



Engineered Nanomaterials, Plants, Plant Toxicity and Biotransformation

: A review

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Abstract

α -aminophosphonate oxadiazoles (5a-m) were prepared in high yields by reacting of 1,3,4-oxadiazole acetohydrazide (3) with Interaction between engineered nanomaterials and plants is important; Because plants have direct contact with water, soil, and therefore the atmosphere, the potential pathway for higher species to encounter these nanomaterials is thru the organic phenomenon that plants form the most ring and source of. the aim of the article, Plant Toxicity and Biotransformation, is to boost our understanding of a number of the interactions of engineered nanomaterials with plants, including their toxicity to plants and biotransformation or biodegradation of nanomaterials within the plant system. Mechanisms of nanomaterial toxicity to plants and biological access to nanomaterials aren't yet well understood. it's clear that in these circumstances, further evaluations of the interaction of nanomaterials and plants, likewise because the development of latest methods for characterizing nanomaterials in vivo, are necessary so as to create sustainable use of nanotechnology.

Keyword: in vivo, plant system, nanotechnology, animal cells, Biotransformation, nanomaterial toxicity.

1. Note

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2. Introduction

With the rapid development and widespread use of nanotechnology, engineered nanomaterials will

inevitably enter the environment and will pose a threat to ecological species. The environmental importance and biological effects of those nanomaterials as new pollutants have become the focus of attention[1-5]. The toxic effects of nanomaterials on the cells of humans and animals have been well studied but to the compromise of their effect on plants. Plants serve as a connection tool between the environment and the biosphere[6-9]. Additionally, as final receptors, environmental pollutants don't seem to be only directly stricken by nanomaterials but also affect their deformation and destiny and are the most route for exposure of upper

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species to nanomaterials through bioaccumulation within the organic phenomenon [10-14]. The environmental results of nanotechnology are divulged when nanomaterials interact with plants, but, to date, such interaction has not received enough attention. Most research studies on the toxicity of plant have put focus on toxicity symptoms hence there are few studies in which the mechanisms of plant toxicity, uptake, displacement and bioaccumulation change; as a results, a scientific review of the reports published during this field is important [15-20]. Therefore, cooperation seems necessary to exist between materials scientists, biologists and toxicologists in order to enhance the utilization of nanomaterials and minimize their adverse effects on health and also the environment [21-25]. The interactions between engineered nanomaterials (ENMs) and plants are of particular importance, as plants directly interact with soil, water, and the atmosphere, and serve as a potential pathway of ENMs exposure for higher species through the food chain. The aim of this paper is to extend our current understanding about interactions between ENMs and plants, including Phyto-toxicity, uptake, translocation, and biotransformation of ENMs in plant systems. The mechanisms underlying ENMs phytotoxicity and bioavailability are not well understood. It is clear that more investigations are urgently required in the area of ENMs–plants interactions, as well as the development of novel techniques for in vivo characterization of ENMs to enable these fields to keep pace with the sustain-able implementation of nanotechnology.

2. Interaction (interaction) between nanomaterials and plants

2.1. nanomaterials

Nanomaterials are materials that are but 100 nm in size in a minimum of one in all their dimensions and are engineered specifically for various applications [26-31]. These materials differ from their bulk counterparts in terms of extent and better reactivity and also are subject to quantum confinement. Engineered nanomaterials consist mainly of the followin types: 1) Carbon nanomaterials such as (CNTs), (C60) and graphene's (Figure 1); 2) Metal-based nanoparticles including Zero-Valente metals (such as Au, Ag, Fe, etc.), metal oxides (including ZnO, TiO₂, CeO₂, etc.) and metal salts (such as Nano silicates, ceramics, etc.); 3) Quantum dots

(QDs) (such as CdSe, CdTe, etc.); 4) Nano polymers (including dendrimers, polystyrene, latex), etc. [32-38]. Engineered nanomaterials enter the environment intentionally or accidentally at the time of manufacture or use. To support the sustainable development of nanotechnology, it's necessary to assess potential risks supported by comprehensive research in order to shed light on all corners of this issue.

