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Research Note

Interactive effects of salicylic acid and silicon on some physiological responses of cadmium-stressed maize seedlings

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Abstract

The impacts of salicylic acid and silicon on some physiological parameters of maize seedlings in the presence of cadmium (100 µM) were investigated. Inhibitory effects of cadmium on seedling growth resulted in reduced shoot and root fresh weight, low percentage of relative water content, low chlorophyll, free proline and soluble sugars contents and a low rate of lipid peroxidation. Results indicated that salicylic acid and silicon alleviate the inhibitory effects of cadmium on maize seedlings by increasing both their chlorophyll content and fresh weight. Although individual treatments of salicylic acid and silicon reduced plants free proline, soluble sugars and cadmium uptake and lipid peroxidation rate, they improved root and shoot fresh weights in both cadmium stressed and unstressed seedlings. When combined, salicylic acid and silicon alleviated the inhibitory effects of cadmium on seedlings significantly.

Keywords: Cadmium; maize; physiological response; salicylic acid; silicon

1. Introduction

Cadmium is not an essential element for plant growth [1] and is considered as an extremely significant pollutant due to its high water solubility and its toxicity effects on living organisms [2]. Its uptake and accumulation in plants pose a serious health hazard to humans and living cells via the food chain [3-5]. Cd inhibits photosynthesis [6], transpiration [7], carbohydrate metabolism [8], and other metabolic activities [9].

Silicon (Si) is the second most abundant element in the earth crust, yet its role in plant physiology has been poorly understood and attempts to associate Si with metabolic or physiological activities have been inconclusive [10]. Although Si has not been classified as an essential element for higher plants, it has been shown to be beneficial for plant growth [11, 12]. Silicon has been shown to ameliorate the adverse effects of heavy metals on plants.

Salicylic acid (SA) is an endogenous growth regulator with many physiological functions. It is implicated in hardening responses to abiotic stressors and mediates some positive acclimation response to abiotic stresses, such as heavy metals, herbicides, low temperatures, and salinity [13-17].

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The main objective of the present study was to investigate the interactive effects of SA and Si on the alterations of some physiological parameters observed in Cd-stressed maize seedlings.

2. Materials and methods

2.1. Plant materials and treatments

Seeds of *Zea mays* (var KSC.704) were surface sterilized by 5% (w/v) sodium hypochlorite for 20 min and then divided into two groups. One-half of the seeds were presoaked in salicylic acid solution (500µM) for 6 h and then both groups were allowed to germinate for 3 days at 24° C in petri dishes under sterile conditions. After germination, the germinated seeds were transferred to pots containing a mixture of sand and perlite (1/1, w/w). The seedlings were allowed to grow for another 6 days in a greenhouse at 27/18 °C (day/night), with a 16h light/8h dark photoperiod and 60% relative humidity and were irrigated with nutrient solution. Eight different treatments with three replicates were used during a seven-day experiment as follows: control, Cd, SA, Si, SA+Si, Cd+SA, Cd+Si, and Cd+SA+Si. The concentrations of Cd and Si used were 100 µM and 50 mM, respectively. Cd and Si were added as ($CdN_2O_6.4H_2O$) and ($Na_2SiO_3.9H_2O$) respectively, and were added to the growth media on day 6; fully developed leaves were used for biochemical analysis . Shoots and roots fresh weights were used for analysis of plants growth. Root and shoot FW were shown as g/pot (each pot contained three seedlings).

2.2. Lipid peroxidation measurement

Lipid peroxidation was assayed [18] and calculated [19] as the references.

2.3. Proline, soluble sugars and chlorophyll measurement

Free proline and soluble sugars in 0.1 g of leaf samples were analyzed according to Bates [20] and Nelson [21] methods, respectively. Leaf chlorophyll was extracted by acetone and measured spectrophotometrically using Arnon's equation [22].

2.4. Cadmium uptake

For determination of Cd, the harvested plant samples were rinsed with deionized water and then oven dried at 70°C for 48 h. The dried material was ashed at 550 °C for 24 h. The ash residue was kept in 65% HNO3 for 4 h, and then more HNO3 was added until a clear solution was obtained. The amount of Cd was determined using an atomic absorption spectrophotometer (Varian, spectra AA-220 model) according to Wickliff method [23].

