

Influence of CeO₂ and ZnO Nanoparticles on Cucumber Physiological Markers and Bioaccumulation of Ce and Zn: A Life Cycle Study

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S Supporting Information

ABSTRACT: With the dramatic increase in nanotechnologies, it has become increasingly likely that food crops will be exposed to excess engineered nanoparticles (NPs). In this study, cucumber plants were grown to full maturity in soil amended with either CeO₂ or ZnO NPs at concentrations of 0, 400, and 800 mg/kg. Chlorophyll and gas exchange were monitored, and physiological markers were recorded. Results showed that, at the concentrations tested, neither CeO₂ nor ZnO NPs impacted cucumber plant growth, gas exchange, and chlorophyll content. However, at 800 mg/kg treatment, CeO₂ NPs reduced the yield by 31.6% compared to the control ($p \leq 0.07$). ICP-MS results showed that the high concentration treatments resulted in the bioaccumulation of Ce and Zn in the fruit (1.27 mg of Ce and 110 mg Zn per kg dry weight). μ -XRF images exhibited Ce in the leaf vein vasculature, suggesting that Ce moves between tissues with water flow during transpiration. To the authors' knowledge, this is the first holistic study focusing on the impacts of CeO₂ and ZnO NPs in the life cycle of cucumber plants.

KEYWORDS: CeO₂ NPs, cucumber, gas exchange, growth, ZnO NPs

■ INTRODUCTION

Nanotechnologies are used in many new materials and devices with a vast range of applications. At the same time, the fast development of nanotechnologies raises concerns about the toxicity and environmental impact of nanoparticles (NPs). CeO₂ NPs are applied as diesel fuel-borne catalysts, in cosmetics, and as polishing agents.¹ ZnO NPs are widely used as UV absorbers; thus, they are applied in personal care products, directly in contact with the human body. This has generated concerns about health effects and motivated studies regarding possible NP toxicity.^{2–5}

Previous reports have shown that CeO₂ and ZnO NPs exhibit varying degrees of toxicity to ryegrass (*Lolium perenne*), alfalfa (*Medicago sativa*), mesquite (*Prosopis* sp.), lettuce (*Lactuca sativa*), corn (*Zea mays*), cilantro (*Coriandrum sativum* L.), and cucumber (*Cucumis sativus*).^{6–13} However, in most of the reported studies, the analyzed parameters included germination rate and root elongation in seedlings. So far, few studies have reported the effects of NPs in fully developed plants or have monitored physiological changes during the whole life cycle of plants exposed to NPs.^{14,15} In addition, little is known about the translocation of edible and reproductive tissues. Wang et al. observed the toxicity of CeO₂ NPs in tomato plants from seed germination to fruit maturity and found that CeO₂ NPs had inconsequential or a slightly positive effect on tomato growth and production.¹⁴ However, CeO₂

NPs reduced leaf count and diminished soybean yield.¹⁵ In addition, synchrotron analyses have shown the presence of CeO₂ NPs in soybean grains.¹⁶

Cucumber (*Cucumis sativus*) is a garden vegetable consumed worldwide, and its consumption is increasing annually.¹⁷ The cucumber plant has large leaf area, high transpiration rate, and requires more water than grain crops,¹⁸ which could represent higher NP uptake. Schwabe et al. compared the translocation of CeO₂ NPs between pumpkin (*Cucurbita maxima*) and wheat (*Triticum aestivum*) cultivated in hydroponic solution and found that Ce was translocated to the pumpkin shoot but not to the wheat shoot.¹⁹ They found that pumpkin transpired more water (200 mL) than wheat (50 mL) during 8 days of treatment. Ding et al. revealed that transpiration dependent translocation from the root to the shoot is the main Ce bioaccumulation process.²⁰ Another study suggested that once CeO₂ NPs enter into the vascular cylinder, they could move through the vascular bundle along with water flow.¹² Thus, we assumed that transpiration-dependent translocation may be an important mechanism for NP accumulation in cucumber plants.