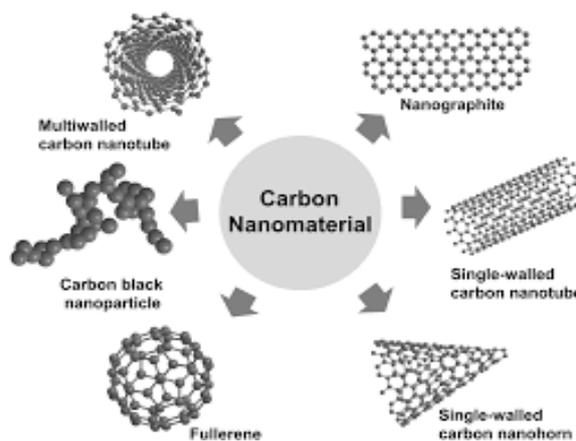


Figure 1. Different structures of carbon-based nanomaterials [2].

2.2. Plant poisoning due to engineered nanomaterials

Preliminary studies on the interaction of nanomaterials and plants have mainly focused on the plant toxicity aspect of nanomaterials. Plant toxicity of nanomaterials varies betting on the type of nanomaterial and plant species [39-41]. Tests of plant toxicity are commonly performed at two phases of plant development: 1) during germination, during which the germination rate and root elongation are measured; 2) During seedling growth, the elongation of roots and stems and their dry weight are measured. The traits of the ultimate stages of the expansion process are the foremost appropriate comparison criteria for nanomaterials and plants [42-46]. Recently, criteria like leaf number, chlorophyll content, moreover as cytotoxicity and genomic toxicity are utilized in plant toxicity assessments. So far, a large range of nanomaterial effects on plants are reported. In most studies, experimental species (including ecologically or economically important monocotyledonous, dicotyledonous, and non-crop species) are wont to assess plant toxicity in accordance with EPA guidelines. the foremost

studied nanomaterials were carbon nanomaterials and metal nanomaterials that had the very best production and application [47-51].

2.3. Carbon nanomaterials

Toxicity reports of carbon nanomaterials have been published for a variety of different plant species, but due to experimental conditions and different plant species, conflicting results have been obtained [52-56]. There is a general consensus that the high degree of functionalization of carbon nanotubes significantly reduces their toxic effects.

Cañas et al. [3] studied the effects of functional and inactive carbon nanotubes on root elongation of six plant species (cabbage, carrots, cucumbers, lettuce, onions, and tomatoes). They concluded that the plant toxicity of inactivated carbon nanotubes was greater than that of functionalized ones. Also, using a scanning electron microscope (SEM), it was understood that carbon nanotubes are sucked on the root surface but do not accumulate in the plant.

Stampoulis et al. [4] revealed that multi-walled carbon nanotubes with a concentration of 1000 mg / L under hydroponic culture did not affect the rate of germination of *C. pepo* but decreased biomass by 60% compared to the witness.

Liu et al. [5] showed that fullerenes (C₇₀ (COOH)₂) 4-8 inhibited root elongation in *Arabidopsis* and induced abnormal root gravitropism.

Lin et al. [6] showed that C₇₀ and multi-walled carbon nanotubes of natural organic origin cause a one-month delay in flowering in rice, indicating that carbon nanomaterials interfere with water and nutrient uptake.

Liu et al. [7] investigated changes in the cell wall of the tobacco plant (*Nicotiana tabacum* L. Cv. Bright Yellow) exposed to water-soluble carboxyfullerenes (C₇₀ (COOH)₂) 2-4. Deposition of these nanomaterials on the cell wall resulted in the inhibition of cell growth and disruption of the cell wall and membrane. This study provided direct evidence of the change in cell wall composition of plant viable cells by fullerenes.

Avanasi et al. [8] also investigated soil uptake, decomposition, and plant uptake of fullerene using C₆₀ solutions labeled with carbon 14 (¹⁴C). They showed that C₆₀ released into the environment is not very bioavailable to plants (about 7%), but may persist in soil for more than a year. According to Begum et al. [9], graphene substantially prevented plant growth and biomass production (cabbage, tomatoes, red spinach and lettuce) in comparison to

controls. The mechanism of this plant toxicity included oxidative stress [38-40]. Furthermore, the positive effect of carbon nanomaterials on plants has also been reported.

Miralles et al. [10] showed that a concentration of 2560 mg / kg of multi-walled carbon nanotubes increased germination and elongation of wheat and alfalfa roots. There are many other reports on the positive effects of nanomaterials on plant growth and development, but this article focuses more on toxicity.