2.5. Statistical analysis and computations

The experimental designs were randomized complete block with three replicates. The collected data was imported to Microsoft Excel program for calculations and graphical representation. SPSS software (version 17.0) was used for analysis of variance. The means were compared using Duncan's multiple range tests at $P < 0.05$.

3. Results and discussion

Treatment of maize seedlings with 100-μM Cd reduced both shoots and roots fresh weight significantly (Figs. 1 and 2). The reduction in root fresh weight was more than that of the shoots, which could be due to the higher accumulation of cd in root tissues (Fig. 3). Those reported by Blum [24] and by Liang et al. [13] support our results. Cd is easily taken up by the roots tissues and is accumulated in both roots and shoots.
At the cellular level, Cd exerts toxic effects on cell metabolism. Preventation of Cd uptake by plant roots alleviates plant tissues from the adverse effects of this toxic metal [25].

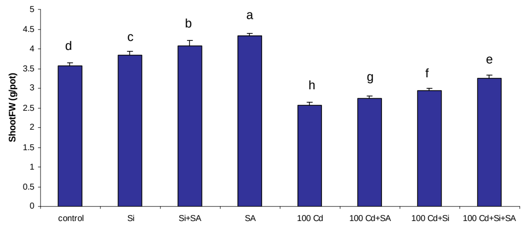

Fig. 1. Effects of Si, Si+SA and SA in the presence and absence of Cd (100 μm) on maize seedlings shoot fresh weight

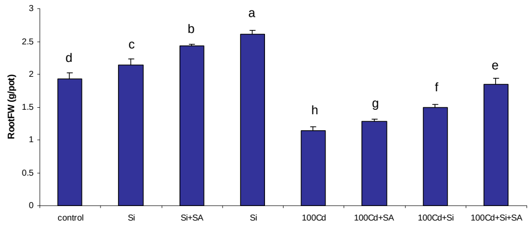

Fig. 2. Effects of Si, Si+SA and SA in the presence and absence of Cd (100 μm) on maize seedlings root fresh weight

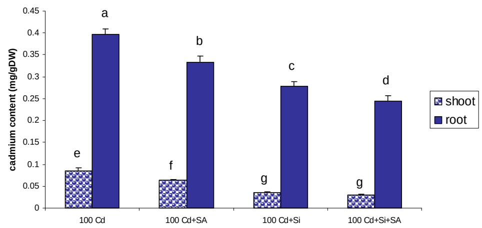

Fig. 3. Effects of SA, Si and Si+SA and on the cadmium content of roots and shoots of maize seedlings (seedling were treated with 100 μm Cd)

In our study the presence of SA, Si and SA+Si reduced the adverse effects of Cd on roots and shoots fresh weight significantly (Figs. 1, 2 and 3). Chlorophyll pigments, as the main component of photosynthetic apparatus, play a pivotal role in plant metabolism and energy supply. In our study, Cd treatments reduced the amount of chlorophyll significantly (Fig. 4).

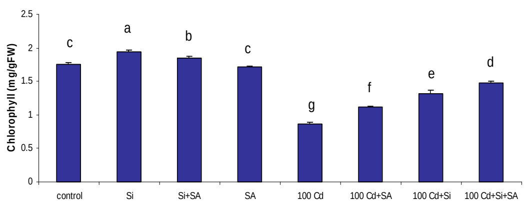

Fig. 4. Effects of Si, Si+SA and SA in the presence and absence of Cd (100 μm) on maize seedlings leaves chlorophyll content

However, in the presence of SA, Si and SA+Si, the inhibitory effects of Cd on leaves chlorophyll content was considerably reduced (Fig. 4). Parasad et al. [26] have reported that high Cd levels, by interfering with protochlorophyllide biosynthesis in *Riccia* sp. inhibit the leaves chlorophyll content. Inhibition of carotenoid pigments as protectants of photodynamic damage to chloroplasts apparatus by Cd could also result in less leaves chlorophyll content [27]. Heavy metals are known to induce the production of reactive oxygen species (ROS) in higher plants [28]. Cd treatment increased plants malondialdehyde (MDA) content by 72.7%. However, in the presence of Si, SA and Si+SA, the amounts of MDA content reduced by 24.7, 10 and 10% respectively (Fig. 5).