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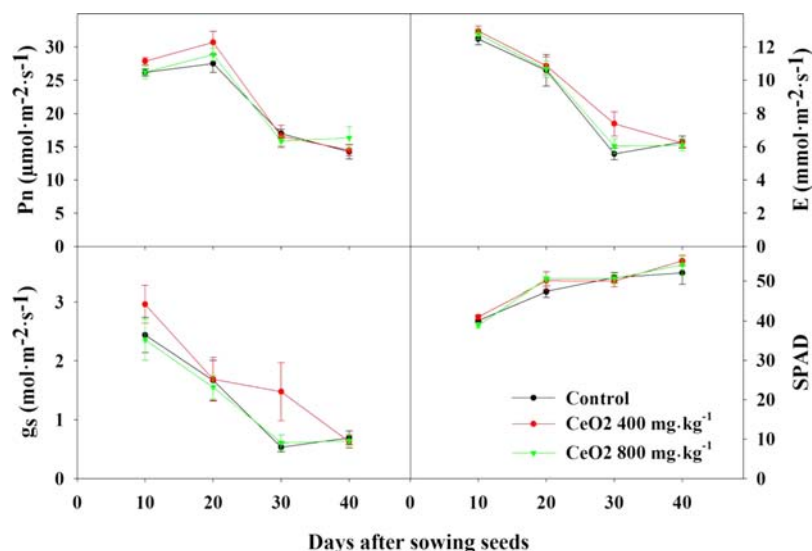


Figure 1. Leaf net photosynthesis (P_n), transpiration rate (E), stomatal conductance (g_s), and relative chlorophyll content (SPAD) of cucumber plants grown in substrate containing CeO₂ NPs at 0–800 mg/kg. These parameters were recorded every 10 days after germination. Error bars represent \pm standard error.

The phytotoxicity of NPs has been evaluated through growth parameters. Recently, the chlorophyll content has been included as an indicator for phytotoxicity assays.^{10,21–23} Silica particles (10–20 nm) were shown to decrease the chlorophyll content of *Scenedesmus obliquus* in the logarithm growth phase.²¹ Chlorophyll *a* fluorescence yield is a sensitive method to evaluate NP toxicity.²⁴ Perreault et al. evaluated the CuO NP toxicity in *Lemna gibba* by using chlorophyll *a* fluorescence.²⁵ However, the monitoring of plant photosynthetic processes during the entire life cycle under NP stress has yet to be reported.

The aims of this study were to characterize the physiological impact of CeO₂ and ZnO NPs on cucumber grown in soil medium, and the potential bioaccumulation of Ce and Zn in the fruit. Quantification and localization of Ce and Zn in the fruit were performed by using ICP-OES, ICP-MS, and μ -X-ray fluorescence (μ -XRF).

MATERIALS AND METHODS

Characteristics of Nanoparticles. The ZnO and CeO₂ NPs (Meliorum Technologies, Rochester, NY) were obtained from the University of California Center for Environmental Implications of Nanotechnology (UC CEIN). Previous characterization showed that ZnO and CeO₂ NPs have primary sizes of 10 ± 1 nm and 8 ± 1 nm, respectively.²⁶ Other NP characteristics were previously published.²³ NP suspensions were prepared by adding previously weighed amounts of NPs to specific deionized water volume. The suspensions were sonicated in an ice-cooled water bath (Crest Ultrasonics, Trenton, NJ) at 25 °C for 30 min and immediately applied to soil. The soil field capacity was previously determined in order to avoid leaching.

