RESEARCH ARTICLE | MAY 09 2023

Green synthesis of sulfur nanoparticles using garlic (*Allium sativum*): Fungicidal activity and plant growth effects on chili plants (*Capsicum annum* L.)

Khairan Khairan ✉; Firna Aryati; Putri Dika Suhayla; ... et. al

AIP Conference Proceedings 2480, 040015 (2023)
https://doi.org/10.1063/5.0121749

Green Synthesis of Sulfur Nanoparticles Using Garlic (*Allium sativum*): Fungicidal Activity and Plant Growth Effects on Chili Plants (*Capsicum annum* L.)

Khairan Khairan[1,2,3,4,a)], Firna Aryati[4,b)], Putri Dika Suhayla[4,c)], Rina Sriwati[3,5,d)], Khalijah Awang[6,e)]

[1)]*Department of Pharmacy, Universitas Syiah Kuala, Banda Aceh, 23111, Indonesia*
[2)]*Pusat Riset Obat Herbal, Universitas Syiah Kuala, Banda Aceh, 23111, Indonesia*
[3)]*Atsiri Research Centre, Universitas Syiah Kuala, Banda Aceh, 23111, Indonesia*
[4)]*Department of Chemistry, Universitas Syiah Kuala, Banda Aceh, 23111, Indonesia*
[5)]*Department of Plant Diseases, Universitas Syiah Kuala, Banda Aceh, 23111, Indonesia*
[6)]*Department of Chemistry, Faculty of Science, University of Malaya, Kuala Lumpur, 50603, Malaysia.*

[a)]*Corresponding author: khairankhairan@unsyiah.ac.id*
[b)]*virnavirna6@gmail.com*
[c)]*Putridkshylaa@gmail.com*
[d)]*rin_aceh@yahoo.com*
[e)]*khalijah@um.edu.my*

Abstract. Green synthesis of sulfur nanoparticles (gSNP) using aqueous garlic extract (*Allium sativum*) have been effectively conducted from sodium thiosulfate pentahydrate. The gSNP were characterized using FT-IR, XRD, SEM-EDX spectroscopy. The results show that gSNPs show high purity and spherical existence with an average particle size of about 54.73 nm. The fungicidal activity of gSNP revealed that at a concentration of 800 ppm they exhibited the highest activity against *Colletroticum capsici* with a 14 mm diameter of the inhibition zone after 48 hours of incubation. The obtained results reveal that applying gSNP by foliar spray at a concentration of 400 ppm produced a healthier chili plant (*Capsicum annum* L.) in terms of plant (height, amount of leaves, length of root and weight of root, compared with a control (plant treated with the growth hormone; indole acetic acid, or IAA).

INTRODUCTION

Recently, the study of nanoparticles has grown rapidly due to their wide application in pharmaceuticals, biomedical technology, electronics, optics, and agriculture. Nanoparticles are materials that have one dimension with a diameter smaller than 100 nanometers, including nanodots (quantum dot), nanowires, and nanotubes [1].

Sulfur is a non-metallic atom and possesses antibacterial, anticancer [2], and insecticidal properties [3]. Sulfur is an essential macronutrient for plant growth, development, and response to environmental changes [4]. Sulfur is considered a safer element compared to other metals because of its ability to form organosulfur compounds in plant tissues [5]. The activity of sulfur atoms in cells is enzymatically triggered by an electron transfer to a sulfur atom where the sulfur atom is reduced to form reactive sulfur species (RSS). The RSS generated could cause toxicity in cells and inhibit cell growth [6].

In the past decade, a lot of research has been done on sulfur nanoparticles (SNPs). Synthesis by using a plant extract or green synthesis is one of the prime methods used to synthesize nanoparticles. This method was selected because it is the easiest, simplest, most convenient, it requires the least amount of reaction time, and it is eco-friendly [6]. Paralikar and Rai mentioned that green synthesis using plant extracts has more potential as compared to microorganisms because plant extracts can produce metal ions [7]. Plant extracts have the capability to reduce or

The 4th International Conference on Science and Technology (ICST) 2021
AIP Conf. Proc. 2480, 040015-1–040015-9; https://doi.org/10.1063/5.0121749
Published by AIP Publishing. 978-0-7354-4463-8/$30.00

accumulate metal ions to form nanoparticles on the surface of the cells, organs, and tissues. The metal accumulated by a plant extract can be restored after harvesting through sintering and smelting methods [8]. The type of metabolites and the kind of plant are very important factors in the synthesis process of nanoparticles as they greatly affect the morphology of the nanoparticles. The types of metabolites and the kind of plants that are often used for nanoparticle synthesis are *Azadirachta indica* [9], *Aloe vera* [10], *Punica granatum* [11], *Parkia speciose* [12], *Talaromyces purpurogenus* [13], and *Allium sativum* [14].

