

Glycine-chelated zinc lowered foliar phytotoxicity than excess zinc sulfate and improved zinc use efficiency in two sweet potato cultivars

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ABSTRACT

Although applying widely used ZnSO_4 at high spraying rates provides high Zn concentration in some crops, it usually causes leaf burn and yield and fertilizer losses. Whether chelated Zn fertilizers can improve Zn concentration in edible roots of sweet potato while concomitantly avoiding apparent adverse effects deserves detailed study. Field experiments were conducted in 2019 and 2020 on two sweet potato cultivars to study the effects of foliar spraying of three typical Zn sources (i.e. zinc sulfate heptahydrate [ZnSO_4], EDTA-chelated zinc [ZnEDTA], and glycine-chelated zinc [ZnGly]) at normal to high spraying Zn rates of 1.19, 2.39, 4.78 and 7.16 kg ha^{-1} . The results showed that excess ZnSO_4 spray caused greater damage to the leaves of cultivar Tianfeng1 than to those of cultivar Pushu90, whereas both chelated Zn fertilizers reduced the extent of foliar burn on the two cultivars. ZnGly achieved a higher biofortification level than ZnEDTA and higher yield level than ZnSO_4 . ZnGly facilitated the most efficient storage of Zn in the edible roots, and the highest Zn use efficiency was observed under ZnGly treatment at spraying rates of 1.19 or 2.39 kg ha^{-1} . In summary, the high fertilizer efficiency and safety threshold spraying rate of ZnGly are potentially useful for Zn biofortification of sweet potato.

1. Introduction

Zn is an essential nutrient for human health (Kieffer and Schneider, 1991; Deshpande et al., 2013). As per recent reports, Zn might even facilitate the fight against the unprecedented global burden of COVID-19 infection (Hunter et al., 2020; Skalny et al., 2020; Yasui et al., 2020). However, because of the low Zn level in common staple food crops, billions of people in developing countries cannot obtain the adequate Zn balance, thereby facing risks of hidden hunger or Zn-deficiency disorders, with clinical manifestations such as growth retardation, hypozincemia, and impaired immunity (Prasad, 2012; Garg et al., 2018; Geyik et al., 2020). As there are no functional Zn storage organs in the human body (Gibson, 2012), an adequate Zn dietary intake is a requirement for human health.

Sweet potato (*Ipomoea batatas* L.) is a very important source for food and nutrition security in many low-income countries and rural regions owing to its low maintenance requirement and high yield potential (Adetola et al., 2020; Laurie et al., 2015; Viliamu et al., 2018). However, Zn concentration in the edible roots of sweet potato is almost the lowest among all staple food crops (Adetola et al., 2020; Laurie et al., 2015; Lim et al., 2013), and maximizing it would be of great significance to

sweet potato growing regions and for global Zn nutritional balance. Production of Zn-enriched varieties of sweet potatoes, via genetic or other modification methods, have not yet been reported (Chen et al., 2021; Garg et al., 2018). Application of Zn fertilizer is an effective method to grow Zn-biofortified sweet potatoes globally, independent of constrictive crops, cultivars, and soils (Cakmak, 2007; Montalvo et al., 2016; Montalvo et al., 2016; Zou et al., 2012).

ZnSO_4 is the most widely used Zn fertilizer (Li et al., 2009). Although few studies have focused on sweet potato (Sun et al., 2018), numerous studies have demonstrated that ZnSO_4 application can increase the Zn concentration in edible parts of many crops (Liu et al., 2016; Prasad et al., 2014; Hilário et al., 2020), especially by foliar spraying (Zou et al., 2012; Wang et al., 2012; Zhou et al., 2020). Movability and availability of inorganic Zn sources are influenced by soil conditions; this effect is partly attributable to the superiority of foliar spraying over soil application. Moreover, anthropogenic chelators are being adopted as new Zn sources and the most common chelator used is ethylenediaminetetraacetic acid (EDTA). In terms of Zn biofortification effects, EDTA-chelated Zn (ZnEDTA) is better than ZnSO_4 for soil applications (Chatterjee and Mandal, 1985; Karak et al., 2005; Zhao et al., 2016), whereas it is not as good for foliar application (Wei et al.,

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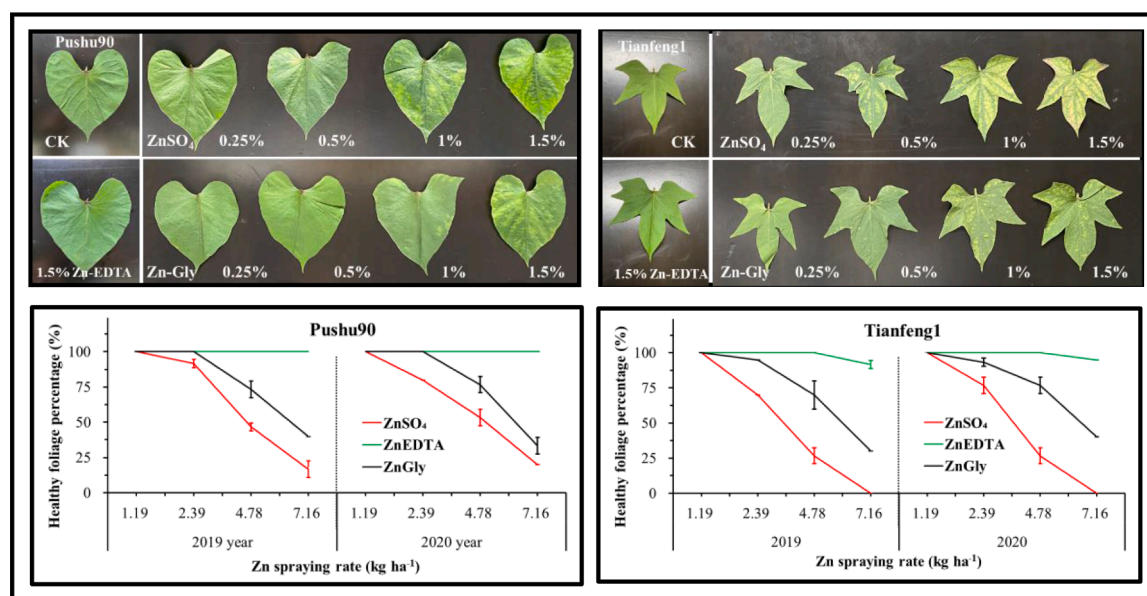


Fig. 1. Zn fertilizer-induced foliar burn and healthy foliage percentage.