Anjum et al. [11], showed tolerance of growing bean seedlings (*Vicia faba* L.) to different concentrations (0/100, 200, 400, 800 and 1600 mg / L) Single-bilayer oxide graphene plates graphene oxide sheet (0.5 to 5 microns) as well as related potential mechanisms. Also, both positive and negative effects of graphene oxide concentration in beans were revealed. Negative and significant effects of graphene oxide concentrations (in order of magnitude: 1600 > 200 > 100 mg / l graphene oxide) by reducing the growth parameters and activity of redox enzyme systems, in addition to increasing electrolyte leakage, H₂O₂, and lipid and protein oxidation was shown. Positive effects of graphene oxide (respectively: 800 > 400 mg / l) in the form of improving bean health based on indicators of reducing electrolyte leakage, H₂O₂, and lipid and protein oxidation, in addition to increasing the activity of the redox system, increasing the relative content of seed water an increase in praline was also shown [57-62]. These results indicate the complex interaction of carbon nanomaterials with farm plant species and their understanding requires further studies.

2.4. Metal based nanomaterials

Plants are directly exposed to the environmental elements of water, soil and atmosphere, and all three of these elements can be the basis for plants to be exposed to engineered nanomaterials. Different kinds of engineered metal oxide nanomaterials with varying features have been developed for using in biotechnology, agriculture and industry that are likely to be transferred and bio accumulated throughout the food chain [63-66]. Metal-based nanomaterials have a variety of effects on plants, with both positive and negative effects reported. Plant toxicity of nanomaterials depends on their properties, plant species as well as environmental conditions. In some cases, inconclusive results have been obtained even for the same nanomaterial [67-70]. For example, some

articles have reported the positive effects of titanium nan oxide on spinach growth, improved light absorption, increased activity of rubisco-activating enzymes, and reduced oxidative stress induced by UV-B radiation for chloroplasts [12].

Ghosh et al. [13] reported that a concentration of 4 mmol / L of titanium nan oxide could induce DNA laddering (DNA laddering): And micro contact in Album cape root cells. Wang et al. [14] indicated that the penetration of titanium nan oxide into Arabidopsis Taliana cells causes the micro tubular network to disintegrate, resulting in overloading of the proteasome system and isotropic growth of root epidermal cells.

Clement et al. [15] reported that the crystalline form of titanium nan oxide anatine is more toxic than its rutile form for flax (*Linum usitatissimum*). Due to their lipophilicity, rutile nanoparticles produced larger masses in the aqueous medium, resulting in less toxicity than the anatine form.

Cerium nanoxide is another metal nanoparticle that is considered as an insoluble compound in the environment. Most reports indicate that the nanomaterial is non-toxic to plants, but some other reports indicate its effects on the activity of antioxidant defense enzymes; However, seedlings may not show signs of toxicity. In one of the first reports, root growth in maize (*Z. mays*) and cucumber (*C. sativus*) increased significantly in the presence of cerium oxide nanoparticles, but was delayed in alfalfa (*Medicago sativa*) and tomato (*Lycopersicum esculentum*). However, in all four species and in all treatments, nCeO₂ concentration (0-4000 mg / L) increased the longitudinal growth of the stem [16]. The researchers also reported the effects of nCeO₂ genotoxicity on soy based on the emergence of new bands in the RAPD test [17].

Priester et al. [18] showed that high concentrations of nCeO₂ reduce growth and yield as well as stop nitrogen fixation of soybeans grown in soil. Ma et al. [19] presented an example of nCeO₂ concentration-dependent effects on Arabidopsis. Plant biomass increased significantly at 250 ppm nCeO₂ but decreased by 85% to 500-500 ppm. Also, the production of chlorophyll, anthocyanin and MDA (malondialdehyde) at high concentrations was affected. The mechanism of plant toxicity of nanomaterials is not yet fully understood [71-73]. One of the reasons for the plant toxicity of nanomaterials may be related to the release of toxic

ions, especially nanomaterials that easily produce enormously heavy metal ions and are one of the biggest challenges in Nano system studies. Decomposition of metal-based nanomaterials in the environment requires special attention. Zinc oxide (nZnO) nanoparticles are a clear example of this class of nanomaterials [74-77]. In some studies, the toxicity of nZnO has been attributed to the ions released from them, while in others, the toxicity of ZnO nanoparticles has been considered. Similar cases have also been observed for other metal-based nanomaterials such as silver, copper, copper oxide and alumina nanoparticles. None of these studies have revealed the toxicity differences between the nanomaterials themselves and the ions released from them, as well as the effect of adsorbed ions on the nanoparticles, and requires further study on the toxicity mechanisms of metal-based soluble nanoparticles. The different distribution and formation of Ce and La in cucumber indicated that nLa₂O₃ acts ironically, while the behavior of nCeO₂ is in the form of a particle or a combination of ions and particles. Further decomposition of nLa₂O₃ than nCeO₂ may be the cause of significant differences in their transfer behaviors and plant toxicity in cucumber[78-82].