Substrate and Greenhouse Conditions. Two cucumber plants were grown in each 5.8 L Poly-Tainer container (22.5×19.5 cm²) containing local regular loam sand soil (3.7% clay, 12.2% silt, 84.1% sand, and 0.04% organic matter content, pH 7.9), sand (Quikrete Premium Play Sand, Atlanta, GA), and Sunshine Mix #4 (SunGro Hort., Bellevue, WA) at a ratio of 1:1:3 by volume. The bulk density of the substrate was 0.76 g·cm⁻³. CeO₂ NP and ZnO NP suspensions were prepared and applied to the soil to have final concentrations of 400 or 800 mg NPs/kg soil. Each treatment was replicated five times; four replicate/treatments were used to record the growth parameters, and the fifth one was used for synchrotron μ -XRF studies. Plants were grown for 53 days in a greenhouse. The temperatures in the

greenhouse were maintained at 30.5 ± 4.7 °C (mean \pm standard deviation) during the day and 25.8 ± 2.8 °C at night. The daily light integral (photosynthetically active radiation) was 17.3 ± 3.6 mol·m⁻²·d⁻¹.

Gas Exchange. Photosynthesis, respiration, and transpiration are indicators of plant health. In this study, leaf net photosynthetic rate (P_n), stomatal conductance (g_s), transpiration (E), and relative chlorophyll content (SPAD) were monitored every 10 days after sowing until fruit production occurred.

The (P_n), (E), and (g_s) of five cucumber plants per treatment were measured by placing the fully expanded leaf in the cuvette of a portable gas exchange system (CIRAS-2; PP Systems, Amesbury, MA). Environmental conditions in the cuvette were controlled at a leaf temperature of 25 °C, photosynthetic photon flux (PPF) of 1000 $\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, and CO₂ concentration of 375 $\mu\text{mol}\cdot\text{mol}^{-1}$. The data was recorded when environmental conditions and gas exchange parameters in the cuvette were stable. These measurements were taken on sunny days between 1000 and 1400 h, and plants were well watered to avoid water stress.

Chlorophyll Content Measurement. A hand-held SPAD chlorophyll meter (Minolta Camera, Japan) was used to measure the relative chlorophyll content of all plants. For each plant, two healthy, fully expanded leaves were chosen.

Growth Data. Plants were open pollinated in the greenhouse, and the harvest occurred when the cucumber fruits were in marketable size (53 days after treatment). The length of shoots, number of leaves, and fruits were recorded. Shoots were severed at the substrate surface and then divided according to tissue type—stem, leaves, and fruits, and their mass was immediately measured. The fruits were categorized into three groups, based on diameter and length: large (50.6–59.7 mm in diameter, 22.2–19.0 cm in length), medium (29.3–43.0 mm, 13.1–17.9 cm), and small (8.3–22.2 mm, 3.5–11.8 cm). Leaf area was determined using a LI-3100C area meter (LI-COR Biosciences, Lincoln, NE). The dry weight (DW) of roots, stem, leaves, and fruits was determined after oven-drying at 65 °C to constant weight. Before drying, the root system was removed by first carefully breaking apart the soil with a metal scopula, cleaned with running tap water, followed by rinsing (1 min, three times) in deionized water.

ICP-OES and ICP-MS Analysis. All dried tissues were ground to pass a 40-mesh screen with a stainless Wiley mill (Thomas Scientific, Swedesboro, NJ). Powder samples were digested with concentrated plasma-pure HNO₃ and H₂O₂ (30%) (1:4) using a microwave acceleration reaction system (CEM Corp; Mathews, NC). The digestion was performed following the EPA method 3051. For Ce