2012; Golden et al., 2016). Another chelated Zn sources are based on natural organic chelators, such as amino acids, which are biodegradable, nontoxic, and highly compatible with environment-friendly agriculture (Dolev et al., 2020). Some studies have indicated that the Zn-biofortified performance of amino acid-chelated Zn was greater than that of either ZnSO_4 or ZnEDTA (Wei et al., 2012; Ghasemi et al., 2013; Mohammadi and Khoshgoftarmanesh, 2014; Tabesh et al., 2020). However, comparative performance analyses among the three typical Zn sources on sweet potatoes have been scarce.

With an increase in the spraying rate of ZnSO_4 , Zn concentration in the edible parts also steadily increased in rice (Xu et al., 2021), wheat (Zhang et al., 2012; Dapkekar et al., 2018), corn (Xu et al., 2022), and green bean (Hilário et al., 2020). However, excess ZnSO_4 causes visible foliage phytotoxicity in corn (Drissi et al., 2015; Golden et al., 2016), and even causes substantial yield loss in tomato (Kaya and Higgs, 2002). Hence, the foliar Zn spray spraying rate is usually $\leq 2.39 \text{ kg ha}^{-1}$ to prevent foliar burn, which is empirically used for all other Zn sources as well. Golden et al. (2016) reported that ZnEDTA caused a lower extent of foliar injury in corn, compared to that caused by ZnSO_4 . It was possibly because the pH and electrical conductivity (EC) of ZnEDTA are more suitable for leaf spray applications than those of ZnSO_4 . However, the effect of physicochemical properties of Zn fertilizers has not been tested. Previous studies have mostly focused on a narrow range of frequently used spraying rate to compare the effects of different Zn sources. The effects of a wider range of spraying rates of chelated Zn sources on sweet potato are not clear.

The aim of this study was to evaluate the effects of foliar sprays of three common Zn sources (namely, ZnSO_4 , ZnEDTA , and ZnGly), at normal to high spraying rates, on the foliar burn, photosynthetic performance, tuber yield, and concentration of trace elements in sweet potato. The specific objectives of the present study were: (1) to identify chelated Zn sources that allow spraying at higher rates with less severe foliar burn than ZnSO_4 , (2) to identify an optimal Zn source with the highest fertilizer efficiency, and (3) to elucidate whether the difference in solution pH and EC is the determining factor for foliar burn induced by Zn sources. The results may help balancing yield and Zn concentration of tuber root to meet food and nutrition security, especially for sweet potato-salience regions.

Table 1

Values of pH and EC (electrical conductivity, mS cm^{-1}) of different Zn sources at different Zn spraying rates (25 °C).

Zn source	Zn spraying rate (kg ha^{-1})							
	1.19	2.39	4.78	7.16	1.19	2.39	4.78	7.16
ZnSO_4	pH 5.87 EC 0.94	pH 5.69 EC 1.71	pH 5.50 EC 3.24	pH 5.41 EC 4.12				
ZnEDTA	pH 5.95 EC 1.26	pH 5.94 EC 2.08	pH 5.79 EC 4.03	pH 5.82 EC 6.25				
ZnGly	pH 4.62 EC 0.95	pH 4.41 EC 1.74	pH 4.26 EC 3.27	pH 4.22 EC 4.27				

2. Materials and methods

2.1. Experimental site and soil analysis

The two-year experiment was performed from May 2019 to October 2020 in the Panzhai village of Meijiang Town, Zhejiang Province, China ($29^{\circ}19' \text{ N}$, $119^{\circ}43' \text{ E}$). The site is subject to a typical subtropical monsoon climate; monthly average temperature and rainfall data from the nearby meteorological station are provided in Fig. S1 of the Supplementary Materials. The soil is classified as Stagnic Anthrosol in Chinese Soil Taxonomy. Three core samples were randomly collected each year from the topsoil (0 to 30 cm) for chemical analysis before seedling transplant. The soil analysis methods referred to methods of Lu (2000) and the results of soil analysis are provided in Table S1 of the Supplementary Materials.

2.2. Experimental design

Two sweet potato cultivars with different potential yield and leaf size were chosen: the normal-yield cultivar Pushu90 has heart-shaped leaves with entire margins, and the high-yielding cultivar Tianfeng1 has small palmate leaves (Fig. 1). The experimental design was a factorial combination of three Zn foliar sources (ZnSO_4 , ZnEDTA , and ZnGly) and four spraying rate gradients (1.19 , 2.39 , 4.78 and 7.16 kg ha^{-1}) arranged in a randomized complete block with 3 repetitions. A total of three spraying intervals of 7 days were carried out (beginning on 29 August 2019 or 2 September 2020) during the root expansion stage. The spray amount in each plot was 700 L ha^{-1} for each spraying. The pH and EC of different Zn solutions are shown in Table 1. Control (CK) seedlings of each cultivar were sprayed with water. Surfactant Tween 80 was added at