3. Biotransformation of nanomaterials in plants

Nanomaterials are highly reactive and dynamic compared to bulk materials because of their unique physicochemical properties. In biological and ecosystem systems, nanomaterials inevitably interact with biological and natural compounds and undergo physicochemical changes like accidental coverage by natural organic matter and biomolecules, dissolution, and regenerative reactions [20]. Normally, metal-based nanoparticles like CuO, ZnO, and Ag may dissolve, ion release, and chemically deform by showing reaction to organic or inorganic materials (such as sulfides and phosphates) that are abundant within the environment and living organisms [21-22]. Nanomaterials may additionally physically react with mineral ions, biomolecules, and natural organic matter, reducing their aggregation and altering their surface chemistry properties. As a result, the behavior, fate, and toxicity of nanomaterials are further modulated and even determined by these processes instead of the nanoparticles themselves; therefore, the mechanisms and extent of those transformations are important for understanding and

predicting the possible risks of nanomaterials to human health and therefore the environment (Figure 2). However, to date, most nanoparticle toxicity studies have focused on the fate, distribution, and toxicity of intact or transformed nanoparticles [83-87]. Our knowledge of the kind, rate, and rate of transformation of nanomaterials within the environment and biological systems, additionally because the effect of this variation on their behavior and toxicity, remains largely obscure.

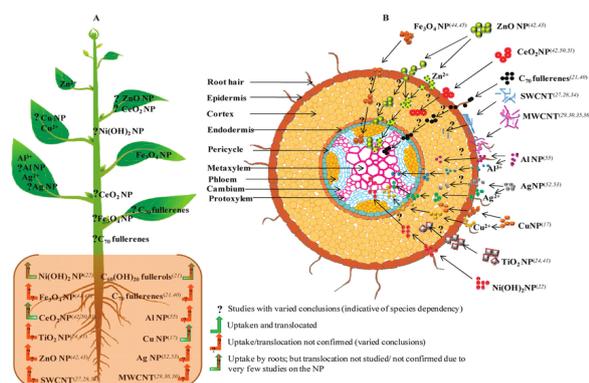


Figure 2. Absorption, transport and biotransformation path of different nanoparticles in a plant system [22].

Studies on the phytotoxicity of nanomaterials are about a decade old, while no research on plant biotransformation of nanomaterials has been conducted until the last one or two years. Lopez-Moreno et al. indicated that nZnO nanoparticles changed to Zn^{2+} in the form of nitrite or zinc acetate in soybean roots, while nCeO₂ remained unchanged [17,88-90]. However, some studies have presented contradictory results on the fact that nCeO₂ can be deformed in plants [23,1-93]. In plant systems, nanomaterials may undergo several types of deformation with the help of plant compounds. The root can produce high amounts of secretions such as mineral ions, minute molecular organic matter (such as phenols, aldehydes, amino acids and organic acids) and high molecular weight pectin (such as polysaccharides and fatty acids) that create a fine environment around the root. It produces the so-called "rhizosphere" [24,94-100]. It is well established that root secretions in the rhizosphere performs a substantial role in deciding the behavior and toxicity level of heavy metals. For instance, organic acids and pectin's in root secretions are likely to bring in unchangeable chelates with heavy metals such as Pb^{2+} , Cu^{2+} , Cd^{2+} , etc., thus restricting

their uptake into the root [25,101-103]. Nanomaterials have also the potential to experience such physicochemical deformation by interaction with root secretions; because in most cases, nanomaterials have direct contact with the roots. These types of deformations affect the ultimate fate and toxicity of nanomaterials to plants.

Many studies have been performed on the biotransformation of nanomaterials of rare earth oxides and the important role of root secretions in biotransformation has been identified. For example, in one study, large numbers of lanthanum phosphate (LaPO₄) clusters were observed in the intercellular regions (Figure 3), as well as the vacuoles and cytoplasm of cucumber roots under five-day La₂O₃ treatment, indicating significant deformation of nanoparticles in plants.

