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Growth of Celery (*Apium graveolens* var. dulce) as Influenced by Phosphite

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Although Phosphite (PO_3; Phi) has recently been marketed as "superior P fertilizer" for a wide range of crops, the claim regarding its potential as a fertilizer is controversial. In this study, effects of Phi at both low (0.2 mM) and high (2 mM) rates on growth and phosphorus (P) supply of celery (*Apium graveolens* var. dulce) were investigated under either low phosphate (PO_4; Pi) (0.1 mM) or high Pi (0.5 mM) supply. The effect of Phi was found to be strongly dependent on the P nutrient status of the plants. Addition of Phi at both rates did not influence growth of high Pi–supplied plants. However, Phi at 2 mM significantly decreased both shoot and root growth of low Pi–supplied plants. Plants grown in this treatment showed bleaching of young leaves and death of root tips and root hairs as typical toxic symptoms of Phi. Shoot P concentration was highly increased by root application of Phi, suggesting that Phi was well absorbed by roots and mobile inside the plants but also corroborating that Phi did not provide P nutrition to celery plants. No beneficial effect on plant growth was detected from the use of this material.

INTRODUCTION

Phosphite (PO_3; Phi), a reduced form of phosphate (PO_4; Pi), is recently widely marketed either as fungicide or as a superior source of plant phosphorus (P) nutrient (McDonald *et al*., 2001; Rickard, 2000). Although it is commonly accepted that Phi products are excellent fungicides, the claim regarding their potential as a fertilizer is controversial. Numerous studies indicated that Phi has negative effects on growth and development of plants and did not serve as a source of plant nutrient (Carswell *et al*., 1996, 1997; Forster *et al*., 1998, Abel *et al*., 2002; Schroetter *et al*., 2006), but rather it interferes with Pi starvation responses of plants (Ticconi *et al*., 2001; Varadarajan *et al*., 2002; Carswell *et al*., 1996, 1997; Abel *et al*., 2002). In contrast, there were also evidences that Phi application could replace Pi in some plants such as citrus and avocado crops suffering from P deficiency (Lovatt, 1990a; 1990b). Beneficial effects of Phi on growth of cucumber and on the yield and quality of Satsuma orange in Japan (Watanabe, 2005), Valencia orange in Florida (Albrigo, 1999) were also reported. Rickard (2000) summarized studies on commercial Phi fertilizers and concluded that these materials produced consistent improvements in yield and quality for many different crops. Although the effects of Phi on plant growth are still being debated, crop producers in many countries are applying Phi formulations that are labeled as P fertilizers (McDonald *et al*., 2001; Rickard, 2000). Interest in using Phi as part of a total production package is also increasing (Lovatt and Mikkelsen, 2006). Some researchers (Rickard, 2000; Lovatt and Mikkelsen, 2006) suggested that the beneficial effects of Phi were not related to the molecule's fungicidal properties but to other growth–stimulating potential. The negative effects of Phi in many studies were claimed due to inappropriate use of this material, such as use as the sole source of P or in excessive amounts (Rickard, 2000; Watanabe, 2005; Lovatt and Mikkelsen, 2006).

This research therefore was conducted to investigate the potential effects of Phi as a P fertilizer on growth and nutrition supply of celery plant, a high P requirement crop.

MATERIALS AND METHODS

Seedling preparation

Celery seeds (*Apium graveolens* var. dulce cv. Mini White) obtained from Takii & Co. Ltd., were germinated in vermiculite in a 20 ˚C–controlled room. Seedlings were watered daily and received a half–strength Hoagland solution twice per week. To guarantee the uniformity of the seedlings and to reduce the shock to the plants during the transfer to the hydroponic treatment solutions, after growing in vermiculite for 5 weeks, the seedlings were transplanted into aerated–25%–strength Hoagland solution (pH 6.0) in 4–L hydroponic culture containers for 6 days of pre–treatment.

Experimental design and cultivation

The experiment was carried out from June 8th to July 8th, 2007 in the same room. The experiment had a total of 6 treatments, a combination of two Pi levels (0.05 mM as deficient level and 0.5 mM as sufficient level) and three Phi levels (0, 0.2 and 2 mM), arranged in completely randomized design with three replications. After pre–treatment, uniformed seedlings were selected and transferred into 7–L hydroponic pots (6 plants pot^{-1}) (= day 1 of the experiment) containing the same Hoagland solution (pH 6.0) but P was modified according to the treatments. The nutrient solutions renewal was adjusted

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from every 7 days to every 5 days as the plants grew up and the pH was maintained from 6.0 to 6.5 by addition of 1 M H_2SO_4 or 1 M NaOH as needed. In hydroponic system (both pre–treatment and treatment period), the plants are suspended from a floating platform, and the roots hang down into the aerated nutrient solutions. Plants were harvested at 30 days after transplanting into the treatment solutions.

