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Mutual effects and in planta accumulation of co-existing cerium oxide nanoparticles and cadmium in hydroponically grown soybean (Glycine max (L.) Merr.)†

Lorenzo Rossi, Hamidreza Sharifan, Weilan Zhang, Arthur P. Schwabb and Xingmao Ma*

Cadmium (Cd) is a metal toxic to humans even at very low concentrations. Elevated levels of Cd in soil due to various anthropogenic activities have led to higher Cd concentrations in various crop tissues, making it a food safety concern. With the progressive production and continued accumulation of engineered nanoparticles (ENPs) in agricultural soils, understanding the effect of ENPs on the uptake and accumulation of metals by plants is imperative. The goal of this study was to determine the mutual effects of cerium oxide nanoparticles (CeO$_2$NPs) and Cd$^{2+}$ on their uptake and accumulation by soybean seedlings (Glycine max (L.) Merr.) in a hydroponic system. Soybean seedlings were exposed to four treatments (1.0 mg L$^{-1}$ Cd$^{2+}$, 1.0 mg L$^{-1}$ Cd$^{2+}$ + 100 mg L$^{-1}$ CeO$_2$NPs, 100 mg L$^{-1}$ CeO$_2$NPs and 0 mg L$^{-1}$ Cd and CeO$_2$NPs as the control) for 10 days. At termination, plant roots and shoots were separated and the concentrations of Cd and Ce in these tissues were determined. In addition, the amounts of ionic Ce and particulate Ce within soybean roots were determined. Significant interactions between co-existing CeO$_2$NPs and Cd were found with regard to their accumulation in plant tissues. While CeO$_2$NPs did not affect the total Cd associated with soybean roots, they significantly reduced the translocation of Cd from roots to shoots by 70%. In contrast, the co-presence of Cd lowered the concentration of Ce in soybean roots by 45% but significantly increased the concentration of Ce in soybean shoots by 60%. The altered plant accumulation of co-existing Cd and Ce was attributed to various physical, chemical and biological processes that occurred in the plant rhizosphere. Specifically, the co-presence of Cd and CeO$_2$NPs led to higher excretion of plant root exudates, which might have altered the chemical environment in the plant rhizosphere and enhanced CeO$_2$NP dissolution, leading to different plant accumulation of both chemicals.

Introduction

Cadmium (Cd) is a ubiquitous environmental pollutant toxic to humans. Chronic exposure to Cd has been shown to be associated with a variety of health problems such as renal dysfunction, cancer, osteoporosis, and cardiovascular disease.\cite{1,2} Consumption of Cd-tainted food is a predominant pathway for human exposure to Cd due to its efficient soil-to-plant transfer.\cite{3,4} Food consumption accounts for almost 90% of Cd exposure in the general non-smoking population, rendering
it an urgent task to understand processes which may potentially aggravate the plant accumulation of Cd.²

With the current embrace of nanotechnology in many industries including agriculture, engineered nanoparticles (ENPs) are increasingly detected in agricultural soils. They affect plant physiological and biochemical processes at various levels and alter the plant uptake of co-existing environmental pollutants.⁵,⁶ Earlier efforts in investigating ENPs and co-occurring environmental pollutants are mostly focused on organic contaminants.⁸ Few studies have evaluated the effects of ENPs on the plant uptake of co-existing heavy metals. The plant uptake of heavy metals involves complex transport processes and some are highly regulated. For example, Cd²⁺ is taken up by plant roots both apoplastically and symplastically.⁴,⁷ In the symplastic pathway, Cd²⁺ may diffuse into plant root cells passively or be actively transported in them via different transporters embedded in the root plasma membrane.⁵,⁷,⁸ Two types of Cd²⁺ transporters have been identified including the more specific Cd²⁺ ZIP transporters (zinc-regulated transporter/iron-regulated transporter-like proteins) and the less selective calcium permeable channels.⁷,⁹,¹⁰ Once Cd is in the plant root cells, it can be translocated to plant leaves. The translocation from roots to shoots is greatly affected by the vacuolar compartmentation of Cd–phytochelatin complexes as well as the transporters involved in the xylem loading of Cd.⁴,⁷,⁸,¹¹,¹²

ENPs may potentially interrupt many of the physiological processes involved in heavy metal uptake and transport. Consequently, investigating the impact of ENPs on the plant uptake of co-existing heavy metals is of great interest. Cerium oxide nanoparticles (CeO₂NPs) have been broadly used over the past years, particularly as a fuel additive.¹³,¹⁴ The main pathways for CeO₂NPs to enter into agricultural soils include deposition of vehicle exhausts, municipal run-off and land application of bio-solids.¹³,¹⁴ Simultaneous exposure to both CeO₂NPs and heavy metals such as Cd²⁺ is increasingly possible for crops. Due to the disruptive impacts of CeO₂NPs on the cellular membrane as well as the plant physiology and root anatomy¹⁵,¹⁶ CeO₂NPs may substantially change the plant uptake and accumulation of heavy metals.