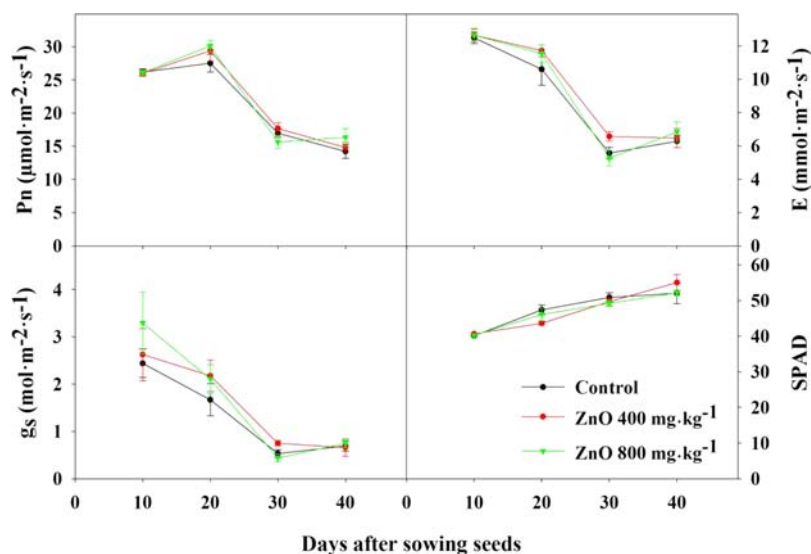


Figure 2. Leaf net photosynthesis (P_n), transpiration rate (E), stomatal conductance (g_s), and relative chlorophyll content (SPAD) of cucumber grown in substrate contaminated with ZnO NPs at 0–800 mg/kg. These parameters were recorded every 10 days after germination. Error bars represent \pm standard error.

determination in roots and Zn in all tissues, the digested samples were analyzed by inductively coupled plasma-optical emission spectroscopy (ICP-OES, Optima 4300 DV; Perkin-Elmer). For Ce determination in aerial tissues, the digested samples were analyzed by using an inductively coupled plasma-mass spectrometer (ICP-MS, ELAN DRC II; Perkin-Elmer). Standard reference materials from National Institute of Standards and Technology 1547, 1570a, and 2709a were used to validate the digestion and analytical method obtaining recoveries between 90% and 99%.

μ -XRF Analysis. Fruits and leaves were carefully washed with DI water to eliminate any surface contaminants. Then, they were transversally cut and frozen in liquid nitrogen for 30 min. The samples were fixed with Tissue Tek (Sakura Finetek USA, Torrance, CA) and sectioned with a cryomicrotome (Triangle Biomedical Sciences, Durham, NC) at -20 °C to a thickness of 30 μm . Subsequently, the samples were mounted onto Kapton tape and freeze-dried for 1 h in a Labconco freeze-dryer (FreeZone 4.5, Kansas City, MO) with operating conditions of -53 °C and 0.140 mBar pressure.

X-ray fluorescence imaging was obtained at beamline 10-2 at the Stanford Synchrotron Radiation Lightsource (Menlo Park, CA). Standard operating conditions were 3 GeV beam energy and 80–100 mA beam current. Incident X-ray energy was set to 10 KeV, and a Si (111) monochromator was used. SMAK²⁷ software was used for data analyses.

Data Analysis. The treatments, two nanoparticles at two concentrations and five replicate/treatments, were established in a completely random design. All data including growth, photosynthetic parameters, and mineral nutrients were analyzed by using PROC GLM in SAS software (version 9.1.3, SAS Institute, Cary, NC).

RESULTS AND DISCUSSION

Impact of CeO₂ and ZnO NPs on Gas Exchange and Chlorophyll Content. In this study, neither ZnO NPs nor CeO₂ NPs produced visible signs of toxicity in cucumber plants during the growth period. Data for total chlorophyll content (SPAD), leaf net photosynthetic rate (P_n), transpiration rate (E), and stomata conductance (g_s) at different growth stages of the CeO₂ NP treated plants are shown in Figure 1. As shown in this figure, total chlorophyll content gradually increased in both control and CeO₂ NP treated plants, but no significant differences among them were observed. Gas exchange (P_n , E , and g_s) was also not affected by CeO₂ NPs, even at high

exposure level. Previous reports have shown that CeO₂ NPs are strongly adhered to the root surface of cucumber, pumpkin, and wheat.^{13,19} Thus, very likely, the CeO₂ NPs, per se, did not affect the leaf gas exchange. However, the adsorbed NPs in the root surface may interfere with water transport inducing leaf response. Asli and Neumann observed a reduced water flow in corn plants treated with titanium dioxide NPs.²⁸ In the present study, the water content in cucumber fruit was not significantly changed by CeO₂ NPs, which suggests that these NPs did not affect the water flow in cucumber (Supporting Information, Table S1).

