

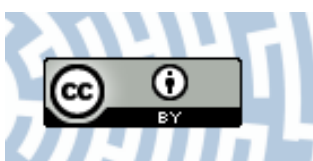


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Author: Anna Milewska-Hendel, Robert Gawecki, Maciej Zubko, Danuta Stróż, Ewa Kurczyńska

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REVIEW

Diverse influence of nanoparticles on plant growth with a particular emphasis on crop plants

Anna Milewska-Hendel^{1*}, Robert Gawecki¹, Maciej Zubko², Danuta Stróż², Ewa Kurczyńska¹

¹ Department of Cell Biology, Faculty of Biology and Environmental Protection, University of Silesia in Katowice, Jagiellońska 28, 40-032 Katowice, Poland

² Institute of Materials Science, University of Silesia in Katowice, 75 Pułku Piechoty 1a, 41-500 Chorzów, Poland

* Corresponding author. Email: anna.milewska@us.edu.pl

Abstract

The article describes the current knowledge about the impact of nanoparticles on plant development with a particular emphasis on crop plants. Nanotechnology is an intensively developing field of science. This is due to the enormous hopes that have been placed on the achievements of nanotechnology in various areas of life. Increasingly, it has been noted that apart from the future benefits of nanotechnology in our everyday life, nanoparticles (NPs) may also have adverse effects that have not been sufficiently explored and understood. Most analyses to date have been focused on the influence of nanomaterials on the physiological processes primarily in animals, humans and bacteria. Although our knowledge about the influence of NPs on the development of plants is considerably smaller, the current views are presented below. Such knowledge is extremely important since NPs can enter the food chain, which may have an influence on human health.

Keywords

apoplast; crop plants; development; nanoparticles; plants; plasmodesmata; symplast

Nanotechnology, nanomaterials, and nanoparticles

The father of nanotechnology is the Nobel Prize-winning physicist Richard Feynman who pointed out in his lecture in 1959 that it was possible to obtain nanomaterials [1], but the first time the term “nanotechnology” was defined by Norio Taniguchi in 1974 [2]. The definition of nanomaterials is still evolving and currently assumes that nanoparticles (NPs) are insoluble or biopersistent materials (objects) that are produced intentionally and that have one or more external dimensions or an internal structure on a scale from 1 nm to 100 nm [3]. Within this group of materials are NPs, which have at least two dimensions on the nanoscale [4]. The basis of the 100 nm limit is the fact that the novel properties that differentiate particles from bulk material typically develop at a critical length scale of under 100 nm [5]. However, it must be taken into account that according to the researchers the current limit of 100 nanometers, which is the basis for the dimensions of NPs, is now out of date and that any new regulations that are created should be based on the newer, more advanced systematics. This is because sometimes additives have almost identical characteristics as “normally” produced chemicals when reduced to the nanoscale and that the synthesized NPs of the same material have completely different properties depending on the size of the particles.

Nanotechnology is an innovative and promising field of interdisciplinary studies. It opens up a wide range of possible uses in various areas of industry and science, such as medicine, pharmacology, electronics, biology, and plant breeding [6,7]. The rapidly developing commercial and industrial usage of nanotechnology has led to the increased emission of nanoparticles into the environment and inevitably to different effects on living organisms, which have been analyzed less in the case of plants [8]. The current state of the art indicates that nanomaterials may cause adverse effects and that these are not yet fully explored and understood. A new field of knowledge, “nanotoxicology”, which has recently developed [3], has confirmed the need to analyze the influence of nanomaterials on living organisms. However, before the degree of toxicity of nanoparticles can be determined, detailed analyses of the uptake of nanoparticles by living organisms and their movement within the body at different levels of organization – the organs, tissues, cells, and molecular level – should be described.