Most nanomaterials are easily taken in and massed on the root surface. This physical deformation restricts the adsorption of nanomaterials by the roots and their subsequent displacement. On the other hand, the suction of nanomaterials on the root surface causes them to directly connect with root secretions and therefore increases the likelihood of their deformation. Dissolution of metal-based nanomaterials is the most prominent deformation process that affects the behavior and fate of this type of nanomaterial in plants [49-50,104-108]. For example, nZnO biotransformation has been studied in many studies. All of these studies using synchrotron-based techniques (XANES) have shown that zinc oxide nanoparticles are not absorbed and internalized intact in plants and are often in the form of Zn^{2+} such as zinc citrate in soy, zinc phosphate in Wheat grown in sand and citrate, histamine and zinc phytate are present in groundnuts grown in the soil. At least part of the toxic effect of zinc oxide nanoparticles is due to the released Zn^{2+} ions. Organic acids secreted from plant roots play a significant part in their plant biotransformation by intensifying the dissolution of nanoparticles [109-11].

Oxidation and reduction are important reactions that usually occur in plant-soil systems. Many nanomaterials contain metal components with the ability to change capacity, which can be reduced-oxidized and subsequently bio transformed by interaction with reducing agents in plants [46-48].

Conclusive studies which have been conducted on the ecological transformation of silver nanoparticles have been summarized in a review article [20]. Though, there is only one study which has been

performed on the transformation of these nanoparticles in plants [26] which showed that silver nanoparticles are oxidized as Ag₂S or Ag₂O in the roots of *Lolium multiflorum*.

Wang et al. [13] observed that copper oxide nanoparticles are able to return from the stem to the root, during which they are partially reduced to Cu₂S and Cu₂O.

Cerium oxide nanoparticles are among the most nanoparticles whose transformation has been studied in plants. CeO₂ nanoparticles are considered as very stable nanoparticles in their environment and are used as model nanoparticles compared to other nanoparticles that are easily dissolved and unstable (such as ZnO, Ag, etc.) [27].

Zhang et al. [23, 28] found that nCeO₂ is not very firm and is likely to be lowered and transformed into Ce (III) species. Using transmission electron microscopy, it was found that large amounts of needle-like clusters were present in the intercellular and epidermal space of cucumber root cells under 21-day treatment with nCeO₂. Combined EDS analysis showed that these clusters consisted of Ce and P in an approximately 1: 1 atomic ratio and may be in the form of CePO₄[43-45]. This hypothesis was later confirmed by XANES and STXM analyzes that provided two-dimensional distribution and cluster formation (Figure 4). Mass studies with XANES showed that Ce is mainly present as CePO₄ and CeO₂ in the roots, while Ce and CeO₂ are present as carboxylates in the stems and leaves (Figure 5). The combination of the above results and subsequent simulations revealed to some extent the mechanism of deformation and displacement of nCeO₂ in cucumber. CeO₂ releases Ce³⁺ ions with the help of organic acids and reducing agents in root secretions, which are subsequently converted to CePO₄ and Ce carboxylates. Some of the released Ce³⁺ ions are immobilized by phosphates, which are found in abundance in nutrient solutions and plant tissues. The remaining of the Ce³⁺ ions are transferred from the roots to the stem or are stabilized by the carboxylic compounds of the woody vessel during transfer. This study contributed to our understanding of the behavior of nanomaterials in plants. Determining the transformation of metal-based nanoparticles in plants, like other types of nanoparticles (such as polymer nanomaterials and carbon nanomaterials), is difficult because of the substantial background of the plant matrix and the shortage of effective approaches of

spotting [112-115]. Although, there is no study which revealed the biotransformation of these nanomaterials in plants, this potential deformation should not be ignored[116-122]. A number of extracorporeal research studies have exposed the likelihood of transforming carbon nanomaterials. For example, carbon nanotubes decompose from horseradish in the presence of the natural peroxidase enzyme [29]. Graphene oxide can also be reduced by bacterial respiration [30].

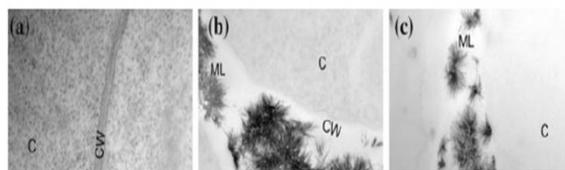


Figure 3. TEM images of different sections of cucumber root under control conditions (a) and 5-day treatments with concentrations of 2000 mg / l La₂O₃ (b) and 200 mg / l LaCl₃ · 6H₂O (c) after germination. CW: cell wall, C: cytoplasm, ML: middle lamella and IS: intercellular space [19].

