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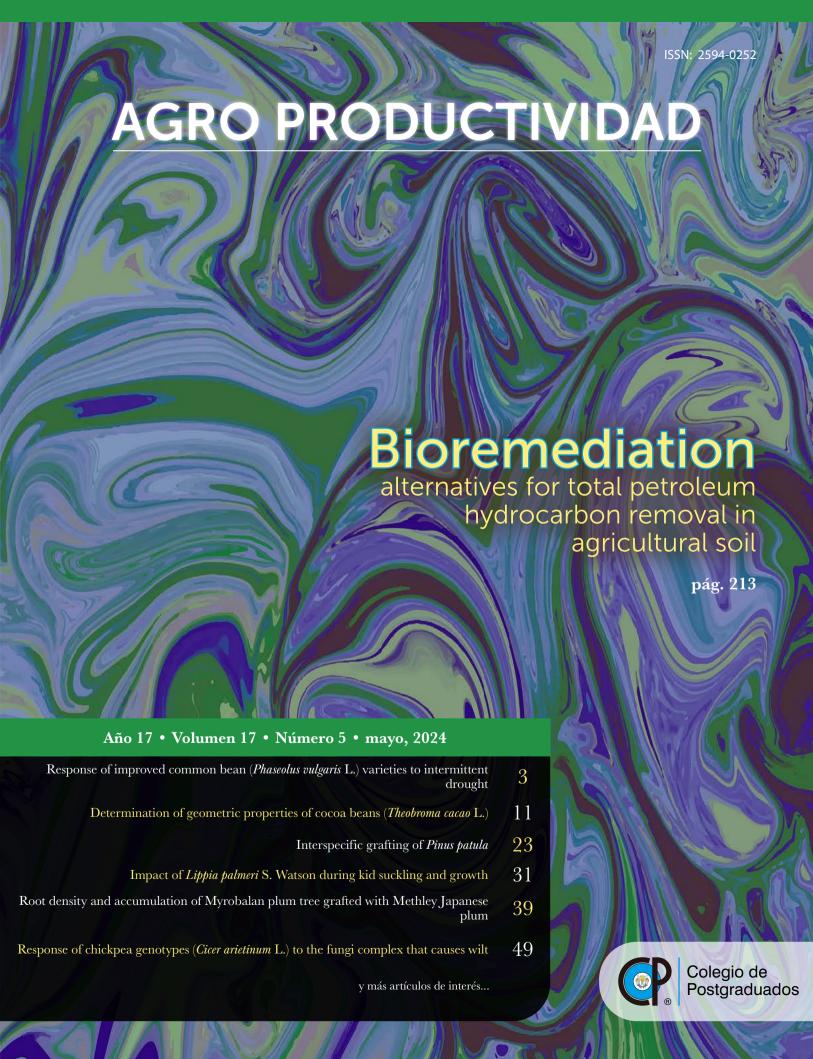
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Biofortification of forages through the application of selenium nanoparticles

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ABSTRACT

Objective: This review paper examines the application of nanoparticles in agriculture, through biofortification with Se NPs in plants.

Methodology: A comprehensive review of the literature on the use of Se NPs in plant biofortification was made.

Results: According to documented studies, the foliar application of NPs significantly improves the morphological and physicochemical characteristics of plants. They promote root development, plant growth and increase the content of raw protein and ether extract in forages. An increase in the production of amino acids and essential fatty acids is also observed.

Limitations: The findings are promising, but there is still slight research on how NPs affect the environment and the safety of their use in the food chain. Further studies are needed to fully understand the long-term effects.

Conclusions: The application of Se NPs for biofortification can improve the nutritional quality of forages, contributing to more efficient and sustainable livestock production. However, to ensure the safety of its implementation it is necessary to continue investigating.

Keywords: sustainable agriculture, animal nutrition, nanotechnology.

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Livestock farming faces the challenge of increasing its production in the face of expectations of world population growth due to the products and by-products that come from these animal sources and are required to feed the population, with forecasts of reaching close to 10 billion people in 2050 [1]. The expectations to meet this challenge are high, but this requires more efficient livestock production with fewer polluting emissions, both greenhouse gases and substances that contaminate soils and waters.