Shankar and Makarov reported that various metabolites including proteins, phenolic acid, alkaloids, sugars, polyphenols, and terpenoids play an important role in the bioaccumulation and bio reduction of metal ions yielding nanoparticles [6]. For example, amino acids are able to bind to metal ion via C- and N-terminus. Other side groups such as cysteine (thiol groups) are also responsible for reduction of metal ion to form nanoparticles [15].

Garlic has been known to contain high levels of organosulfur compounds which can potentially be used as herbal medicine for the treatment of various diseases and physiological disorders [16]. It has been demonstrated by previous studies to exhibit anticancer, antivirus, insecticidal, and antimicrobial properties [17]. Garlic contains high levels of organosulfur compound including allicin, diallylsulphide (DAS), diallyldisulphides (DADS), and diallyltrisulphides (DATS) [14, 17, 18].

Chili (*Capsicum annum* L.) is an important vegetable crop all over the world. The Food and Agriculture Organization Corporate Statistical Database (FAOSTAT) reported that total production of chili pepper increased by 25% from 2006 to 2016. Researchers have reported that chilies can be used as antioxidant and anti-inflammation agents due to a large vitamin C and carotene content, and also some minerals such as potassium, magnesium, and iron [19].

Colletotrichum is one of the most important plant pathogenic fungi causing anthracnose disease. This disease economically reduced the productivity of the chili plant (*Capsicum spp*.) up to 50% in worldwide in 2008 [20]. For decades, farmers have used synthetic fungicides to reduce this disease. However, the continuous use of pesticides can cause an emergence of pathogenic resistance, environmental harm, and a danger to consumers.

Salem showed that green synthesis using *Punica granatum* was able to accumulate sulfur nanoparticles that measured up to 20 nm. Salem also showed that sulfur nanoparticles with a concentration of 200 ppm produced the best growth effects on a tomato plant compared to a tomato plant treated with control and growth hormone IAA [11, 14]. Our previous research revealed that *Allium sativum* was able to accumulate sulfur nanoparticles up to 50.76 nm. In this study, we synthesized sulfur nanoparticles using *Allium sativum* and evaluated its activity against *Colletroticum capsici*. We also explored the effect of the sulfur nanoparticles on the growth parameters of chili plants (*Capsicum annum* L.).

MATERIALS AND METHODS

Materials

Sodium thiosulfate pentahydrate ($Na_2SO_3.5H_2O$) was purchased from Merck (Germany), nystatin and dithane M-45 were used as positive controls for antifungal assay, distilled water was prepared using Water Distillation GFL2004, garlic (*Allium sativum*) was obtained from the Lambaro traditional market, Aceh Besar, Indonesia. The seeds of the chili plant variety Profit F1 were obtained from Ulee Kareng traditional market, Aceh Besar, Indonesia.

Preparation of Aqueous Garlic (*Allium sativum*) Extract

The clove of *Allium sativum* (100 gram) was chopped and air dried for ± 4 weeks. The dried garlic was then crushed into powder. 20 g of the garlic powder was dissolved in 500 mL of aquadest, boiled for 20 minutes, and filtered. The filtrate was centrifuged for 5 minutes at 1200 rpm to remove biomaterials. The filtrate was then used as media to synthesis sulfur nanoparticles.

Green Synthesis of Sulfur Nanoparticles (gSNP).

Green synthesis of sulfur nanoparticles (gSNP) was prepared according to Salem [11]. Briefly, 24.8 g of sodium thiosulfate pentahydrate ($Na_2S_2O_3.5H_2O$) were dissolved into 500 mL of aqueous garlic extract and stirred for 10 minutes at 400 rpm at room temperature. Then, 500 mL of distilled water was added. After that, 10% HCl solution was added drop by drop using dropping funnel to precipitate the sulfur particles. The precipitated sulfur particles then centrifuged at 1000 rpm for ten minutes at ambient temperature. The supernatant was removed, and the sulfur particles were subsequently washed successively using aquadest and absolute ethanol using centrifugation to remove biological materials from the garlic. Sulfur particles that were free of biological material were then vacuum-dried at 80ºC for 6 hours. The gSNP produced after drying were powdery, soft and yellow in color. The gSNP produced were then freeze-dried, and kept at room temperature for further analysis. Under acidic conditions, the garlic extract together with sodium thiosulfate will undergo a disproportionation reaction into sulfur and sulfonic acid particles, as shown below:

$$Na_2S_2O_3.5H_2O + 2HCl \rightarrow 2NaCl + S(s) + SO_2 + 6H_2O$$

Characterization of Sulfur Nanoparticles (gSNP)