In addition, CeO₂NPs display a high adsorption capacity for hazardous heavy metals such as arsenic, lead and Cd.¹⁷,¹⁸ Consequently, CeO₂NPs may serve as carriers for Cd²⁺ to enter into plant tissues or retain Cd²⁺ in the plant rhizosphere when they are not taken up by plants.¹⁹,²⁰ The complex interactions of CeO₂NPs and other metals suggest that CeO₂NPs are likely to alter the plant uptake and accumulation of co-present heavy metals. A few available studies in the literature with different ENPs corroborate this assumption. For example, TiO₂NPs (100 mg L⁻¹) were shown to significantly reduce the phytotoxicity of Cd to rice (Oryza sativa L.), which is likely due to the adsorption of Cd on TiO₂.²¹ However, comprehensive studies and mechanistic insights into ENP–heavy metal interactions in the plant rhizosphere are lacking.

The plant uptake of ENPs may be affected by various abiotic stresses. Salt stress and nutrient deficiencies increased the plant uptake of ENP elements.²²–²⁴ ENP accumulation in plant tissues has been a food safety concern because dietary consumption of ENP contaminated plant tissues is an important pathway for human exposure to these emerging chemicals. As a result, the plant uptake of ENPs in the presence of heavy metals (a source of abiotic stress) would complement the research.

The objectives of this research were to investigate the reciprocal effects of CeO₂NPs and Cd on the plant uptake of these materials by soybeans and to explore the underlying physical and chemical mechanisms leading to the altered plant uptake of these materials. Soybean (Glycine max. (L.) Merr.) was used in this study because it is a main protein source of the human diet that can accumulate metals (including Cd) in the grains.²⁵ Soybean was reported by the Food and Agricultural Organization (FAO) as the fifth most produced crop worldwide that provides about 30% vegetable oil and 77% nitrogen fixation. The concentration of Cd in soybean seeds is positively correlated with its concentration in the soil.²⁶ The concentration of CeO₂NPs (100 mg L⁻¹) was chosen based on many previous nanotoxicity studies with terrestrial plants, which used concentrations mostly in the range of 1–1000 mg L⁻¹.²⁷ CeO₂NPs at this concentration display a significant impact on plant physiological processes but not lethal effects.²⁸,³⁰ Cd sulfate (CdSO₄) was purchased from Fisher Scientific Inc. (Pittsburgh, PA) and was dissolved in tap water to obtain a final concentration of 1.0 mg L⁻¹ Cd.

Material and methods

CeO₂NPs and Cd

CeO₂NPs coated with polyvinylpyrrolidone were purchased from US Research Nanomaterials, Inc. (Houston, TX). The average size of the nanoparticles was 41.7 ± 5.2 nm, calculated by averaging the diameter of over 100 individual nanoparticles measured with ImageJ using transmission electron microscopy images of the NPs as reported earlier.²⁷ The pH of the 100 mg L⁻¹ CeO₂NP dispersion was 7.1 and the zeta potential of the nanoparticles was −48.6 mV, determined using a dynamic light scattering Zetasizer Nano ZS90 (Malvern, UK). X-ray photoelectron spectroscopy (XPS) indicated that about 9.7% of Ce on the nanoparticle surface exists as Ce⁵⁺ (Fig. S1†).