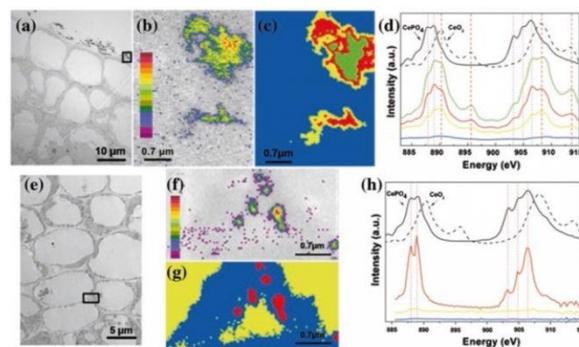


Figure 4. a and e TEM images of root cells; b and f Cerium element maps for rectangular areas in panels a and e obtained with ratios eV 886 and 888. The scale values of the images are estimated based on X-ray absorption measurements and cerium absorption coefficients (in grams per square centimeter). The calculated surface densities are between 5-10 * 1.1 to 5-10 * 4.6 and 6-10 * 2/4 to 5-10 * 8.2 g / cm², respectively. c and g are color-coded maps of cerium components in panels b and f obtained by STXM Ce M post-elevation analysis[41-42]. The order of cerium is green> red> yellow, blue indicates the absence of cerium. Panels d and h are the XAFS spectra obtained from panels c and g, respectively. The upper black spectrum is for standard compounds, while the lower color spectrum

is for root samples. Vertical red dotted lines represent specific CePO_4 peaks and dashed lines indicate nCeO_2 peaks [28].

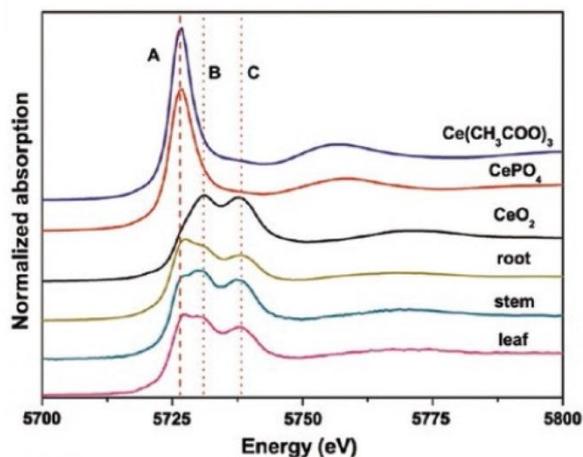


Figure 5. XANES Ce LIII edge (eV 5723) spectra of cucumber roots, stems and leaves treated with 2000 mg / l CeO_2 nanoparticles for 21 days. Spaced lines and vertical dotted lines represent Ce (III) and Ce (IV), respectively [28].

4. Conclusions and perspectives

Excellent plants are sensitive to contaminants such as nanomaterials engineered in the plant-soil system due to accidental discharge of nanomaterials into the environment or the deliberate application of nanotechnology in agriculture and soil refining. Plant toxicity, accumulation and potential magnification of these nanomaterials in the food chain have raised concerns not only for the environmental system but also for human health. Although many efforts have been made to understand the plant toxicity of nanoparticles, our understanding of the mechanism of toxicity and its relationship to the physicochemical properties of nanoparticles is still limited. Important issues to be considered in future research are: 1) A comprehensive study of the physicochemical properties of nanomaterials, because the behavior and toxicity of nanomaterials are strongly influenced by their physicochemical properties such as size, morphology, surface charge and crystal structure. The results obtained from different laboratories may be different even for one type of nanomaterial. Therefore, accurate and complete characterization of nanomaterials before evaluating their toxicity is a prerequisite for understanding their behavior and toxicity in plants; 2) More research is needed on the biotransformation of nanomaterials. Nanomaterials are not able to retain their original chemical form

completely, and most of them undergo some kind of transformation in plants. To understand the mechanism of toxicity and behavior of nanomaterials, it must be noted that their toxicity is due to their intact form or deformed forms; 3) It cannot be denied that short-term studies are a way to understand the mechanism of toxicity and behavior of nanomaterials in plants, but the effect and long-term presence of nanomaterials in plants grown in natural habitats should be evaluated to understand the response of plants to permanent nanomaterials. And also have the life cycle of nanomaterials.

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