Journal of Hazardous Materials 346 (2018) 62–72

Contents lists available at ScienceDirect

Journal of Hazardous Materials

journal homepage: www.elsevier.com/locate/jhazmat

Plant extract–mediated green silver nanoparticles: Efficacy as soil conditioner and plant growth promoter

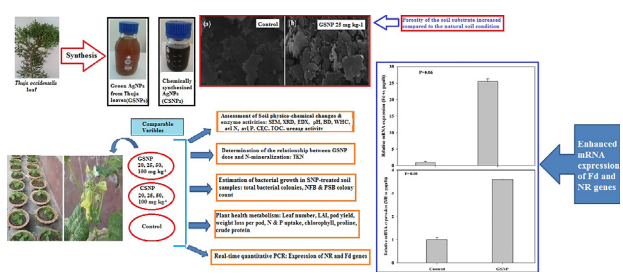
Pallabi Das[a], Shaswat Barua[b], Shuvasree Sarkar[c], Niranjan Karak[b], Pradip Bhattacharyya[d], Nadeem Raza[e], Ki-Hyun Kim[f,*], Satya Sundar Bhattacharya[a,*]

[a] Soil and Agro-Bioengineering Laboratory, Department of Environmental Science, Tezpur University, Assam, 784028, India
[b] Advanced Polymer and Nanomaterial Laboratory, Department of Chemical Sciences (Centre for Polymer Science and Technology), Tezpur University, Assam, 784028, India
[c] Department of Zoology, Visva-Bharati, Santiniketan, West Bengal, India
[d] Agricultural and Ecological Research Unit, Indian Statistical Institute, Giridih, Jharkhand 815301, India
[e] Govt. Emerson College affiliated with Bahauddin Zakaryia University, Multan, Pakistan
[f] Dept. of Civil & Environmental Engineering, Hanyang University, 222 Wangsimni-Ro, Seoul 04763, South Korea

HIGHLIGHTS

- The remarkable antimicrobial activity of silver nanoparticles (SNPs) is well known.
- Extensive industrial use of SNPs has led to their large-scale disposal as waste materials.
- The effects of SNPs on plant metabolism are assessed in terms of NR and Fd expression.
- We provide evidence of an overall beneficial impact of SNPs on soil properties.

GRAPHICAL ABSTRACT



ARTICLE INFO

Article history:
Received 28 March 2017
Received in revised form 5 December 2017
Accepted 7 December 2017
Available online 8 December 2017

Keywords:
Green silver nanoparticles
Soil quality
N availability
Plant health
Fd gene expression

ABSTRACT

Recently, concerns have been raised regarding the ultimate fate of silver nanoparticles (SNPs) after their release into the environment. In this study, the environmental feasibility of plant leaf (*Thuja occidentalis*) extract–mediated green SNPs (GSNPs) was assessed in terms of their effects on soil physicochemical properties and crop growth in comparison to conventionally synthesized silver nanoparticles (CSNPs). Upon application of GSNPs, soil pH shifted toward neutrality, and substantial increments were observed in water holding capacity (WHC), cation exchange capacity (CEC), and N/P availability. The mechanism behind the enhanced availability of N was verified through lab-scale experiments in which GSNP-treated soils efficiently resisted nitrate leaching, thereby sustaining N availability in root zone soil layers. However, retardation in nutrient availability and enzyme activity was apparent in soils treated with 100 mg kg^{-1} of either CSNPs or GSNPs. Remarkable improvements in leaf area index (LAI), leaf number, chlorophyll content, nitrate reductase (NR) activity, and *Phaseolus vulgaris* pod yield were observed after the application of low doses of GSNPs (25–50 mg kg^{-1}). The true benefit of GSNP application to soil was substantiated through experiments on plant uptake of nutrients, NR expression, and ferredoxin gene expression in *P. vulgaris* leaves.

1. Introduction

Nanotechnology is a leading area in modern science. The community is expecting great improvements in living quality with the

* Corresponding authors.
E-mail addresses: kkim61@hanyang.ac.kr (K.-H. Kim), satyasundarb@yahoo.co.in, satya72@tezu.ernet.in (S.S. Bhattacharya).
https://doi.org/10.1016/j.jhazmat.2017.12.020
0304-3894/

continuing growth of nanotechnology. Silver is among the most widely used materials in engineered nanoparticles [1]. The remarkable antimicrobial activity of silver nanoparticles (SNPs) has led to their use in various domains of biomedical research [2]. However, because of their nanoscale size, high reactivity, and unique dissolution properties, their associated environmental influenecs has not yet been fully elucidated.

Extensive industrial use of SNPs has led to their large-scale disposal as waste materials in the soil environment [3]. Their interactions with plant cells, uptake, and toxicological effects are well known [4]. In contrast, relatively little is known about the influence of SNPs on soil quality. A few nanoparticles have been reported to catalyze the formation of organic pollutants in aqueous suspensions and eventually to modify the redox potential of soil organic matter [3]. Recently, multi-walled carbon nanotubes were reported to be used to effectively remediate sediments contaminated with phenanthrene and cadmium [5]. However, SNPs are more mobile in soils enriched in mineral than in organic matter [6]. Moreover, SNPs are also reported to influence the composition of essential oils in some plants [7].