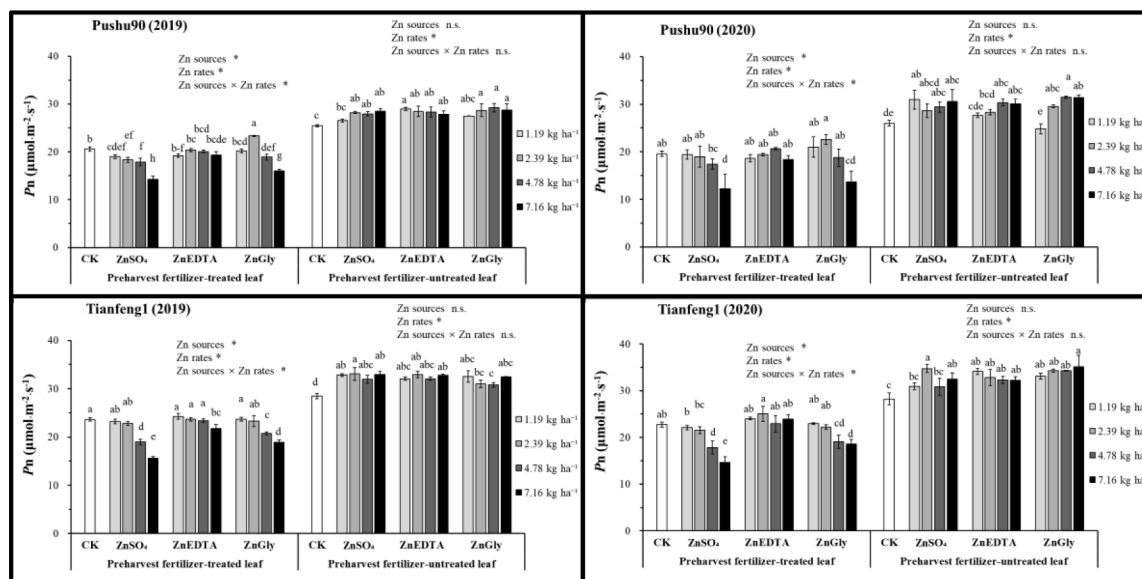


Fig. 2. Effect of foliar spraying of different Zn sources and rates on net photosynthetic rate (Pn) of two sweet potato cultivars in 2019 and 2020. Note: For the two-way ANOVA, * indicates significant differences ($p < 0.05$); n.s. indicates insignificant differences. Values of the same measured leaf followed by different lowercase letters within a cultivar and year are significantly different at the 5% probability level according to the LSD test.

0.01% (v v⁻¹) to all treatments, including CK. Spraying was performed after 17:00 on rainless and windless days with 2 L artificial compression sprayers.

The rectangular experimental field was surrounded by protective rows. The field was divided into 78 plots. Each plot (8 m², 3.33 × 2.40 m) was surrounded by a 0.5 m alley, which facilitated the spraying of foliar fertilization. Each plot had three 0.3 m high ridges, 0.8 m apart, in a north to south orientation. Every ridge was planted with ten 40-day-old sweet potato cutting-seedlings 0.3 m apart from each other. The cutting-seedlings were grown from virus-free roots and transplanted on 23 May 2019 and 26 May 2020. The N (150 kg N ha⁻¹ as coated urea), P (120 kg P₂O₅ ha⁻¹ as triple super-phosphate) and K (225 kg K₂O ha⁻¹ as potassium chloride) fertilizers were incorporated as base fertilizer into the top 0.15 m of the ridge soil in all treatments, prior to manual transplanting. All other management measures, except for foliar fertilizers, were consistent with standard practice in the region.

2.3. Assessment of leaf health status

The third expanded mature leaf (called post-spraying leaf) in the middle ridge of each plot was photographed 1 week after the last spray to compare the extent of damage caused by Zn fertilizers. Healthy foliar areas unaffected by fertilizer-induced damage were visually estimated as a percentage of the total leaf area (Golden et al., 2016). The leaf health status was always assessed by the same researcher in the field. One week before harvest, the photosynthetic performance of the sprayed leaf (called preharvest fertilizer-treated leaf) and the newly third expanded mature leaf (called preharvest fertilizer-untreated leaf) was measured on the right side of the leaf base, using the LI-6400 XT portable photosynthesis system (LI-COR Inc., Lincoln, NE, USA). The measurement was performed within 2.5 h beginning at 9:30 to minimize the variation due to changes in time of day.

2.4. Root yield, sampling, and analysis

All roots in each plot were collected and weighed on 27 October 2019 and 25 October 2020; then, three plants from the middle ridge in each plot were randomly selected for analysis and weighed individually. All representative tuber roots (> 100 g) of each plant were selected and washed in tap water followed by two washes in deionized H₂O. They

were then sectioned into peel (periderm), outer flesh (cortex), and inner flesh (stele) using a ceramic knife. Each part was cut into thin pieces, dried to a constant weight at 65 °C (first at 105 °C for 15 min), crushed into powder, and packed in bags.

All roots and leaf samples destined for micronutrient analysis were ground in a MM301 grinder with an agate mortar (Retsch, Germany) and digested in 16 mL of nitric acid (HNO₃) (guaranteed reagent, GR), perchloric acid (HClO₄) (GR), and hydrogen peroxide (H₂O₂), in a 5:2:1 vol ratio. The digestion solutions were allowed to cool to room temperature (25 °C) and diluted to 50 mL using double deionized H₂O. The concentrations of Zn, iron (Fe), Cd, and Cr were determined using ICP-MS (model 7500a, Agilent, USA). Reference material (rice, GBW-E 080,684) and blank controls were included in each batch of samples.

To quantitatively evaluate the rate of non-use or percentage loss of the Zn fertilizer, Zn loss percentage (ZLP,%)—defined as the percentage of Zn ions in the foliar fertilizer that apparently did not transfer into the roots—was calculated using the following equation (Moreira et al., 2018):

$$ZLP(\%) = 100 - \frac{C_t \times W_t \times 1000 - C_0 \times W_0 \times 1000}{F} \times 100$$

where C_t and C_0 represent the Zn concentration (mg kg⁻¹) in the tuber root of sweet potato treated with or without the foliar Zn fertilizer, respectively; W_t and W_0 represent the weight (t ha⁻¹) of the tuber root of sweet potato treated with or without the foliar Zn fertilizer, respectively; and F is the total Zn ion input (mg ha⁻¹) of the foliar Zn fertilizer.

2.5. Statistical analysis

The results were analysed using STATISTICA software 5.5 and were elaborated using Microsoft Excel 2016. Data were expressed as means ± SD ($n = 3$). Following Shapiro–Wilk test to assess variable normality, the sources of variation included Zn sources, Zn rates, and the interaction Zn sources × Zn rate were evaluated by performing a two-way ANOVA (analysis of variance). The means were compared by the least significant difference (LSD) test at $p < 0.05$.