Forages are the primary support of sustainable agriculture and constitute one of the leading resources for the success of the animal production system [2]. Selenium (Se) is an essential micronutrient for animals; soil typically contains low levels of Se. Plants can benefit their growth and development by increasing their tolerance to different types of stress and synthesizing phytochemicals with antioxidant properties [3, 4]. Therefore, Se



may be present, but in chemical forms, it is not available to organisms; as a result, Se content is deficient in plants, animals, and other organisms in the food chain [5, 6]. Biofortification increases the nutritional value of food crops by increasing the mineral and vitamin content in the plants [7]. Biofortification has been carried out in agricultural products (wheat, sorghum, peas, rice, and vegetables) and some forage varieties, such as alfalfa, due to the lack of Se in the soil [8]. Several biofortification methods are used for plants, but the foliar application method is more effective than soil fertilization [9]. In recent years, the benefits that Se provides for the health of farm animals have been demonstrated, such as improving reproductive performance and immunological and homeostatic function [10]. Worldwide, several studies relate the absence of consumption of this element to decreased development, growth, and reproductive and metabolic problems, especially in ruminants [11, 12].

Nanotechnology and agriculture

Nanotechnology is a multidisciplinary scientific research area that works on the design, characterization, manufacturing, and application of structures created by controlled size and shape manipulation at the nanometric scale (less than 100 nm) that produce complex systems in which they modify some specific characteristics [13]. Due to the above, many applications have been developed, including medicine, genetics, pharmaceuticals, and agriculture. In 2013, the FAO published information on the state of the art of nanotechnology in food and agriculture. Its report documented ten-year research focused on sustainability and solutions to environmental problems. These investigations focus on releasing active ingredients (disease management and crop protection), minimizing fertilization loss and increasing yield, and producing bionano compounds from traditional crops [14]. Reports of such research on possible applications in agriculture have been both positive and negative. However, there are reports in which favorable results have been found, for example, in the case of fertilization, germination, growth promoters, sensors of contaminants and other substances, and recovery and treatment of soil and water [15, 16]a. Our knowledge about the interactions of nanoparticles for agricultural use with the environment still needs to be improved. Due to its complexity, there is still a long way to go to understand it completely. The UN 2013 raised the need to increase food production since by 2050, there is an estimated population of 9.1 billion people. There is currently a trend towards producing energy crops derived from efforts to mitigate climate change and due to the imminent reduction of hydrocarbon reserves. At the same time, in the expert forum organized in 2009 by FAO, "How to feed the world in 2050," it was proposed that it will be necessary to increase cereal production by 70% by 2050 [17]. Nanotechnology could provide tools for modern agriculture and even solve future problems related to food production and energy demand, focusing on sustainability. However, some NPs have been shown to affect the development and yields of crops.

Nanotechnology and caring for the environment

The current food system worldwide is not sustainable and, for decades, has caused environmental damage, including greenhouse gas emissions, the irrational use of drinking water, the contamination of soils and aquifers by nitrogen and phosphorus, the use of pesticides, etc. However, recent research on environmental nanotechnology shows it can be a valuable tool for reducing environmental pollution. [18] developed nano encapsulates and nanocomposites for food and feed additives, biocides, pesticides, and contact materials. [19] applied nanomaterials as catalysts in phytoremediation processes and used stabilizers to improve their performance.

Applications of nanoparticles in agriculture

Applied science in agricultural systems presents a series of adversities and a wide range of challenges, such as a decrease in crop yields, low efficiency in the use of macro and micronutrients, a decrease in soil organic matter, deficiencies in multi-nutritional foods, climate change, the reduction in the availability of arable land, as well as the scarcity of water and labor for the countryside [20]. Various experiments have been carried out regarding the agricultural sector to know the optimal size, shape, and concentration of NPs to be applied to plants trying to improve their penetration and vascular translocation through the vascular bundles of the xylem and phloem.

The literature mentions that concentrations or doses less than 5 ppm of NPs can promote more significant plant growth. The size of the NPs is a relevant factor that significantly intervenes in the action of their penetration and translocation inside the plant tissues; therefore, the larger the size of the NPs, the less penetration will be into the vascular system in the plants when foliar spray techniques are used [21]. When applied to plant leaves, NPs can penetrate plant tissues as an aerosol or spray.

Evidence shows that 14.7% of the nanomaterial applied to plants is lost when using the spray or aerosol technique, compared to 32.5% or more with conventional agricultural products applied traditionally. An alternative is the application of foliar spray for the manipulation of nanoscale materials. When fertilizers are applied in a foliar way, they can easily penetrate the interior of the plants through the opening of the stomata, which have micrometric sizes fluctuating from $10 \,\mu\text{m}$ to $60 \,\mu\text{m}$ [22, 23].

Pastures and forages

Mexico has the eighth-largest cattle population in the world, with approximately 31 million heads, representing 2.31% of the world herd. However, Mexico has a greater diversity of ruminants, such as sheep and goats [24].