Plant analysis

Determination of dry weight: After harvesting, plants were separated into shoots and roots, washed in running tap water and then in deionized water. The dry weight (DW) of shoots and roots were obtained after drying samples for 3 days at 70 °C.

Determination of total P: The dried, ground plant samples were digested using an H_2SO_4–H_2O_2 Kjeldahl digestion method, designed for nitrate–containing samples (Ohyama *et al*., 1991), before measurement of P by the colorimetric procedure of Murphy and Riley (1962).

Statistical analysis

Treatment effects were assessed by analysis of variance (ANOVA) using IRRISTAT for Windows version 4.0 (Biometric Unit, International Rice Research Institute). Mean separation was performed using the least significant difference (LSD) at P=0.05 and P=0.01.

RESULTS

There was highly significant difference in shoot DW between low Pi– and high Pi–supplies (P<0.001). Fig. 1 shows a 2–fold difference in shoot DW (averaged over the three Phi levels) between low Pi– and high Pi–supplied plants. The response of plants to Phi was found different between two Pi levels. Addition of Phi did not influence shoot DW of high Pi–fertilized plants, these plants grew healthy and normally up to 2 mM Phi. However, under low Pi–supply, shoot DW of plants tended to decrease with addition of Phi. The decrease in shoot DW was up to 20% as Phi addition at the rate of 2 mM. Plants grown in this treatment showed Phi toxic symptoms as bleaching of the young leaves (data not shown).

The same trend was found in root growth, as shown

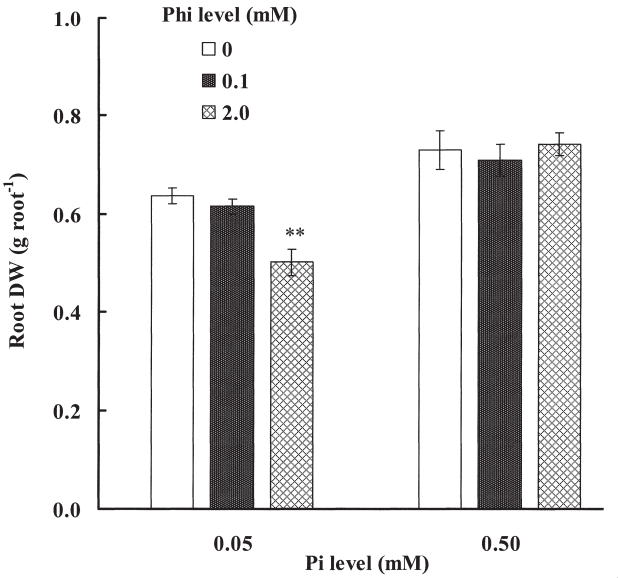

Fig. 2. Effect of different Phi levels on root growth of celery under low Pi– and high Pi–supplies. Bars represent ± SEM (n=3). ** indicates significant differences (P<0.01) from the no Phi treatment under the same Pi level.

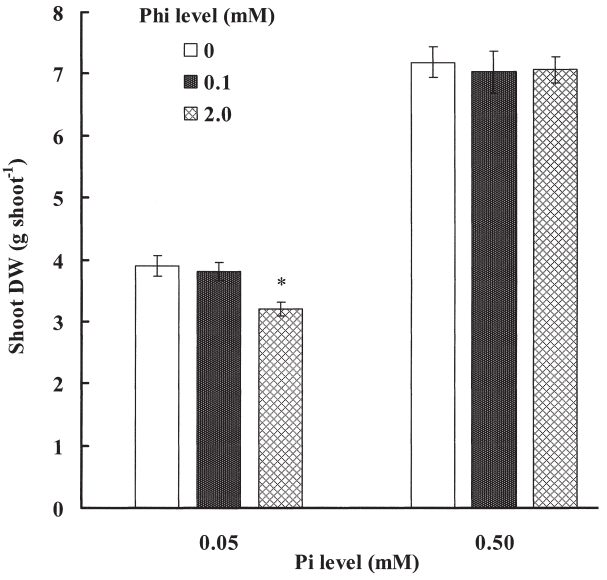

Fig. 1. Effect of different Phi levels on shoot growth of celery under low Pi– and high Pi–supplies. Bars represent ± standard error of the mean (SEM) (n=3). * indicates significant differences (P<0.05) from the no Phi treatment under the same Pi level.