Table 2

Effect of different Zn sources and rates on fresh weight (t ha^{-1}) of edible roots of two sweet potato cultivars.

Cultivar	Year	Zn rate (kg ha^{-1})	Zn source ZnSO ₄	ZnEDTA	ZnGly	Significance ($p < 0.05$)
Pushu90	2019	0	52.24 cde			
		1.19	54.35 bcde	54.07 e	62.81 ab	Zn sources n. s.
		2.39	57.09 abcd	58.07 abc	63.79 a	Zn rates n. s.
		4.78	49.72 de	55.79 a- e	56.69 a-e	Zn source n. s.
		7.16	50.73 cde	52.80 cde	51.85 cde	× Zn rates
		0	51.54 d			
	2020	1.19	52.49 cd	58.62 ab	59.65 ab	Zn sources *
		2.39	55.24 bcd	58.03 abc	61.18 a	Zn rates *
		4.78	55.01 bcd	61.69 a	58.64 ab	Zn source n. s.
		7.16	50.54 d	57.4 abc	55.08 bcd	× Zn rates
	2019	0	60.56 a-e			
		1.19	57.97 b-f	55.61 ef	58.85 b-f	Zn sources *
		2.39	58.95 b-f	62.71 abc	65.14 a	Zn rates *
		4.78	57.36 cdef	63.72 ab	61.91 abcd	Zn source n. s.
		7.16	53.27 f	61.88 abcd	56.52 def	× Zn rates
		0	64.45 abc			
Tianfeng1	2020	1.19	62.47 a-e	65.55 ab	65.52 ab	Zn sources *
		2.39	59.85 cde	63.26 a- e	66.77 a	Zn rates n. s.
		4.78	58.54 de	63.71 abcd	60.79 bcde	Zn source n. s.
		7.16	58.14 e	65.48 ab	61.41 a-e	× Zn rates

Note: For the two-way ANOVA, * indicates significant differences ($p < 0.05$); n. s. indicates insignificant differences. Values followed by different lowercase letters within a cultivar and year are significantly different at the 5% probability level according to the LSD' test.

3. Results

3.1. Zn fertilizer-induced foliar burn

The fertilizer-induced foliar burn showed visible yellow mottles and/or streaks, which were usually found between leaf veins (Fig. 1). The foliar burn occurred only on fertilizer-touched leaves; thereafter, all newly grown leaves appeared healthy. Control and all ZnEDTA treated leaves of both cultivars (only 7.16 kg ha^{-1} ZnEDTA treatment is shown in Fig. 1) showed little, if any leaf damage. The yellow mottles on the leaves increased with Zn spraying rate. In the most severely injured leaves treated with 7.16 kg ha^{-1} ZnSO₄, necrotic margins were visible only on Tianfeng1, implying that burn on Tianfeng1 leaves became more severe than that on Pushu90 ones. For Pushu90, the safe threshold spraying rates for spraying ZnSO₄ and ZnGly were 2.39 and 4.78 kg ha^{-1} , respectively, while for Tianfeng1 the safe threshold levels were 1.19 and 2.39 kg ha^{-1} , respectively. In short, both chelated fertilizers reduced ZnSO₄-induced foliar burn and showed a higher safety threshold of spraying rate than that of ZnSO₄.

3.2. Net photosynthetic rate

As shown in Fig. 2, the net photosynthetic rate (Pn) in fertilizer-treated leaves was lower than that in the new, fertilizer-untreated leaves for the two sweet potato cultivars. The Zn sources, Zn rates, and cultivars have significant effects on fertilizer-treated leaves (Table S2). The Pn in leaves of Pushu90 was lower than that of Tianfeng1, regardless of leaf types. Both ZnSO₄ and ZnGly application reduced Pn in fertilizer-treated leaves of both cultivars with increasing Zn rate, while ZnEDTA application had little influence on Pn. In addition, the reduction in Pn by ZnGly was less than that recorded for ZnSO₄, which was consistent with the differences of Zn sources in foliar burn in Fig. 1.

3.3. Fresh yield

The root yield of Pushu90 showed a parabolic trend with increasing Zn rate, while that of Tianfeng1 showed a decreasing trend (Table 2). No apparent Zn sources \times Zn rates interaction was implicated. At 1.19 kg ha^{-1} rate, the root yields in ZnSO₄ and ZnEDTA groups were similar to the yield observed under the control treatment, while the yield under the ZnGly treatment was significantly increased in Pushu90. The best yield was consistently obtained by the application of ZnGly at 2.39 kg ha^{-1} , while the lowest yields were consistently observed at 4.78 kg ha^{-1} .

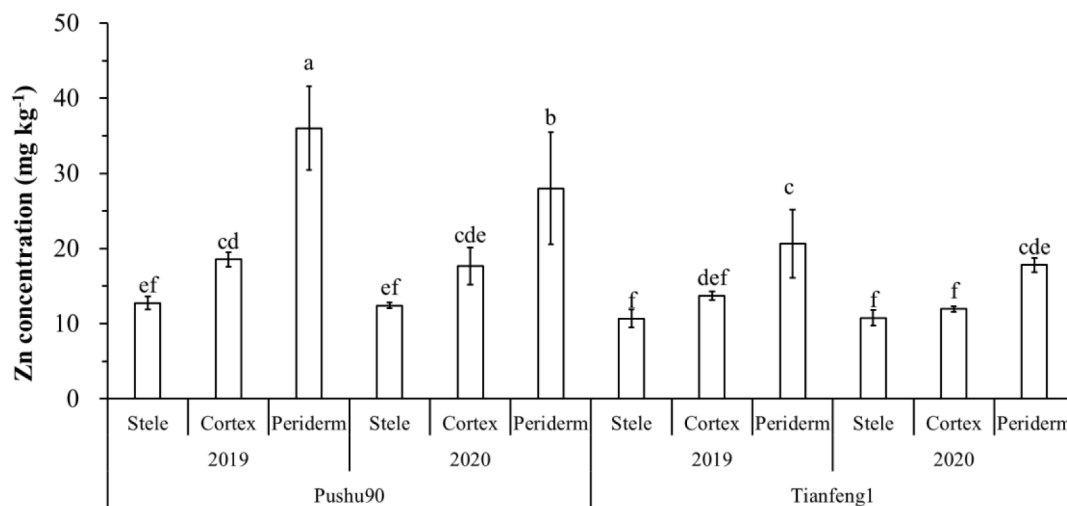


Fig. 3. Distribution of Zn concentration in different root parts of two sweet potato cultivars. Samples are of control plants (without foliar Zn fertilizer treatment). Bars with different lowercase letters are significantly different at the 5% probability level according to the LSD' test.