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Plant species and growth conditions

Glycine max (L.) Merr. (soybean) cv. Tohya seeds were purchased from Johnny’s Selected Seeds (Winslow, ME). The soybean seeds were germinated in moist sand for 5 days. After germination, young seedlings were individually transplanted into 50 mL plastic centrifuge tubes (Becton, Dickinson and Co., Franklin Lakes, NJ, USA) filled with 25% Hoagland solution³¹ (Phyto Technology Lab, Shawnee Mission, KS). The transpiration rate was recorded in terms of daily water loss
Determination of dissolved and particulate Ce in soybean tissues was performed following a published protocol.\textsuperscript{33} Enzymatic extraction of ionic and particulate Ce from plant tissues was assumed to contain large molecular weight molecules from exudates such as proteins, polysaccharides and border cells. Low molecular weight molecules in exudates such as short-chain aliphatic acids were expected to remain in the supernatant and were decanted. The combined pellets were re-suspended in 40 mL DI water and altogether six such tubes were prepared. They were divided into three groups. Different combinations of Cd and CeO$_2$NPs were introduced to these tubes. The first group contained 30 mg L$^{-1}$ Cd$^{2+}$, the second group 100 mg L$^{-1}$ CeO$_2$NPs and the third group both

Cadmium and cerium content analyses

An aliquot of 0.25 g subsample of dry roots and 0.5 g of the shoot tissues were ground and digested with 4 mL of 70% (v/v) nitric acid using a DigiPREP MS hot block digester (SCP Science, Clark Graham, Canada) at 95 °C for 4 hours following the EPA method 3050b.\textsuperscript{34} After cooling to room temperature, 2 mL of 30% (w/v) H$_2$O$_2$ were added to the mixture and heated in the hot block at 95 °C for 2 hours. The Ce and Cd amounts in a final concentration of 1% nitric acid were quantified by ICP-MS.

Total organic carbon determination

Total organic carbon (TOC) in the growth media was measured as an indicator of the excretion of plant root exudates. The growth medium at termination was first diluted with tap water to 50 mL. The diluted solution was then shaken for 12 hours on a shaker table. Subsequently, the TOC in those samples was measured using a TOC analyzer (Shimadzu TOC-VWP Analyzer, Japan) following reported procedures.\textsuperscript{35,36} The TOC in the growth medium was then obtained by correcting the measured TOC with the dilution factor for each sample. Three replicates from each treatment were measured.

Investigating the association of Cd with CeO$_2$NPs and plant root exudates

The potential chemical interactions of Cd with CeO$_2$NPs and biomolecules in plant exudates were examined. Forty-two soybean seedlings were grown in 20% strength Hoagland solution in 50 mL centrifuge tubes for 21 days. Transpired water was replenished with Hoagland solution of the same strength during cultivation. At termination, plant seedlings were removed, and the growth medium containing root exudates was centrifuged at 10,000 rpm for 10 min. Afterwards, the supernatant was decanted and the pellet at the bottom of each centrifuge tube was gently collected and transferred to a new tube in which the pellet was combined with pellets from six other tubes to further concentrate them. The pellets were assumed to contain large molecular weight molecules from exudates such as proteins, polysaccharides and border cells. Low molecular weight molecules in exudates such as short-chain aliphatic acids were expected to remain in the supernatant and were decanted. The combined pellets were re-suspended in 40 mL DI water and altogether six such tubes were prepared. They were divided into three groups. Different combinations of Cd and CeO$_2$NPs were introduced to these tubes. The first group contained 30 mg L$^{-1}$ Cd$^{2+}$, the second group 100 mg L$^{-1}$ CeO$_2$NPs and the third group both
chemicals at the same concentrations. The tubes were shaken for 24 h on a shaker table and then centrifuged at 10,000 rpm for 15 min. The supernatant was then decanted, and the residual was desiccated at 90 °C for 4 hours to form a dried pellet for X-ray photoelectron spectroscopy (XPS). XPS was carried out using an Omicron ESCA (Scienta Omicron GmbH, Germany) connected to a probe spectrometer with polychromatic Mg Kα X-rays (hν = 1253.6 eV). The ground sample was mounted on a stainless-steel sample holder using double sided adhesive conductive carbon tape. The X-ray was mounted on a stainless-steel sample holder using double sided adhesive conductive carbon tape. The X-ray was performed at 225 W where the pass energy was set to 100 eV for survey scans and 50 eV for specific regions to ensure sufficient sensitivity under a base pressure of 10−6 Pa. The calibration of the binding energies of the core levels was fixed at 284.8 eV for the main C 1s B.E. peak. The peaks of the XPS spectrum were fitted by a Gaussian function using Origin software 2017 (OriginLab, Northampton, MA).