Contrary to the CeO₂ NPs, the ZnO NPs have been found to release ionic Zn into the soil solution.³ However, ZnO NP treated plants did not show any disturbance on leaf gas exchange and chlorophyll content, even at the highest ZnO NPs concentration treated (800 mg/kg) (Figure 2). Zn is an essential micronutrient involved in several physiological processes, yet at concentrations above 200 mg/kg tissue, it causes phytotoxicity in *Bacopa monniera* and *Lolium perenne L. cv Apollo* leaves.^{29–31} A previous study showed that the seedling growth of *L. perenne* (ryegrass) was retarded and that the plants had shorter roots and shoots when treated with 50 mg/L ZnO NPs.⁸ They also reported that “at 1000 mg/L ZnO NPs, the epidermis and root cap were broken, the cortical cells were highly vacuolated and collapsed, and the vascular cylinder also shrank.” In this study, the Zn concentrations in cucumber leaves were 409 and 564 mg/kg, respectively, at 400 and 800 mg/kg treatment, which is two to three times higher than the threshold value (200 mg/kg) reported in other studies.³¹ However, there were no signs of toxicity in the plants. This could be due to the slow release of Zn ions from the ZnO NPs. In our previous work, we found that the release of Zn from the ZnO NPs into soil solution was a slow process linked to plant uptake.²³

Impact of CeO₂ and ZnO NPs on Cucumber Growth and Yield. None of the treatment significantly affected the numbers of leaves, leaf area, stem length, and dry weight of roots, stems and leaves, compared to those of the control, indicating no impact of CeO₂ NPs on cucumber plant growth (Table 1 and Table S2). The yield of cucumber grown in 400

Table 1. Effect of CeO₂ and ZnO NPs on Cucumber Growth Parameters and Biomass Accumulation^a

treatment	leaf count	leaf area (cm ²)	shoot length (cm)	dry weight (g)			
				root	stem	leaves	fruit ^b
control	129.0 ± 11.2	16986 ± 473	249.8 ± 26.2	1.9 ± 0.3	34.3 ± 1.6	44.0 ± 1.4	1402.4 ± 83.7
400 mg/kg CeO ₂ NPs	127.0 ± 9.1	16447 ± 972	215.8 ± 31.8	2.1 ± 0.3	35.5 ± 2.8	43.2 ± 2.4	1480.2 ± 137.1
800 mg/kg CeO ₂ NPs	157.5 ± 14.6	18027 ± 2223	237.9 ± 35.9	2.6 ± 0.2	37.8 ± 3.5	47.3 ± 3.4	958.5 ± 190.0
<i>p</i> value	0.13	0.61	0.79	0.12	0.36	0.38	0.07
400 mg/kg ZnO NPs	141.8 ± 4.6	16916 ± 666	250.9 ± 5.7	2.1 ± 0.2	34.7 ± 1.1	43.9 ± 1.4	1410.6 ± 122.9
800 mg/kg ZnO NPs	137.5 ± 15.7	16107 ± 1125	254.1 ± 19.5	3.1 ± 0.4	34.4 ± 2.6	43.0 ± 2.1	1487.1 ± 101.5
<i>p</i> value	0.6	0.44	0.87	0.02	0.98	0.67	0.52

^aThe data are the means of four replications ± standard error. Plants were grown to full maturity in a mesocosm in organic soil containing CeO₂ or ZnO NPs at 0, 400, and 800 mg/kg. ^bThe data represent fresh weight.