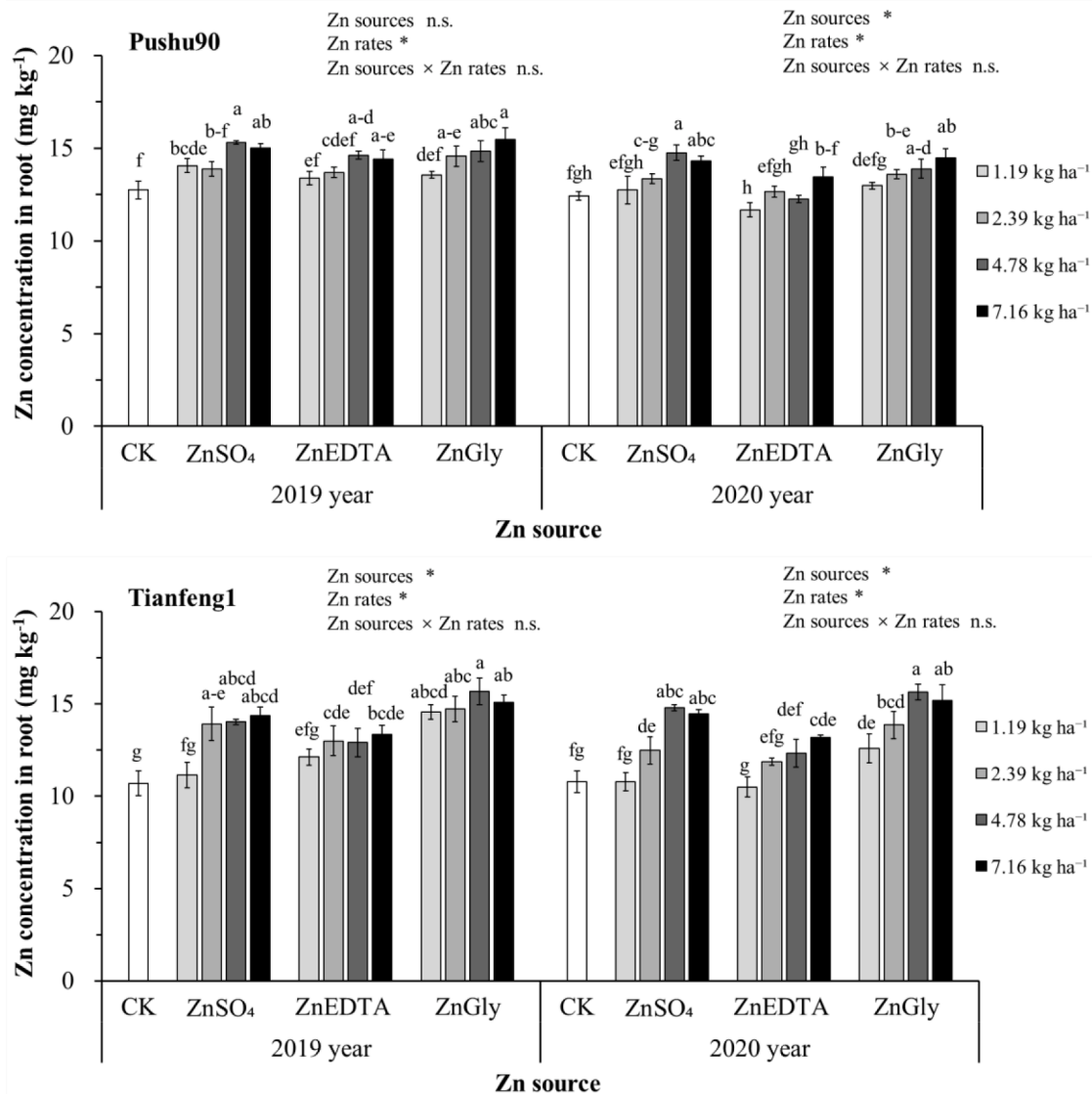


Fig. 4. Effect of foliar spraying with different Zn sources and rates on Zn concentration in the stele of edible roots of two sweet potato cultivars. **Note:** For the two-way ANOVA, * indicates significant differences ($p < 0.05$); n.s. indicates insignificant differences. Values followed by different lowercase letters within a cultivar and year are significantly different at the 5% probability level according to the LSD' test.

or 7.16 kg ha⁻¹ ZnSO₄. Although this treatment caused significant reduction in root yield compared to the best yield, it did not significantly reduce the yield relative to the control treatment. Overall, ZnSO₄ treatment had a relatively stronger negative effect on root yield than either ZnEDTA or ZnGly, especially when the Zn rate reached 4.78 kg ha⁻¹.

Cultivars and years have also significant effects on tuber yield (Table S2). The root yield of Pushu90 was higher ($p < 0.05$) than that of Tianfeng1, for that the leaf Pn of Pushu90 were higher than that of Tianfeng1. For both cultivars, the fresh yields in 2019 were higher than that in 2020, for that the sunshine hours in 2019 were higher than that in 2020 (Fig. S1).