Statistical analyses

Data were subjected to analysis of variance using a 2 × 2 factorial experiment set-up. Two-way ANOVA was carried out to differentiate the significance of independent variables: CeO2NPs and Cd. One-way ANOVA was performed and mean separation between treatments was obtained via the Games–Howell nonparametric post hoc test. Data were analyzed using the IBM SPSS Statistics software (Version 22.0. Armonk, NY).

Results

Growth analysis and plant uptake of CeO2NPs and Cd

Neither CeO2NPs nor Cd at the applied concentrations, separately or in combination, caused any significant changes in the root and shoot dry biomass of soybean seedlings (Fig. 1). The Ce concentration increased in roots and shoots of CeO2NPs treated plants compared to the controls (Fig. 2). Interestingly, the co-existence of Cd markedly decreased the concentration of Ce in soybean roots but not in shoots (Fig. 2). The Ce associated with soybean roots decreased by 45% in CeO2NPs + Cd treated plants but increased in the shoots by 44% compared to plants treated with CeO2NPs alone. Two-way ANOVA showed a significant interaction between CeO2NPs and Cd for Ce accumulation in soybean tissues. The total Cd associated with soybean roots was unaffected by the presence of CeO2NPs (Fig. 3). In contrast, the presence of CeO2NPs led to a significant decrease of Cd in plant shoots by 78%. Significant interactions between CeO2NPs and Cd were only found in the shoots for Cd accumulation.

Speciation of CeO2NPs in soybean roots

To gain further insights into the mutual effects of CeO2NPs and Cd, in planta speciation of Ce in plant roots was determined. The concentration of ionic Ce in soybean roots increased by 65% in the presence of both CeO2NPs and Cd compared to soybeans exposed to CeO2NPs alone. (Fig. 4A). Conversely, in the presence of CeO2NPs + Cd, the concentration of particulate Ce decreased by 45% (Fig. 4B), and the CeO2NPs × Cd interaction was significant. The total Ce concentrations in the growth media and root washing solutions were determined through acid digestion and ICP-MS quantification. The Ce concentration in the growth medium was not altered by the presence of Cd and no interactions were found (Fig. 4C), but the total Ce concentration in the washing solution, presumably all forms of Ce deposited on the root surface, was increased by 47% in the co-presence of CeO2NPs and Cd3+, compared to plants exposed to CeO2NPs (Fig. 4D).

Total organic carbon and pH

The TOC in the growth media exposed to CeO2NPs and/or Cd was higher than in control plants. The TOC in the growth medium from the joint treatment of CeO2NPs and Cd was noticeably higher than that from the treatments with CeO2NPs or Cd alone, even though the differences were not statistically significant (Fig. 5). The pH values in the growth media and on the root surface were significantly reduced by the joint exposure to CeO2NPs and Cd (Table 1), and interactions between CeO2NPs and Cd were found based on the pH on the root surface. However, no significant interactions were found between CeO2NPs and Cd in the growth media (Table 1).

X-ray photoelectron spectroscopy

The cadmium spectra are shown in Fig. 6. Two fitting peaks (u and u′) at the binding energy of 404.82 eV and 411.82 eV were identified and were assigned to Cd3d5/2 and Cd3d3/2,
Fig. 2  Total cerium concentration in roots (A) and shoots (B) of Glycine max grown in the presence of 100 mg L\(^{-1}\) CeO\(_2\)NPs and 1 mg L\(^{-1}\) Cd. Means labeled with different letters are significantly different based on the Games–Howell nonparametric post hoc test (\(p < 0.05\)). Error bars represent the standard deviation (\(n = 3\)). Two-way ANOVA is reported in Table S1†.

Fig. 3  The cadmium concentration (mg kg\(^{-1}\)) in roots (A) and shoots (B) of Glycine max increased in the presence of 100 mg L\(^{-1}\) CeO\(_2\)NPs and 1 mg L\(^{-1}\) Cd. Means labeled with different letters are significantly different based on the Games–Howell post hoc test (\(p < 0.05\)). Error bars represent the standard deviation (\(n = 3\)). Two-way ANOVA is reported in Table S1†.