In the tropics of Mexico, the exploitation of livestock, specifically cattle, is carried out mainly by grazing native grasses and introducing grasses for livestock feeding. Pastures are grass and legume plants that grow in the pasture and serve to feed livestock. At the same time, forage is defined as any grass or legume harvested to be supplied as food to animals, whether green, dry, or processed. They can have different nutritional characteristics (hay, silage, stubble, saccharin, ammonification) [25, 26].

Selenium and Selenium nanoparticles

Selenium (Se) is a trace element that is scarce in the Earth's crust and is distributed in all parts of the Earth, being an element of volcanic origin. It accompanies sulfur and is found in clay soils (Figure 1). It is a by-product of the industrial manufacture of sulfur and sulfuric acid. Chemically, it forms with hydrogen and oxygen, the same compounds as sulfur H₂SeO₄, H₂SeO₃, H₂Se, and SeO₂. It can also replace sulfur in certain amino acids (cysteine, methionine) [27]. Selenium is an element found constantly but in small quantities in animal tissues. Selenium is one of the essential micronutrients for animals and is very important for animal nutrition [28]. Selenium is critical because it is necessary to form proteins and be a cofactor of antioxidant enzymes such as glutathione peroxidase (GPX), which reduces reactive oxygen species (ROS). Se deficiency in mammals causes physiological and nutritional deficiencies and pathologies since Se forms at least 25 selenoproteins that fulfill antioxidant, antiviral, and antitumor functions [29-31]. The synthesis and application of selenium nanoparticles (SeNPs) present several advantages, including chemical stability, biocompatibility, low toxicity, increased bioactivity, improved targeting, and versatile means to control the release profile.

Biofortification of plants with selenium nanoparticles

Biofortification increases the nutritional value of food crops by increasing the mineral, vitamin, and mineral content of conventional plant crops. This action is called agronomic biofortification. Biofortification involves the application of different minerals or nutrients in a foliar or edaphic manner, taking advantage of plant management, soil factors, and plant characteristics to obtain a higher content of critical micronutrients in the edible portion of the plant. The importance of Se in plant nutrition has been known for a long time. There are increasing ways to use Se NPs in agriculture, such as edaphic form (addition of Se to soil), hydroponic, and plant foliar cultivation. Plants can benefit from the use of Se NPs in a variety of applications, including (1) as care and management of pests and diseases caused by infectious microorganisms such as bacteria and fungi, (2) as near-essential trace elements, through the promoting plant biochemical pathways and thereby refining the

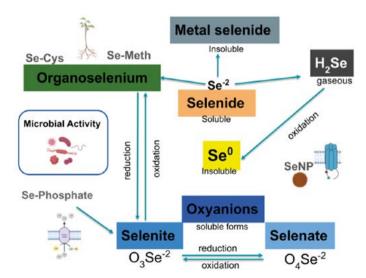


Figure 1. Changes in Selenium and selenium nanoparticles (SeNP) in the soil environment. Selenium (Se), a natural soil element, is found in the environment and organisms in two forms: inorganic (selenate, selenite, elemental Se and selenide) and organic (mainly Se-Meth selenomethionine and Se-Cys selenocysteine).

progress, yield and nutritional value of crops, (3) biofortifying crops using Se to increase their content, (4) alleviating abiotic stress and (5) increase the nutraceutical quality of consumable foods [32].

Uptake of Se NPs in plants

Concerning the phyto-uptake of SeNPs and their translocation, determining the safety and toxicity of NPs provokes a deep understanding of their uptake in plants (Figure 2). There are different ways for the plant to capture this trace element in the form of NP. An absorption mechanism of SeNPs occurs through the cell wall and penetration of the plasma membrane. Only NPs with sizes smaller than 100 nm can enter through the different pores (stomata) and pass through the cell wall successfully. The plant cell walls act as a barrier, preventing the entry of external substances or materials, including NPs, into the plant cell walls [33-35]. It has been shown that NPs can adhere to surfaces such as plant roots and could influence plant chemical and physical absorption [33]. One of the explanations that demonstrates the most outstanding scientific solidity is the translocation of nanomaterials, which can move intracellularly and extracellularly between plant tissues until they reach the xylem [36]. Once the NPs are within the plant's vascular system, the designed NPs could be transported to the aerial parts along with plant water transpiration and nutritional flux in nutrient transmission. [37] applied selenium nanoparticles in grass plants, Festuca arundinacea, reporting significant differences in biomass parameters, root length, and leaf length (p<0.05) between the treatments (1.5, 3.0, and 4.5 ppm NPsSe) and the control. The increase in root length, leaf length, and biomass production of se-treated plants could be attributed to the application of selenium nanoparticles; Se stimulates the growth and development of plants because it improves the synthesis of photosynthetic pigments and organogenesis [3, 38]. Some authors [39] observed that foliar application of different amounts of SeNPs affected morphological characteristics such as shoot length, root size, and fresh weight of the bean plant. In other study [40] reported that by applying SeNPs foliar, plant height, fresh weight of the shoots, and root size were increased when concentrations of 10 mg SeNPs were used in wheat plants, attributing that the SeNPs stimulated the organogenesis and growth of the plant roots.