Table 2. Concentration of Ce and Zn in Various Cucumber Tissues at Harvest^a

treatment	fruit	leaf	stem	root
Ce Concentration (mg/kg Dry Tissue)				
control	0.06 ± 0.03	0.83 ± 0.37	0.6 ± 0.1	4.5 ± 4.4
400 mg/kg CeO ₂ NPs	0.09 ± 0.04	1.72 ± 0.41	4.6 ± 0.9	317.4 ± 41.0
800 mg/kg CeO ₂ NPs	1.27 ± 0.44	2.69 ± 0.65	9.9 ± 5.6	551.2 ± 96.6
<i>p</i> value	0.014	0.019	0.061	<0.0001
Zn Concentration (mg/kg Dry Tissue)				
control	46.4 ± 5.7	55.5 ± 2.0	43.8 ± 2.7	169.6 ± 14.5
400 mg/kg ZnO NPs	102.7 ± 4.6	409.4 ± 20.2	205.1 ± 6.8	1416 ± 121.4
800 mg/kg ZnO NPs	110.7 ± 3.3	563.9 ± 25.7	262.2 ± 16.4	2150 ± 98.2
<i>p</i> value	0.0001	<0.0001	<0.0001	<0.0001

^aThe data are means of four replications ± standard error.

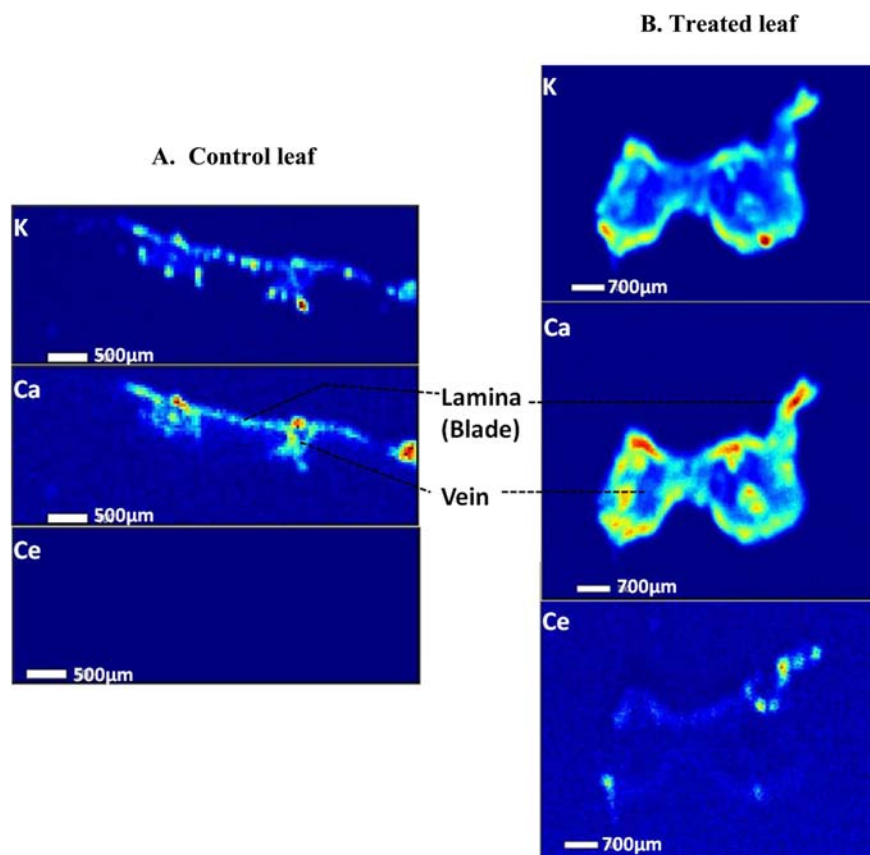


Figure 3. μ -XRF maps from transversally cut cucumber leaves showing normalized K, Ca, and Ce intensities. The image presents leaves exposed to (a) 0 and (b) 800 mg/kg CeO₂ NPs. Maps are presented in a temperature color display where the red color represents higher intensity, and the dark blue color represents the absence of the element.

mg/kg CeO₂ NPs was not impacted (Table 1). However, the high concentration (800 mg/kg) of CeO₂ NPs decreased the yield by 31.6% compared to that of the control, which was statistically significant at $p \leq 0.07$. Priester et al. reported that CeO₂ NPs diminish soybean yield.¹⁵ Cerium is a rare earth element (REE) that has been applied in agriculture as fertilizer for crop production in China since 1980s.^{32,33} The positive effect of REEs on plant growth and yield has been reported in previous studies.^{34–36} However, in the present study we found that CeO₂ NPs decreased the cucumber yield at an 800 mg/kg dose.