Fig. 4  Cerium ions (A) and cerium particulates (B) in roots. Total cerium concentration in the growth medium (C) and in the root washing solution (D) of Glycine max grown in the presence of 100 mg L\(^{-1}\) CeO\(_2\)NPs and 1 mg L\(^{-1}\) Cd. Means labeled with different letters are significantly different based on the Games–Howell's post hoc test (\(p < 0.05\)). Error bars represent the standard deviation (\(n = 3\)). Two-way ANOVA is reported in Table S1†.
respectively.37,38 No Cd specific peaks were found in CeO₂ samples, but Cd peaks were detected in samples containing part of the root exudates as well as 30 mg L⁻¹ Cd and 100 mg L⁻¹ CeO₂NPs. Some Cd might have complexed with large molecules such as proteins and polysaccharides in plant root exudates or adsorbed on the surfaces of border cells, and some Cd might have adsorbed on the surface of CeO₂NPs. The evidence provided here is only qualitative, and quantitative characterization on the interactions between Cd and exudates and CeO₂NPs requires additional study, yet the results provided here do shed light on the possible mechanisms for the interactions of CeO₂NPs and Cd in the plant root rhizosphere.

Discussion

Interest in the interactions between ENPs and co-present environmental pollutants in different environmental systems are growing28,29,39 because these interactions may affect their accumulation in plant tissues. Consumption of food crops contaminated with various environmental pollutants is a significant pathway for human exposure to these materials.40–42

In this study, we examined the reciprocal effects of CeO₂NPs and Cd in an aqueous system, avoiding many of the complications caused by soil particles and allowing a mechanistic investigation on the interactions of CeO₂NPs and Cd in the soybean rhizosphere. CeO₂NPs, Cd and their interactions did not affect the overall dry weight and water transpiration, confirming that CeO₂NPs and Cd at the tested concentrations do not have any detrimental effects on plant growth and biomass, separately or together. Similar results have been reported in the literature. In fact, Gui et al.41 reported that lettuce (Lactuca sativa L.) treated with 100 mg kg⁻¹ CeO₂NPs grew much faster than the controls. Similarly, Cao et al.43 demonstrated that 100 mg kg⁻¹ CeO₂NPs improved the net.

Table 1 Root surface and growth medium pH values of Glycine max grown in the presence of 100 mg L⁻¹ CeO₂NPs and 1 mg L⁻¹ Cd. Means ± SD (n = 3) labeled with different letters are significantly different based on the Games–Howell post hoc test (p < 0.05).

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Root pH</th>
<th>Medium pH</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>7.40 ± 0.10bc</td>
<td>7.69 ± 0.13bc</td>
</tr>
<tr>
<td>CeO₂NPs</td>
<td>7.53 ± 0.06a</td>
<td>7.80 ± 0.06a</td>
</tr>
<tr>
<td>Cd</td>
<td>7.40 ± 0.10bc</td>
<td>7.66 ± 0.13bc</td>
</tr>
<tr>
<td>Ce NPs + Cd</td>
<td>7.20 ± 0.10b</td>
<td>7.50 ± 0.07b</td>
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<tr>
<td>Two-way ANOVA</td>
<td>p</td>
<td>p</td>
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<tr>
<td>CeO₂NPs</td>
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<td>0.633</td>
</tr>
<tr>
<td>Cd</td>
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<td>0.025</td>
</tr>
<tr>
<td>CeO₂NPs × Cd</td>
<td>0.015</td>
<td>0.057</td>
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</table>

Fig. 5 Total organic carbon (TOC) in Glycine max grown in the presence of 100 mg L⁻¹ CeO₂NPs and 1 mg L⁻¹ Cd. No statistical differences were found by the Games–Howell post hoc test (p < 0.05). Error bars represent the standard deviation (n = 3). Two-way ANOVA is reported in Table S1.†

Fig. 6 X-ray photoelectron spectroscopy spectra indicating the presence of cadmium (30 mg L⁻¹) on the surface of CeO₂NPs (100 mg L⁻¹) or its association with large molecular weight molecules in soybean root exudates. The horizontal axis shows the associated binding energy [eV] and the arbitrary unit (a.u.) represents the frequency of the peaks. Two peaks at B.E. 404.82 eV and 411.82 eV were identified as characteristic peaks of Cd3d₅/₂ and Cd3d₃/₂, respectively.
posed to them separately. The altered apoplastic barriers on the cell wall of endodermal cells, compared to plants existing chemicals differed. Cd led to a significant decrease of Ce in the roots but a significant increase in the shoots. For Cd, however, the co-occurrence of CeO2NPs significantly reduced its concentration in the shoots but did not affect Cd associated with the roots. This was attributed to the complicated physical, chemical and biological processes occurring concurrently in the soybean rhizosphere. First of all, the pH in the rhizosphere and on the plant root surface decreased in the co-presence of these two materials. The pH in the rhizosphere is affected by various biological processes and the lower pH observed in this study might be partially attributed to the increased root exudation which contains a broad spectrum of acids. A lower pH generally favors the dissolution of metals. In addition, the low molecular weight acids in plant root exudates can function as reducing agents and therefore enhance the reduction of Ce4+ to Ce3+, facilitating Ce uptake and transport to plant leaves as ionic Ce. In agreement with our results in Ce speciation, a higher concentration of ionic Ce was detected in soybean roots (Fig. 4). Ionic Ce is much more efficiently transported from roots to shoots than CeO2NPs. Consequently, a higher Ce concentration was detected in soybean shoots exposed to both CeO2NPs and Cd than in those exposed to CeO2NPs alone, even though the total Ce within root tissues was lower when the plants were exposed to both materials simultaneously.