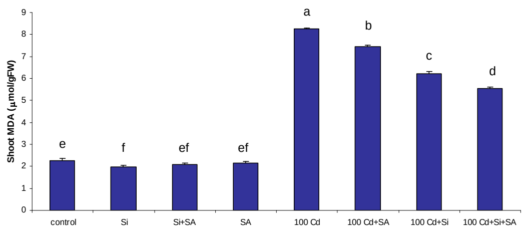

Fig. 5. Effects of Si, Si+SA and SA in the presence and absence of Cd (100 μm) on maize seedlings MDA content

MDA is the decomposition product of polyunsaturated fatty acids of biomembrane induced by ROS and its accumulation is an indication of oxidative stress in plants [29] in response to Cd treatment. The amount of soluble sugars in the leaves of plants treated with Cd increased by 41.4% as compared to control. In the presence of Si, SA and Si+SA, the amounts of soluble sugars in Cd-stressed plants decreased by 19, 9 and 23.2% respectively (Fig. 6).

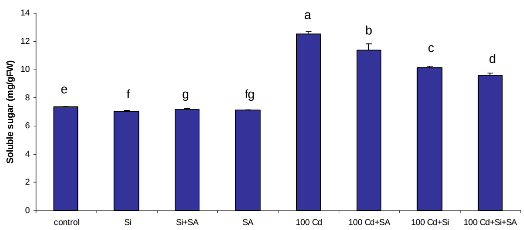

Fig. 6. Effects of Si, Si+SA and SA in the presence and absence of Cd (100 μm) on maize seedlings leaves soluble sugar content

There was a 60% increase in the proline content of Cd-stressed plants. However, Si, SA and Si+SA treatments reduced the amount of proline in Cd – stressed plants by 41.2, 9.67 and 52.47% respectively (Fig. 7). Proline also accumulates in higher plants in response to ROS production and protects plants against the oxidative damage caused by free radicals [30]. Proline is also known to activate the Krebs cycle reactions, thus enhancing the plants energy turnover [31]. Pretreatment of seeds with SA is known to protect plants against the inhibitory effects of Cd on growth, photosynthesis, and chlorophyll content and against the damage caused by Cd [17].

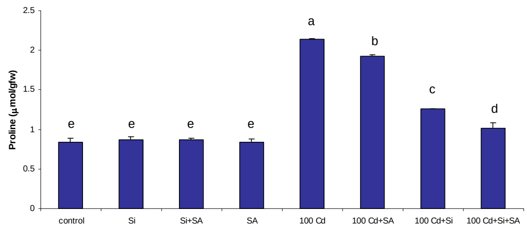

Fig. 7. Effects of Si, Si+SA and SA in the presence and absence of Cd (100 μm) on maize seedlings leaves proline content

It is suggested that the beneficial effects of SA during the early growth stages could be due to its role in preventing the accumulation of free radicals [17]. However, Metwally and co-workers [14] reported that SA alleviates Cd toxicity not at the level of antioxidant defense, but by affecting other mechanisms of Cd detoxification. Deposition of Si in several parts of a plant, especially in root reduces the apoplastic flow of Cd and provides binding sites for metals, resulting in a decrease uptake and translocation of toxic metals from roots to shoots. Si-alleviated effects have been associated with an increase in its antioxidant defense abilities [32, 33]. Data presented in figs. 1 to 7 indicate that although Si and SA can individually protect maize seedlings from the adverse effects of Cd on maize seedlings physiological parameters, the combination of Si and SA have more alleviating effects on Cd toxicity. In addition, Si and SA in the absence of Cd treatment have some beneficial effects on maize seedlings growth (figs. 1, 2 and 4).

Acknowledgments

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