ZnO NPs did not negatively affect growth related parameters (Table 1) in the above ground plant parts. Moreover, the root dry biomass was significantly increased by ZnO NPs at both 400 and 800 mg/kg ($p \leq 0.02$). The root dry biomass increased by 10.5% and 63% for 400 and 800 mg/kg ZnO NPs, respectively. This suggests that at the concentration tested, in an organic soil, the ZnO NPs do not affect the growth of cucumber plants. A similar result was reported for soybean, where ZnO NPs increased root biomass.¹⁵ Dimkpa et al. also reported that ZnO NPs did not have a significant impact on wheat plant biomass.³⁷ In addition, the cucumber yield was not affected by ZnO NPs (Table 1). The results suggest that the concentrations tested were not toxic for cucumber. Conversely, the toxicity of ZnO NPs to various plants in several cultivation conditions has been previously reported. Lin and Xing reported that ZnO NPs at 2000 mg/kg significantly reduced cucumber root length.⁶ Lin and Xing also reported that the ZnO NPs at the concentration of higher than 50 mg/L reduced ryegrass growth with shrinking of root tips and vacuolation of root epidermal and cortical cells.⁸ ZnO NPs (5g/110 kg) have also been reported to reduce wheat growth in agricultural soil.³⁸ The difference with our results could be due to variation in cultivation conditions and concentrations.

Distribution of Ce in Cucumber Plants and Evaluation of Potential Hazards to Human. The ICP-OES and ICP-MS data showed the presence of Ce in roots, stems, leaves, and fruit after treatment with 400 and 800 mg/kg CeO₂ NPs (Table 2). The concentration of Ce in cucumber tissues followed the sequence root > stem > leaf > fruit. The presence of Ce in cucumber fruit indicates the possibility of introducing Ce ions/CeO₂ NPs into the food chain. It is reported that both CeO₂ NPs and Ce³⁺ ions are difficult to translocate to plant stems; only a small percentage will go from root to stem.^{13,39} In this study, the Ce translocation rate from root to stem was only 1.44% and 1.79% in plants treated with 400 and 800 mg/kg CeO₂ NPs, respectively. This concurs with previous reports which indicate that the majority of NPs are loosely adhered to the plant root surface.^{13,39} It was noted that the translocation rate from the stem to the leaf was about 37% and 27%, respectively, for the 400 and 800 mg/kg CeO₂ NP treatments. This may indicate that once Ce enters into the stem, it is easier to transport it to the leaf. The reason may be that CeO₂ NPs move smoothly in the vascular cylinder with the water flow to the tip of the vascular bundle.¹²

In this study, by using synchrotron μ -XRF, we analyzed the distribution of Ce in cucumber leaf and fruit. The μ -XRF Ce intensity map of the leaf is shown in Figure 3. Figure 3A (control) shows the distribution of K and Ca in both the lamina and the vein. As seen in this figure, Ce was not detected in the control. However, in CeO₂ treated plants Ce was mainly localized in the vasculature of the leaf vein (Figure 3B). This corroborates our previous assumption that Ce can move with

water flow and is transported to the leaf through the vasculature. μ -XRF analysis did not show Ce in cucumber fruit due to the large area, low concentration of Ce, and limited synchrotron beamtime (data not shown). By using XANES, Zhang et al. reported that Ce species in cucumber root exist as CeO₂ and CePO₄, while in the stem, 86.4% was as CeO₂ and 13.6% as cerium carboxylates, and in the leaf, 78.5% was as CeO₂ and 21.5% as cerium carboxylates.¹³ More recently, XANES studies have shown that soybean plants exposed to CeO₂ NPs store most of the Ce as CeO₂ NPs and that a small percentage is biotransformed to Ce(III).¹⁶ Thus, this suggests that, although at low concentration, cucumber fruit may have CeO₂ particles. To this end, it is impossible to determine the level of risk to human health from this amount of CeO₂ NPs detected in cucumber fruit; however, a study showed that in rodents, Ce "is absorbed by the gastrointestinal tract and distributed to other organs."⁴⁰ Zinc distributions in fruits of ZnO NP treated plants were not analyzed in the synchrotron since Zn is a natural macronutrient present in the plant.