3.4. Distribution of Zn concentration in tuber root

Regardless of years, elements, or cultivars, Zn concentration were consistently found in the following order: stele < cortex < periderm (Fig. 3). Distribution of Fe, Cd and Cr concentration also followed the same rule (Table S3). Zn concentration in roots of cultivar Pushu90 were usually higher than those in roots of Tianfeng1. The concentration of Zn

in the cortex was almost 0.5-fold higher than those in the stele. Similarly, the concentration of Zn in the periderm were about 1.0-fold higher than those in the stele. The stele is the bulk but with lowest Zn concentration in the edible root. Therefore, the most meaningful bio-fortification result would be to improve Zn concentration in the stele.

3.5. Zn concentration and Zn amounts in tuber root

The Zn concentration in the stele first increased, then stabilised with a further increase in the application rate (Fig. 4). No apparent Zn sources × Zn rates interaction could be implicated, although the fertilizer sprays enhanced Zn concentration in the stele. At 1.19 kg ha⁻¹ rate, Zn sources did not consistently exert a positive effect on Zn concentration in the stele. At 2.39 kg ha⁻¹, ZnEDTA and ZnSO₄ did not significantly improve Zn concentration in the stele, except for Tianfeng1 in 2019. However, ZnGly significantly increased Zn concentration in the stele, except for that in Pushu90 in 2020. At 4.78 and 7.16 kg ha⁻¹, differences among the three Zn sources were not significant for Pushu90; however, Zn concentration in the stele of Tianfeng1 under the ZnGly treatment was significantly increased compared to that for ZnEDTA

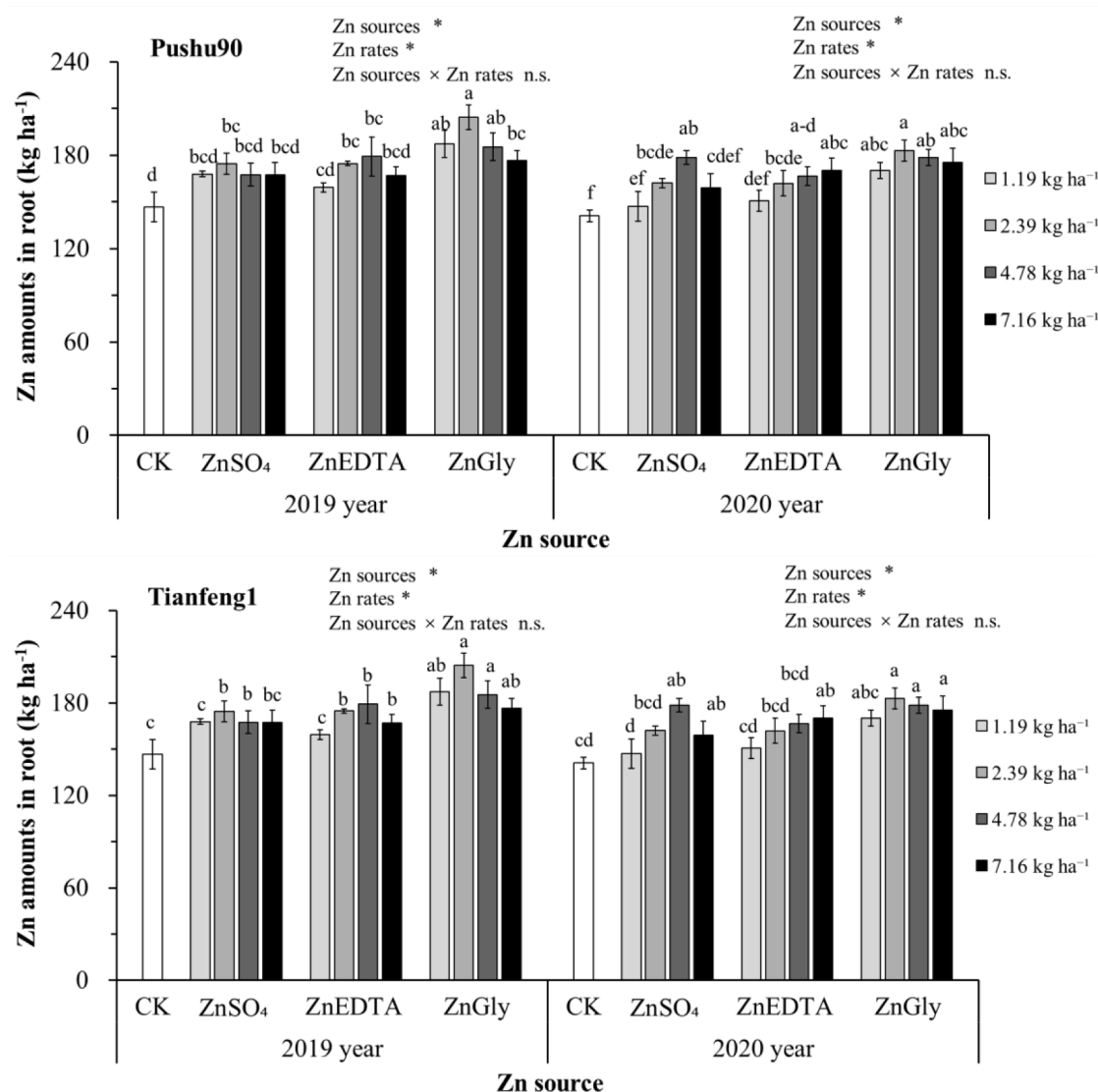


Fig. 5. Effect of foliar spraying with different Zn sources and rates on Zn amounts in the roots of two sweet potato cultivars. **Note:** For the two-way ANOVA, * indicates significant differences ($p < 0.05$); n.s. indicates insignificant differences. Values followed by different lowercase letters within a cultivar and year are significantly different at the 5% probability level according to the LSD test.

treatment.

For two cultivars, the Zn concentration change in the stele was different and the interaction of Zn source × Cultivar reached significant level ($p < 0.05$) (Table S2). Relative to the control, the mean annual percent increase in Zn concentration in Pushu90 ranged between 10.87–14.36, 0.64–10.13, and 10.42–14.73 in response to all ZnSO₄, ZnEDTA, and ZnGly spraying rates, respectively. The mean percent increase in Zn concentration in Tianfeng1 was 21.75–24.87, 11.00–20.08, and 32.78–40.35 under all spraying rates of ZnSO₄, ZnEDTA, and ZnGly, respectively. In short, the Zn-biofortified effect of ZnEDTA was lower than that of ZnSO₄ or ZnGly and Tianfeng1 was more readily biofortified than Pushu90.