Although nanoparticles have always been present in the environment and their natural sources include, among others, active volcanoes, forest fires, or dust storms, the development of nanotechnology has contributed to a significant increase in their presence in the environment because they are either produced intentionally and/or as a result of technological processes such as welding, metal smelting, soldering, in combustion engines, in heating and power plants, cooking, grilling, or in laser office devices [9]. Nanomaterials are used in many different industrial fields such as electronics [10,11], medicine [12,13], cosmetology, agriculture, the food industry, and construction [14]. The volume of world production of nanomaterials was 2000 tonnes in 2004 and according to forecasts this number is predicted to increase more than 25 times for the period of 2011–2020 [15].

The rapidly increasing numbers of reports on the accumulation of nanomaterials in the environment point out the fact that the fate of NPs in the environment is not fully known and understood [16]. Analyses have mainly related to the study of the impact of nanomaterials on animals and bacteria and our knowledge about their effects on plants is very poor. Analysis of the effects of nanoparticles on living organisms cannot be compared, for example, to the effects of heavy metals that have been tested, since nanoparticles are different than the basic material in the atomic structure and their physico-chemical and biological properties are also different [17]. The need to identify any threats that are connected with developing nanotechnology is beyond dispute.

Nanoparticles and plant growth

In recent years, numerous studies have been conducted in order to analyze and describe the influence of nanoparticles on plant growth and development and many of them examined this problem on the example of crop plants. Many studies have been performed on the impact of NPs on the physiological/metabolic processes (for review see [18,19]) that directly influence plant growth and that is why this paper is only devoted to the effects of NPs on plant growth and development that have been described.

An investigation of the response of six crop species: barley, maize, rice, soybean, switchgrass, tomato, and tobacco cell cultures to exposure to single-walled carbon nanohorns (SWCNHs) showed that these types of nanomaterials accelerate seed germination of some crops studied and enhance the growth of different organs of corn, tomato, rice, and soybean [20]. Moreover, the growth of tobacco cells increased in response to SWCNHs. Analysis at the genetic level indicated that SWCNHs were able to affect the expression of a number of tomato genes that are involved in the cellular and metabolic response of cells to stress conditions [20]. Jasmine rice that had been treated with different sizes and concentrations of silver nanoparticles (AgNPs) showed a positive correlation between the size of the nanoparticles and a decrease in seedling growth [21]. It was also shown that accumulations of AgNPs were higher in the plant tissues of rice that had been treated with smaller AgNPs (20 nm diameter) and that the nanoparticles were retained in the roots rather than being translocated to the leaves [21]. Other examples of studies that have described the effects of nanomaterials on

plant development include those that have been conducted on the roots and above-ground parts of red spinach, lettuce, and cucumber. These researches showed a considerable reduction in root growth after exposure to 1000 mg/L and 2000 mg/L of carbon nanotubes [7]. Red spinach and lettuce were the most sensitive to such treatment, while rice and cucumber were less sensitive. On the other hand, chili, lady's finger, and soybean were neutral to such treatment [7]. The influence of five different types of nanoparticles such as multi-walled carbon nanotubes, aluminium, alumina, zinc and zinc oxide on seed germination and root growth of radish, rape, ryegrass, lettuce, corn, and cucumber has also been studied. The results obtained showed that seed germination was only affected in the case of ryegrass and corn. The reduction in root growth varied depending on the species and the type of nanoparticles [22]. Analysis of the effect of nano-CuO on rice showed that seed germination was significantly reduced and that there was damage to the root cells, an increase in H₂O₂, an accumulation of proline, and a decrease in the level of carotenoids [23]. The response of asparagus to AgNPs included an increase in the content of ascorbate and chlorophyll [24]. Treatment of soybean with nano-iron particles caused an increase in dry weight and productivity [25]. The application of TiO₂-NPs of different sizes to wheat plants showed that even when NPs were accumulated within the root, there was no influence on seed germination, biomass and transpiration nor there was any modification of photosynthesis or the induction of oxidative stress, and the stimulation of root growth was even observed [26]. A rapid inhibition of leaf growth and transpiration was detected in the case of *Zea mays* L. seedlings after they had been exposed to naturally derived bentonite clay or industrially produced TiO₂ nanoparticles [27]. ZnO nanoparticles in *Lolium perenne* caused a reduction of biomass, shrinkage of the root, and cytological changes in the root cortical cells [22]. Other studies have shown that different concentrations of AuNPs have no effect on seed germination of barley, but they had a strong impact on plant growth, which resulted in lower production of biomass [28]. Analysis of single-bilayer graphene oxide sheets of different sizes supplied at different concentrations showed a dose-dependent effect during the germination of *Vicia faba* L. [29]. A significant negative impact of a high concentration of graphene oxide was indicated by a reduction in the growth parameters of *Vicia faba* plants, although low concentrations improved the health status of the plants [29].