Bioaccumulation of Zn in Cucumber and Evaluation of Potential Hazards to Humans. The bioaccumulation of Zn in different cucumber tissues is shown in Table 2. The concentration of Zn in cucumber tissues from higher to lower followed the sequence root > leaf > stem > fruit. Compared to the control, the variation of Zn in treated tissues was significantly different at $p < 0.0001$ for all cucumber tissues. Different from CeO₂ NPs, ZnO NPs release Zn ions that are taken up and translocated into the fruit. In this particular study, the Zn accumulation in cucumber fruits was 10.3 and 11.1 mg per 100 g of dry cucumber for the 400 and 800 mg/kg ZnO NPs treatments, while the control contained 4.6 mg Zn per 100 g cucumbers. According to the U.S. Department of Agriculture, fresh cucumber contains 0.2 mg of Zn per 100 g of fresh fruit. Converted to dry weight, cucumber fruit contains 95.23% of water, which is equivalent to 4.2 mg of Zn per 100 g of cucumber dry mass.⁴¹ This amount is similar to the average amount of zinc detected in our control sample (4.6 mg of Zn per 100 g of cucumber dry matter). However, ZnO NP treated cucumber plants contain 2 times the Zn compared to that in currently consumed cucumber. According to the Food and Nutrition Board at the Institute of Medicine of the National Academies, the recommended dietary allowance for female adults is 8 mg and 11 mg of Zn for adult males, while the tolerable upper intake level (ULs) for Zn in adults is 40 mg of daily intake.⁴² A recent study using data from The Third National Health and Nutrition Examination Survey (NHANES III), a large US population based cross-sectional study, was conducted to determine whether there is an association between dietary zinc intake and prevalent kidney stone disease defined as self-reported for any previous episode of the kidney. It was found that participants who consumed more than 15 mg/day of zinc were associated with a significant increased risk of kidney stone disease compared to those with lower dietary zinc intake (<7 mg/day). Tang et al. concluded that future prospective studies are needed to clarify the causal relationship between zinc intake and kidney stone formation.⁴³ The frequent consumption of cucumber treated with zinc NPs combined with zinc-rich animal-source foods such as beef or pork and legumes such as beans and chickpeas could result in zinc consumption that is higher than the recommended daily allowance, especially when combined with the daily consumption of a multimineral supplement, which is a common practice for approximately 40% men and women in U.S.⁴⁴ It is

of concern whether the chronic ingestion of cucumber with a Zn concentration that is 2 times higher than its normal amount might have an adverse effect on human health.

In summary, the results of this study have shown that ZnO NPs did not show toxicity to cucumber plants and in organic rich soil at the concentration tested. Also, CeO₂ NPs did not impact vegetative growth, but those NPs reduced the yield. None of the NP treatments affected gas exchange and chlorophyll content, which are indicators of stress. ICP data revealed that both Ce and Zn bioaccumulated in cucumber fruit. More studies are still needed in order to evaluate the significance of the bioaccumulation of Ce and Zn in fruits, especially the impact on the next plant generation.

■ ASSOCIATED CONTENT

Supporting Information

Water content of cucumber fruit from plants grown in soil treated with CeO₂ NPs. This material is available free of charge via the Internet at <http://pubs.acs.org>.

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Notes

Any opinions, findings, and conclusions or recommendations expressed in this material are those of the authors and do not necessarily reflect the views of the National Science Foundation or the Environmental Protection Agency. This work has not been subjected to EPA review and no official endorsement should be inferred.

The authors declare no competing financial interest.

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