The plant uptake of Cd is complicated and as described above, both apoplastic and symplastic pathways contribute to the internalization of Cd into plant root cells and then in the vascular system. The presence of CeO2NPs can affect both pathways. The co-occurrence of CeO2NPs and Cd altered the formation of “root apoplastic barriers” by depositing suberin on the cell wall of endodermal cells, compared to plants exposed to them separately. The altered apoplastic barriers could affect Cd entrance with the water flow along the cell wall. It is also possible that CeO2NPs affect the plant symplastic pathway by altering the activities of ion transporters or the integrity of root cell membranes, leading to different Cd accumulation in plants. Such mechanisms have not been extensively explored, but some previous studies have shown that CeO2NPs may disrupt root membrane integrity.

The adsorption of organic compounds on ENP surfaces plays a crucial role in the altered plant accumulation of organic compounds. A similar process could occur between ENPs and heavy metal ions; ENP surfaces have a high capacity for adsorption of heavy metals. The adsorption capacity of Cd on CeO2NPs was four times higher than that on TiO2NPs. In our study, while a detailed adsorption study was not performed, we have conducted a qualitative study in which CeO2NPs and Cd were mixed in growth media containing some actual root exudates. Cd was detected in dry pellets containing CeO2NPs, suggesting a likely adsorption of Cd on CeO2NPs (Fig. 6). Because of the relatively less efficient root-to-shoot transport of CeO2NPs, the adsorbed Cd would penetrate into roots with CeO2NPs but was less effectively transported to shoots. This possible interaction between Cd and CeO2NPs is consistent with the lower Cd concentration in plant shoots in the co-presence of CeO2NPs and Cd, while Cd in the roots was unaffected. In addition, the co-existence of CeO2NPs and Cd resulted in greater release of exudates. Cd may form complexes with organic exudates outside plant roots. The results concerning the potential complexation between Cd and root exudate molecules were inconclusive due to the weak XPS signals. However, if it is true, this complexation can be another mechanism for the altered Cd uptake in the presence of CeO2NPs because Cd chelates are transported into plant root cells through different protein transporters than Cd2+. Chelated Cd is also more effectively sequestered in the vacuole of plant root cells, reducing the loading efficiency to xylem tissues and therefore resulting in less efficient translocation to shoots. The adsorption of Cd onto biomolecules has been shown to be strongest at pH > 5. The pH in the growth media in this study was above 6, suggesting that affiliation between Cd and large size biomolecules in root exudates was possible. However, further evidence is needed to substantiate this hypothesis.

In summary, the Cd × CeO2NPs interaction was statistically significant for plant accumulation of co-existing Cd and CeO2NPs. The co-presence of Cd with CeO2NPs led to higher excretion of plant root exudates, lowered the pH values on the root surface and enhanced CeO2NPs dissolution, and therefore resulted in higher accumulation of Ce in plant shoots compared to plants exposed to CeO2NPs alone. A higher Ce content in the root washing solution indicated that a higher amount of Ce was attached to the plant root surface but did not penetrate into the tissues. The presence of CeO2NPs may provide an adsorption site for Cd and reduces its uptake into plants. The elevated release of root exudates may be another possible reason for the reduced Cd in plant shoots due to the possible complexation between Cd and root exudate molecules. The study provides important new insights into the chemical mechanisms for Cd and CeO2NP interactions in the plant root region. Due to the complex processes involved in both materials, future studies should aim to elucidate more molecular mechanisms for the mutual effects of these chemicals on their uptake and accumulation by plants.

Conflicts of interest
There are no conflicts to declare.

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