Field experiments on *Brassica juncea* that had been treated with AuNPs showed a positive effect on plant height, stem diameter, and the number of branches [30]. In *Cucurbita pepo* plants that had been treated with AgNPs and CuNPs, a decrease in growth was observed [31]. Recently, some studies have been performed on the influence of AgNPs on radish sprouts [18]. It was shown that seed germination was not affected although changes in the chemical composition of the cell wall were observed, which confirms that changes due to the influence of NPs take place at the cellular and molecular levels. Treatment of tomato seeds and seedlings with CoFe₂O₄ NPs did not decrease the germination of seeds [32]. The same study showed that such treatment did not have a negative influence on root growth [32].

Quantum dots (QDs) are other nanomaterials that are widely used as promising tools for imaging the structure of cells and in vivo cell tracking. Studies on the influence of QDs on a *Medicago sativa* culture in vitro showed a negative influence of QDs and that the response of plant cells was dose-dependent [33]. Rice root growth was enhanced by silica that had been coated with QDs [34], however, germination was inhibited [35]. To date, studies that are devoted to the analysis of the influence of QDs on plants are rather limited and this means that our knowledge is incomplete. One gets the impression that the harmful effects of QDs on plant development is stronger in comparison to other types of NPs. This indicates that more studies are needed and/or a limitation of their wide scale application is necessary.

In the case of crop plants, knowledge about the bioaccumulation and accumulation of nanoparticles in food crops is limited, although it is important to point out the possibility that nanoparticles may have an influence on human health.

The literature data presented above leads to the conclusion that the results that have been obtained to date are inconsistent (which is probably due to the fact that only a small number of studies have been done, only a few species have been investigated, different growth conditions have been studied and different nanoparticles have been used), and therefore generalizations about the effects of nanoparticles on plant growth

processes cannot be made. At present, a picture emerges which shows that under the influence of nanomaterials there may be stimulation, inhibition, or no effect on the growth processes and thus on plant development, including crop plants.

Routes of NPs entry and movement within the body of plants

A study of *Cucurbita maxima* plants that had been treated hydroponically with Fe₃O₄ showed the absorption, translocation, and accumulation of the nanoparticles in the plant tissues [36], but the same treatment of *Phaseolus limensis* plants did not cause the same reaction as in the case of pumpkin [36], thus indicating that the response of plants to nanoparticles is variable and depends on the plant species, age and internal and external conditions.

In rice plants after treatment with fullerene C70, uptake, translocation, and even its presence in seeds that had developed on the treated plants were described [37].

Catharanthus roseus protoplasts were used to study the influence of multi-walled carbon nanotubes (MWCNTs). It was shown that these nanomaterials penetrated the cell membrane of an identified endosome-escaping uptake mode and that MWCNTs shorter than 100 nm were present in different cell compartments, including the nucleus, plastids, and vacuoles [38]. Artificial polystyrene nano-spheres (40 nm) and CdSe/ZnS quantum dots (20 nm) were taken up by sycamore protoplasts via fluid-phase endocytosis [39]. A very interesting result from these studies was the demonstration of the sequestration of different nanoparticles in different cell organelles [39]. It should be taken into consideration that the response and absorption of NPs or QDs in the system of a protoplast culture may be different in comparison to a cell with walls.

Carbon nanotubes (CNTs) were detected inside tomato seeds, which suggests that the seed coat is not a barrier for this nanomaterial [40]. The results obtained may suggest that CNTs support water uptake inside seeds, which can explain the positive influence on the seed germination and growth of tomato seedlings that was detected [40].