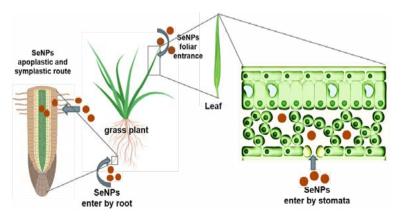


Figure 2. Uptake of Selenium NPs in plants, foliar, and soil route of adsorption.

Proximal composition of fortified forages

This research [41] found significant differences (p<0.05) between the treatments in the variables of crude protein (CP), ether extract (EE), crude fiber (CF), neutral detergent fiber (NDF), acid detergent fiber (ADF), and Nitrogen Free Extract (NFE). The increase in PC content as a plant response to foliar biofortification of NPsSe could be manifested because Se promotes the increase in the synthesis of sulfur amino acids (Cvs and Met) and selenium-aminoacids such as SeCys and Semet, which are incorporated into proteins. However, there is also evidence that other non-protein amino acids are synthesized, such as \(\gamma\)-glutamyl methyl selenium-cysteine (\(\gamma\)-gluMetSeCys), methyl-SeCys and methyl-Semet ([32, 42], where Se was used as a fertilizer, and increased protein was obtained in the root and leaf of alfalfa (Medicago sativa L.). Similarly, [43] found that foliar spraying of selenate produced an increase in the levels of protein content and total root nitrogen in radish (Raphanus sativus). Other research [44] found that foliar supplementation of sodium selenate in wheat plants (Triticum aestivum variety BRS 264) increased total nitrogen (up to 20%) compared to the control. In addition [45] reported that fertilization with Se promotes the increase in fatty acids (oleic, linoleic, and linoleic), increasing the concentration of lipids; however, the metabolic pathway is unclear. Furthermore [46] found that foliar application of sodium selenite and selenate in four rice genotypes (Ariete, Albatros, OP1105, and OP1109) increased the content of total lipids, as well as increased the concentration of oleic acid (C18:1), linoleic (C18:2), and palmitic acid (C16:0). Also, [47] mention that the synthesis of compounds such as the content of starch, total soluble sugars, and reducing sugars are enhanced, caused by the possible increase in the activity of carbohydrate metabolism enzymes. The results obtained in this study are similar to those reported by [42], where Se was used to improve carbohydrate metabolism, finding an increase in these compounds in alfalfa root and leaf (Medicago sativa L). In the same way, [44] found that the carbohydrate content in wheat plants increased when biofortified with selenium, showing this trend in each crop.

Use of nanoelements in Animal Nutrition

Nanotechnology and its applications can revolutionize agricultural production. This technology has had various applications in animal production, including the use of new molecular and cellular tools for animal reproduction, preservation of the animal's identity from birth to the consumer's table ("traceability"), biosecurity of food from animal origin, a better understanding of the phenomena that govern animal nutrition from dietary ingestion to the uptake and use of nutrients, and others [48].

In animal nutrition, as in other areas, it is possible to apply nanotechnology to obtain information about a nutrient or bioactive component and its release at specific sites of action, greater availability and maintenance of adequate levels for more extended periods, greater use of food [49]. Minerals are one of the most widely used supplements in animal nutrition; However, the form in which these minerals are found influences their bioavailability, so if they have low bioavailability, the animal will not use them correctly. An example is Selenium (Se), an essential trace element with a narrow range of beneficial and toxic effects. Recently, nano selenium has attracted the attention of

many researchers due to its high bioavailability and low toxicity. Nonmetric particles exhibit novel characteristics, such as a larger contact surface, high surface activity, high absorption capacity, and low toxicity.

CONCLUSIONS

Biofortification with Se NPs benefits forage grass's morphological and physicochemical characteristics. Se NPs improve root development and synthesis, growth, and grass yield. Applying Se NPs increases the content of crude protein and ether extract.

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