Most of the SNPs used in consumer products enter terrestrial ecosystems through land application of biosolids [1]. The soil system is the eventual sink for released nanoparticles. Hence, the interaction of SNPs with the soil environment can alter the chemistry of the SNPs and/or the soils (dispersibility, dissolution rate, size, agglomeration, surface area, surface chemistry, transformation, and charge), which in turn can influence the stability of the SNPs and their transport through soil systems [1,8]. In addition, plants can accumulate SNPs in their biomass, thereby restricting their transport [9]. Despite the significance of soils as the major pathway of SNP release, reports of practical field research on soil quality and plant health are limited. Rather, most studies have examined the fate and behavior of SNPs in aquatic systems [10,11]. Although the basic properties of SNPs have been characterized, such as their physical, mineralogical, and chemical compositions, relatively little is known about how they influence the physical compositions and mineral distributions of soils [12]. In addition, the responses or sensitivities of plant communities toward SNP exposure vary considerably among species and growing conditions [1,13].

Considering the dearth of basic information regarding the behavior of SNPs in soil–plant systems, we investigated the effects of *T. occidentalis* leaf extract–mediated green synthesis pathway of SNP (hereafter GSNP) on a soil–plant system by applying them at various concentrations. The effects of GSNPs were examined in various respects and compared to the effects of conventionally synthesized silver nanoparticles (hereafter CSNPs) [14]. In the SNP-inoculated soils, we grew *Phaseolus vulgaris* (French bean), a winter vegetable, over the short duration of 70 d. Changes in the physicochemical properties of the soils were assessed in addition to plant growth and yield. In addition, two lab-scale mechanistic experiments were conducted to understand the effects of GSNPs on N mineralization in soil. Here, we report hitherto unknown information regarding the effects of SNPs on soil nitrate retention. In fact, in this investigation, we provide evidence of an overall beneficial impact of SNPs on soil properties. Moreover, we precisely assessed the effects of SNPs on plant metabolism in terms of nitrate reductase (NR) and ferredoxin gene (Fd) expression.

2. Materials and methods

2.1. Preparation of CSNPs and GSNPs

$AgNO_3$ and polyethylene glycol (PEG, Merck, India) were used as received. *T. occidentalis* leaves were collected from a university garden (Tezpur University, Assam, India). *T. occidentalis*–mediated GSNPs were prepared following procedures we recently developed [14]. Stock solutions of both types of nanoparticles (CSNP and GSNP) were serially diluted to obtain the desired concentrations for experiments (20, 25, 50, and 100 mg kg^{-1}). Hereafter, nanoparticle solutions are abbreviated with the nanoparticle type and concentration; for example, $CSNP_{20}$ refers to a 20 mg kg^{-1} solution of CSNPs. Control experiments were conducted in soils not treated with SNPs.

2.2. Experimental setup

Soil samples were collected from typical alluvial soil of Tezpur, India (pH = 6.5 ± 0.6; bulk density (BD) = 1.26 ± 0.2 g cm^{-3}; water holding capacity (WHC) = 52.5 ± 2%; cation exchange capacity = 4.49 ± 0.22 mmol kg^{-1} soil; total organic carbon = 2.1 ± 0.6% ; available N = 314.6 ± 2.2 mg kg^{-1}; available P = 29.6 ± 1.1 mg kg^{-1}). Collected soils were air dried, sieved through a 80 mm mesh followed by a 2 mm mesh, and then used for the study. CSNP and GSNP solutions of various concentrations were mixed with soil samples in earthen pots of 0.45 m height and 0.25 m diameter (2 L volume); the amount of solution added to each sample was 10% by volume (200 mL).

Subsequently, seeds of *P. vulgaris* were sown in each vessel and cultivated following prescribed management practices [15]. We did not apply any chemical fertilizers to the experimental soil in order to avoid their interactions with SNPs. However, well decomposed cow dung manure of 5% by weight was applied uniformly in each pot to provide nutrition to the plants.

2.3. Assessment of physicochemical changes in soil

Samples of control and treated soils ($GSNP_{20}$, $GSNP_{25}$, $GSNP_{50}$, and $GSNP_{100}$) were collected at harvest maturity, namely after 60 days of cropping. The soil samples were then air-dried, ground in an agate mortar, and sieved (<200 mesh) for physicochemical analysis. The ground soil samples were packed in circular sample holders and subjected to X-ray diffraction analysis (XRD; Rigaku Miniflex) under intense Cu Kα radiation (λ = 1.54 Å) over the range of 10–70° 2θ. The surface morphology and elemental composition of each of the control and treated soil samples (GSNP 25, 50, and 100 mg kg^{-1}) were examined by scanning electron microscopy (SEM; JSM-6390 LV SEM, JEOL) paired with energy dispersive X-ray spectrometry (EDX). Moreover, the BD and WHC of each soil sample were analyzed [16].

The effects of each treatment on soil pH, total organic C (TOC), easily mineralizable nitrogen, available phosphorus, and CEC were analyzed according to well established methods [16]. Urease activity was analyzed in the soil according to the method of Tabatabai and Bremner [17].

2.4. Benefit percentage

Soil physicochemical characteristics (pH, nutreint availability (e.g., N and P), cation exchange capacity (CEC), total organic C (TOC), WHC, and BD) are strong indicators of soil quality and fertility [18]. Therefore, we derived a formula (Benefit Percentage) to estimate the overall changes in soil fertility in reference to one of our previous publications [19]. The benefit percentages of different SNP solutions with respect to controls for the soil quality variables of WHC, CEC, TOC, pH, and N/P availabilities were computed using the following equation.

$$\text{Benefit}\,\% = \frac{(\mathbf{Average\ at\ 60\ d}) - (\mathbf{Average\ at\ 0\ d})}{(\mathbf{Average\ 0\ d})} \times 100 \quad (1)$$