, and a humidity 80%. At the end of the incubation, the periphyton colonized on the FCs were removed from the fish tanks and used for the experiments described below.

2.2. Periphyton characterization

The morphology of the phototrophic periphyton was characterized by optical microscopy (OM, Nilcon-80i, Japan) at 400× magnification, scanning electron microscopy (SEM, Quanta200, FEI, Netherlands), confocal laser scanning microscopy (CLSM, Zeiss-LSM710, German), and XRD across an angle range of 0 to 70° 2θ (D/max-^oC, Rigaku Corporation, Japan). Periphyton biomasses used throughout this study were on an oven-dry basis, for which hydrated periphyton were dried at 105 °C for 24 h.

2.3. Inorganic P accumulation with batch experiments

Parameters that were evaluated in the overall set of experiments described below are summarized in Table 1.

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