The higher the total Zn amount in the tuber root, the greater were the storage of Zn ion in the edible roots (Fig. 5). For both cultivars, the highest Zn amounts in tuber roots always appeared in 2.39 kg ha⁻¹ and 4.78 kg ha⁻¹ ZnGly treatments, which was significantly higher than that of CK.

3.6. Zn loss percentage

The lower the ZLP value, the greater the storage of Zn ion from the

fertilizer in the edible roots (Fig. 6). With the increasing rates of ZnGly, the ZLP values were increased; no similar rules were found by ZnGly and ZnEDTA. The ZLP value exhibited a Zn source × Zn rate interaction (Table S2). There were no significant differences among all treatments except 1.19 and 2.39 kg ha⁻¹ ZnGly. The ZLP value for 1.19 and 2.39 kg ha⁻¹ ZnGly was the lowest, regardless of the cultivar and year.

4. Discussion

4.1. Relationships between foliar burn, foliar photosynthesis, and tuber yield

It is generally accepted that the impact of foliar damage can have significant yield implications. Therefore, visible foliage damage can be an important indicator of plant health, photosynthetic capacity, yield potential, and source selection and spraying rate threshold of Zn fertilizers (Du et al., 2014; Kaya and Higgs, 2002). However, in our study, the relationship among Zn fertilizer-induced foliar burn, foliar photosynthesis, and root yield showed a poor correlation because of the different changes in net photosynthetic rate of newly grown, fertilizer-untouched leaves (Fig. 1; Fig. 2; Table 2). The presumably significant reduction in

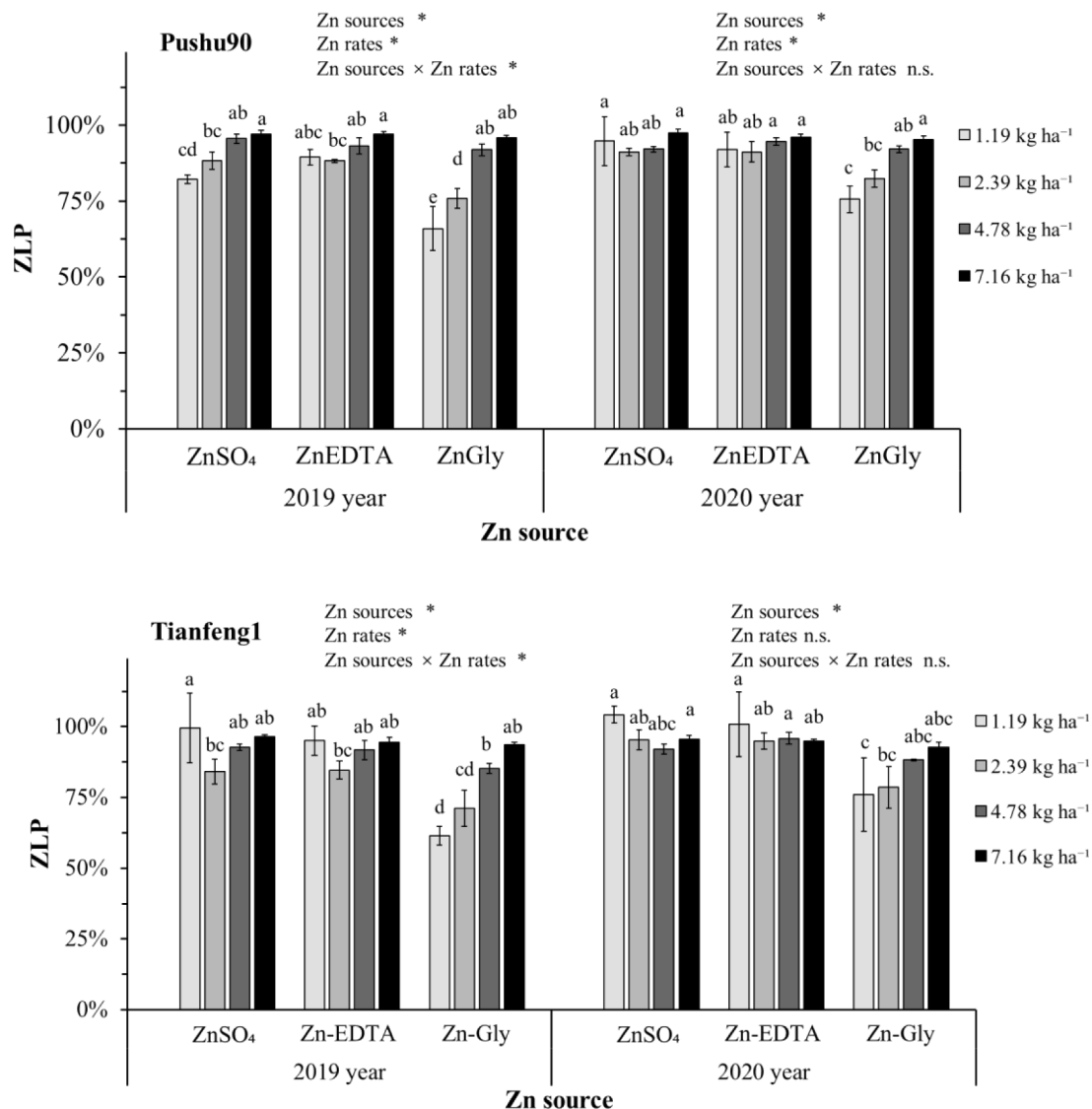


Fig. 6. Effect of foliar spraying with different Zn sources and rates on Zn loss percentage (ZLP) in the roots of two sweet potato cultivars. **Note:** For the two-way ANOVA, * indicates significant differences ($p < 0.05$); n.s. indicates insignificant differences. Values followed by different lowercase letters within a cultivar and year are significantly different at the 5% probability level according to the LSD' test.

root yield caused by foliar burn was partly offset because of compensatory growth effects, especially in Pushu90. Drissi et al. (2015) reported that corn leaf burns from excess ZnSO_4 foliar application did not affect silage yield. Golden et al. (2016) reported that despite high levels of foliar injury observed for some Zn sources at higher spraying rates, no differences in grain yield were observed. In our study, although the worst foliage damage did not cause a yield reduction compared to the control, it did result in an obvious yield reduction when compared to the optimum yield.