It is widely postulated that nanoparticles are collected and accumulated in plants, although the mechanism of such accumulation and the route of the movement of nanoparticles have not yet been fully explained and documented. In studies of this kind, in which nanoparticles are served on the surface of an organ, the chemical nature of the wall and wall pore size, which determine the possibility of nanoparticles breaching the wall, the first barrier in the way environment-plant must be taken into account [41]. The above values indicate that particles with a diameter greater than the cell wall pore size cannot breach the wall barriers. However, much literature data has indicated the localization of nanoparticles inside the plant body. It has been postulated that NPs are adsorbed on plant surfaces [3]. Other ways through which nanoparticles enter the body of plants is not fully understood. It is obvious that NPs can be deposited on aboveground plant parts such as epicuticular cavities or between trichomes. NPs can also enter a plant through the stomata. However, the NPs should be deposited on the outer periclinal walls of the epidermal cells, at least those NPs whose dimensions are greater than the cell wall pore size, and such behavior should also characterize the underground plant organs.

Treatment of *Triticum aestivum* with TiO₂-NPs indicated that roots can only accumulate NPs up to 140 nm in diameter and that NPs are accumulated in the wheat root parenchyma, but translocation to other root tissues, especially to the stele, was not detected, which means that translocation to the shoot is not possible [26].

An accumulation of nanoparticles in the roots of pumpkin [36], soybean, tomato, cucumber, maize, alfalfa, rice, and bean has been detected [42–45]. An interesting conclusion/question that arises from these experiments concerns the mutual correlation between the dimensions of nanoparticles and the pore size of the cell wall, because the statement presented above indicates that NPs can be accumulated in the organs of plants even when their dimensions exceed the cell wall pore exclusion limit.

In *Arabidopsis thaliana* (not a crop plant, but it is important due to the possibility of the use of this plant in explaining the molecular mechanisms that underlie the

influence of NPs on plant behavior because of the large number of mutants and transgenic lines), different doses of Ag nanoparticles of different sizes (20, 40, and 80 nm) affected plant growth and the location of the nanoparticles in tissues and cells was investigated [46]. Studies have shown that under the influence of nanoparticles, the first macroscopically visible sign was a browning of the surface of the root cap cells. Analysis of the distribution of these nanoparticles in the plant showed their presence in the root cap cells and other tissues of the root.

Some authors have postulated the involvement of plasmodesmata (PD) in the movement of NPs within the body of a plant [37,46]. In rice treated with carbon nanoparticles 40–70 nm in size, the PD are engaged in the movement of nanomaterials between the cells of different tissues [37].

In regard to the participation of PD in the movement of nanoparticles with such a large diameter (40 nm), additional studies are necessary. The diameter of the plasmodesmata is 25–50 nm, although it is necessary to take into consideration that the transport channels within the cytoplasmic sleeve have a diameter of between 1.5 and 4 nm [38]. When we assume that nanoparticles move through cytoplasm by diffusion, they should pass the PD via the above-mentioned microchannels. This means that 40 nm nanoparticles cannot go through the PD, at least via the mechanism of simple diffusion. If this is the case, this means that perhaps NPs trigger new mechanisms that allow such a movement through the PD or some other possibilities must be considered as the pathway of the movement of NPs within the body of a plant.

From the above-mentioned results, it appears that nanoparticles can penetrate the cell wall of plants. The most important questions that arise from such results concern the mechanism that regulates a cell's "answer" to the presence of nanoparticles. Namely, if NPs can penetrate the cell wall, which is characterized by a much smaller pore diameter than the dimensions of the particles, it suggests that some adaptive mechanism must have been developed that led to such a modification of the cell wall which allows the penetration of NPs. Without answers to the above questions, our knowledge will not be complete. To answer these questions, more investigations on more plant species and types of nanoparticles must be performed, before any general conclusions can be formulated.

Conclusion and future prospects

Nanotechnology can have positive and diverse effects on living organisms. Because of the possibility of a negative impact of nanoparticles on plant growth, it is necessary to carefully examine the relationship of NPs and plant organisms. Such knowledge is important from both an economic and societal point of view. Thus, the fate of various NPs in different plant species requires further investigation.