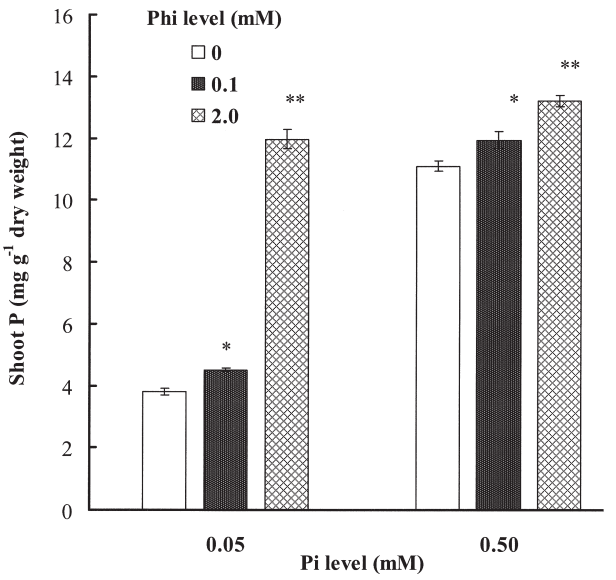

Fig. 3. Effect of different Phi levels on shoot P concentration of celery under low Pi– and high Pi–supplies. Bars represent ± SEM (n=3). Under each Pi level, significant differences from the no Phi treatment at P<0.05 (*) and P<0.01 (**) are shown.

in Fig. 2, under the high Pi–supply, the presence of Phi did not influence root DW. However, under low Pi– supply, application of Phi at 2 mM significantly decreased root DW compared to no Phi treatment ($P<0.05$). The roots of low Pi–fertilized plants treated with 2 mM Phi showed many black spots on the root system and the death of root hairs and root tips (data not shown).

Shoot P concentration was found to increase significantly by application of Phi at both levels. Under low Pi–supply, the increase in shoot P concentration over the control was 19% (for 0.1 mM Phi) and 215% (for 2 mM Phi); whereas under high Pi– supply, they were 8% and 19%, respectively (Fig. 3).

DISCUSSIONS

Although there were numerous studies carried out to determine effects of Phi on plant growth, the results are contradictory. Some researchers (Rickard 2000; Lovatt and Mikkelsen, 2006) claimed that the beneficial effects of Phi were not related to the molecule's fungicidal properties but to other growth–stimulating potential. However, some others (MacIntire *et al*., 1950; Ohtake *et al*., 1996) suggested that the observed nutritional effect of Phi are likely due to indirectly providing P to the plant after its oxidation to Pi in the soil by microbes. The Benejicial effects of Phi on plants in the field trial may also result from other phenomenon, such as Phi's suppression of plant pathogens (McDonald *et al*., 2001). In this research, the experiment therefore was carried out in the controlled room using hydroponic system to minimize the oxidation of Phi to Pi during the experiment as well as the interfering effect of pathogen which usually happened in the field. Our results clearly demonstrated that application of Phi at both levels (0.1 and 2 mM) did not improve plant grown under either Pi deficient or Pi sufficient levels. The effect of Phi on plant growth was strongly dependent on P nutrient status of the plant. High Pi–fertilized plants grew well and normally up to the Phi level of 2 mM whereas for low Pi–fertilized plants, Phi at 2 mM significantly decreased both shoot and root growth (Fig. 1 and 2). The decrease in shoot growth of Pi–deficient plants by Phi was associated with the suppression of root growth, suggesting that the inhibition effect of Phi on root growth and hence the nutrition uptake of roots could be the main reason to contribute to shoot growth reduction of these plants. The results are in agreement with some other studies emphasizing that Phi's phytotoxicity was only evident to Pi–starved but not to Pi–fertilized *Brassica nigra* seedlings (Carwell *et al*., 1996), *Brassica napus* (oilseed or canola) suspension cells (Carwell *et al*., 1997). The negative effects of Phi were suppressed in plants with sufficient Pi. This is likely to be due to a combination of reduced uptake of Phi or its inability to interfere with biochemical reactions in the presence of Pi (Varadarajan *et al*., 2002).

The highly increased shoot P concentration by root application of Phi under both low and high Pi supplies indicated that Phi was well absorbed by roots and mobile in celery plants. Phi was also reported to be well absorbed by roots and mobile in other plants such as *Brassica nigra* (Carwell *et al*., 1996) and maize (Schroetter *et al*., 2006). However, plant species may differ in uptake and translocation of Phi. Our previous research on spinach plants provided evidence that Phi was poorly absorbed by roots of this plants (Thao *et al*., 2008b).

The growth of celery plants were strongly increased in high Pi supply compared to low Pi supply, confirming that the plants in low Pi level suffered from P starvation and therefore strongly responded to P nutrition. However, the increase in shoot P concentration by addition of Phi at either low or high rate did not improve growth of the low Pi–supplied plants. This corroborates evidence that Phi was not significantly oxidized or otherwise metabolized in plants. These results support earlier studies indicated that Phi did not provide P nutrition to *Brassica nigra* (Carwell *et al*.,1996), tomato and pepper (Forster *et al*., 1998), maize (Schroetter *et al*., 2006), komatsuna (Thao *et al*., 2008a) and spinach (Thao *et al*., 2008b).

We conclude that Phi was well absorbed by roots and mobile inside celery plants but did not provide P nutrition to the plants. Phi had also no beneficial effect on growth of celery plants under either P sufficient or P deficient but it rather suppressed both shoot and root growth of Pi–deficient plants.

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