4.2. Variability of degree of fertilizer-induced foliar burn

The pH value of foliar Zn fertilizers is usually adjusted to 5–8 to avoid acid damage, and the Zn (typically $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$) spraying rate is usually lower than 2.39 kg ha^{-1} to prevent salt damage (Li et al., 2009). However, this study demonstrated that a $\text{pH} < 5$ and/or Zn spraying rate $> 2.39 \text{ kg ha}^{-1}$ did not always cause foliar damage (Table 1). Furthermore, although the pH of ZnGly was lower than that of ZnSO_4 and the EC value of ZnEDTA was higher than that of ZnSO_4 , both chelated fertilizers relieved foliar injuries induced by ZnSO_4 .

Rather than pH or EC, the type of Zn source affects the foliar

phytotoxicity in sweet potatoes. ZnSO_4 -induced phytotoxicity was caused by the toxicity of Zn ions. Zn toxicity to plant growth, leaf photosynthesis and mineral composition has been extensively studied by the addition of excess Zn to the rooting medium (Anjum et al., 2015; Chaney, 1993; Prasad, 2012; Saleh and Maftoun, 2008). For foliar application, Qiao et al. (2014) reported that excess ZnSO_4 caused the rate of photosynthesis of rice plants to decrease due to the toxic microenvironment for photosynthesis-related enzymes and inhibition of chlorophyll synthesis. In terms of chelating ability and stability, EDTA was much stronger than glycine. The concentration of remaining free Zn ions in foliar fertilizer ranked in the following order: $\text{CK} < \text{ZnEDTA} < \text{ZnGly} < \text{ZnSO}_4$. Similarly, the degree of fertilizer-induced foliar burns in sweet potatoes followed the same rule (Fig. 1). Golden et al. (2016) reported fertilizer-induced foliar burn in corn seedlings in the severity order $\text{ZnEDTA} < \text{ZnSO}_4 < \text{Zn-Citrate}$ (citric acid chelated Zn), suggesting that the Zn-Citrate chelate itself should be more harmful than the Zn ion. Updated research based on a Zn radio isotope tracer and synchrotron-based X-ray absorption near-edge structures has demonstrated that ZnEDTA was immediately absorbed in the chelated form and moved more slowly than ZnSO_4 in wheat leaves (Doolete et al., 2018). Based on these facts, alleviation of leaf burn, probably caused by

localized Zn toxicity, is not only because of the reduced concentration of Zn ions in foliar fertilizers, but also due to the relative harmlessness of the ZnEDTA and ZnGly complexes compared to Zn ion toxicity. This causal relationship requires further research for confirmation.

4.3. Variability of Zn biofortification level

ZnEDTA was harmless to sweet potato leaves, while the high chelating ability of EDTA resulted in the lowest Zn concentration in edible roots. The weaker ZnEDTA leaf absorption capacity was probably because ZnEDTA chelate had a higher molecular weight and stability than ZnGly chelate and ZnSO₄ molecules (Marešová et al., 2012; Dolev et al., 2020). This ultimately led to the lowest shoot-to-root transfer ability and Zn level in the edible root (Doolette et al., 2018). Despite a few different examples (Kayan et al., 2015), most studies have also shown that Zn-biofortified levels of foliar ZnEDTA application were lower than those of ZnSO₄ (Wei et al., 2012; Golden et al., 2016). In addition, EDTA itself has potential environment and food risks (Claudia and Jaime, 2021; Muthu, 2014). Conversely, as a Zn chelator, glycine might be an effective source of organic nitrogen for plant nutrition with no environmental risks ((Souri and Hatamian, 2018). In short, since the leaves of the target crops were vulnerable to ZnSO₄ toxicity, ZnGly rather than ZnEDTA might be recommended as a foliar Zn-biofortified fertilizer, owing to its superior effectiveness, lower loss rate, and better degradability.

4.4. Differences between cultivars

The tuber yield varied based on the sweet potato cultivars (Table S2). Although a more severe degree of leaf burn was observed, a higher Pn value in Tianfeng1 resulted in higher tuber yield. However, higher tuber yield occurred concomitantly with lower concentrations of Zn in the tuber roots of Tianfeng1, compared to Pushu90, likely due to a dilution effect (Bañuelos and Lin, 2008). Perhaps for the low Zn background value, Zn biofortification on Tianfeng1 was better than that on Pushu90. Pushu90 is a ZnSO₄-tolerant cultivar, whereas Tianfeng1 seems to be a ZnSO₄-sensitive cultivar. Changes in Fe, Cd and Cr concentration in tuber roots were also not consistent between the two cultivars (Fig. S2; Fig. S3; Fig. S4).

5. Conclusion

The Zn source, rather than its pH and EC value, was mainly responsible for foliar burn. Chelated Zn sources induced lesser foliar burn than ZnSO₄ and exhibited a wider working range of spraying rates. These findings can be useful for applications that require high Zn spraying rates or the alleviation of foliar burn in exogenous ZnSO₄-sensitive plants. However, ZnEDTA had relatively minor biofortification effects especially on Pushu90 and entailed potential environmental risk. ZnGly achieved a higher biofortification level than ZnEDTA and higher yield level than ZnSO₄. Moreover, ZnGly facilitated the most efficient storage of Zn in the edible roots. Additionally, glycine is essentially nontoxic to living things, making glycine-chelated fertilizers highly suitable for sustainable agriculture. In the future, the physiological mechanisms of chelated Zn fertilizers for alleviating foliar phytotoxicity of excess ZnSO₄ and enhancing Zn biofortification of crops require further research to aid their extensive application in the field.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.scienta.2022.110880.

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Further reading

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