Undoubtedly, nanotechnology can provide "tools" to improve the properties of plants, their productivity and their resistance to many internal and external factors. However, because of the possibility of harmful effects of NPs on plants, there is a need for a great deal of research that would allow wise use of this new technology. Such a need is due to the fact that the influence of NPs on plant growth can be positive or negative and that this depends on the type of nanoparticle (as it determines their physical and chemical properties), the dosage of NPs, the time of treatment, the plant species, the stage of development, and many other factors (Tab. 1; for review see [47]).

From the results presented above, it appears that much more work is needed to understand the impact of NPs on plants at different organization levels – from morphology, histology, physiology and biochemistry to the molecular level. In order to understand the interaction of NPs with the different molecules that are present in plant cells, studies on the analysis of any changes in gene expression under the influence of nanoparticles are also necessary.

Tab. 1 Summary of the influence of some nanoparticles on some crop plants.

Nanoparticles	No influence	Positive influence	Negative influence	References
Oxide nanoparticles				
Silicon dioxide	<i>Triticum aestivum</i>	<i>Triticum aestivum</i>	<i>Zea mays</i>	[26,27]
Zinc oxide			<i>Zea mays</i> , <i>Lolium perenne</i>	[22]
Iron magnetite	<i>Cucurbita maxima</i>	<i>Glycine max</i>		[26,36]
Nano-CuO			<i>Oryza sativa</i>	[23]
Carbon nanomaterials				
Carbon nanohorns		<i>Zea mays</i> , <i>Solanum lycopersicum</i> , <i>Oryza sativa</i> , <i>Glycine max</i>		[20]
Carbon nanotubes			<i>Spinacia oleracea</i> , <i>Lactuca sativa</i> , <i>Cucumis sativus</i> , <i>Solanum lycopersicum</i>	[7,40]
Graphene oxide (low or high) concentration		<i>Vicia faba</i>		[29]
Metal nanoparticles				
Gold	<i>Hordeum vulgare</i> – seed germination	<i>Brassica juncea</i>	<i>Hordeum vulgare</i> – biomass production	[28,30]
Silver		<i>Asparagus officinalis</i> , <i>Oryza</i> (jasmin rice)	<i>Cucurbita pepo</i> , <i>Raphanus sativus</i>	[18,21,24,31]
Zinc			<i>Lolium perenne</i>	[22]
Copper			<i>Cucurbita pepo</i>	[31]
Quantum dots				
Mercaptopro-panoic acid coated CdSe/ZnS			<i>Oryza L.</i> – seed germination, <i>Medicago sativa</i>	[33,35]
Silica coated CdTe		<i>Oryza sativa</i>		[34]

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Zróżnicowany wpływ nanocząstek na wzrost roślin, ze szczególnym uwzględnieniem roślin uprawnych

Streszczenie

W artykule przedstawiono aktualny stan wiedzy na temat wpływu nanocząstek na rozwój roślin, ze szczególnym uwzględnieniem roślin uprawnych. Nanotechnologia jest intensywnie rozwijającą się dziedziną nauki, co wynika z ogromnych nadziei pokładanych w osiągnięcia nanotechnologii w różnych dziedzinach życia. Coraz częściej wskazuje się, że poza dobrodziejstwami jakie w codziennym życiu przynoszą nam nanomateriały, mogą one również wywoływać niepożądane efekty, a te nie są jeszcze w pełni zbadane i wyjaśnione. Większość przeprowadzonych do tej pory analiz koncentruje się na wpływie nanomateriałów na procesy fizjologiczne przede wszystkim u zwierząt, ludzi i bakterii. Wiedza na temat wpływu nanocząstek (NPs) na rozwój roślin jest znacznie skromniejsza, a obecne badania i poglądy przedstawione zostały w poniższej pracy. Wiedza ta jest bardzo ważna, ponieważ NPs mogą wejść do łańcucha pokarmowego, co może mieć wpływ na zdrowie ludzi.