The gSNP were characterized by using a X-ray diffractometer (XRD-6000 Shimadzu, Japan), with Cu Kα as the source of radiation, and Ni as the filter, with energy of 30 kV/30 mA. Every data point was taken at an angle of $3^o \leq 2\theta \leq 60^o$. The FT-IR analysis spectrophotometer was performed at wave numbers 4000-650 cm-1 using Agilent Technologies Cary 630 model. SEM-EDX analysis was performed using the Quanta FEI 450 SEM machine.

Fungicidal Activity

The fungicidal activity of gSNP was performed using the Kirby-Bauer method [21]. The sterile potato's dextrose agar (PDA) media was poured into petri dishes and allowed to solidify, and the strain of *Colletotrichum capsici* was spread out on the surface of the solidified PDA using a cotton bud. The paper disc (approximately 6 mm in diameter) was laid out on the surface of the agar medium. On each of the discs, 12 µl of gSNP was loaded with the concentrations of 600 and 800 ppm. In this assay, the aqueous garlic extract, nystatin and dithane-M45 were used as positive controls. The dishes were subsequently incubated for 48 hours at 37°C. Then, the inhibition effect of the tested samples were determined. The fungicidal activity was performed in triplicates.

Effect of Sulfur Nanoparticles (gSNP) on Growth of Chili Plant

All studies on chili (*Capsicum annum* L.) were performed in greenhouse compartments at the Department of Plant Diseases, Universitas Syiah Kuala, Darussalam Banda Aceh, Indonesia. The seeds of *Capsicum annum* L., here after abbreviated to *C. annum* L.) were grown on seedling tray. After 4 weeks, the young plants were transferred to the polybags filled with a commercial potting soil. The response of *C. annum* L. to the gSNP through the experiments was performed in April and May of 2019. The gSNP was applied to the chili plant at 200 and 400 ppm using foliar spray application. In this assay, the chili plant was also treated with the growth hormone IAA as a positive control. In each application, the aerial part of the chili plant were wrapped with a plastic sheet to prevent contamination with gSNP. After two months of applications, the growth parameters of *C. annum* L. such as plant height, number of leaves, and length and weight of root were measured. Results were expressed as a mean of the number of experiments indicated. The p-values were determined using Prism 5 software. Significances: ns $p \geq 0.05$, * $p < 0.05$,** $p < 0.01$ and *** $p < 0.001$.

RESULTS AND DISCUSSION

FT-IR Analysis

Figure 1 shows the FT-IR spectrums of aqueous garlic extract and green synthesized sulfur nanoparticle (gSNP). The spectrum was recorded in the range of 4000-650 cm^{-1}. The FT-IR spectrum was conducted to identify garlic's capability as a bioreactor in the synthesis of sulfur nanoparticles. The FT-IR spectrum of aqueous garlic extract showed strong absorption of hydroxyl groups (-OH) and amino group (-NH) at 3410 cm^{-1}, Figure 1A. This absorption is most likely due to the presence of carbohydrates and amino acids in the garlic. The peak at 1670 cm^{-1} is typical of carboxyl group (-COOH) stretches [22], while the peak at 1026 cm^{-1} indicates the presence of thiocarbonyl group (S=O) [23], which are common in many natural sulfur compounds such as allicin.

Figure 1B shows the absorption at 3410 cm^{-1} which could be obtained from biomolecules of garlic that used as media synthesis process. Aqueous garlic extract media plays a role in the process of stabilizing and dispersing sulfur nanoparticles during the synthesis process. The peak at 1670 cm^{-1} was typical of carboxyl groups (-COOH) stretches [25], indicated the presence of thiocarbonyl groups (S=O) [26], which is common in many natural sulfur compounds such as allicin. Figure 1B, it also can be observed that the carbonyl peak at 1670 cm^{-1} has shifted to 1635 cm^{-1}. This shift may suggest that the garlic extract was able to bind sulfur nanoparticles through biomolecules (protein and carbohydrate) and secondary metabolites from garlic. These results indicated that the aqueous garlic extract is capable of binding to sulfur nanoparticles.

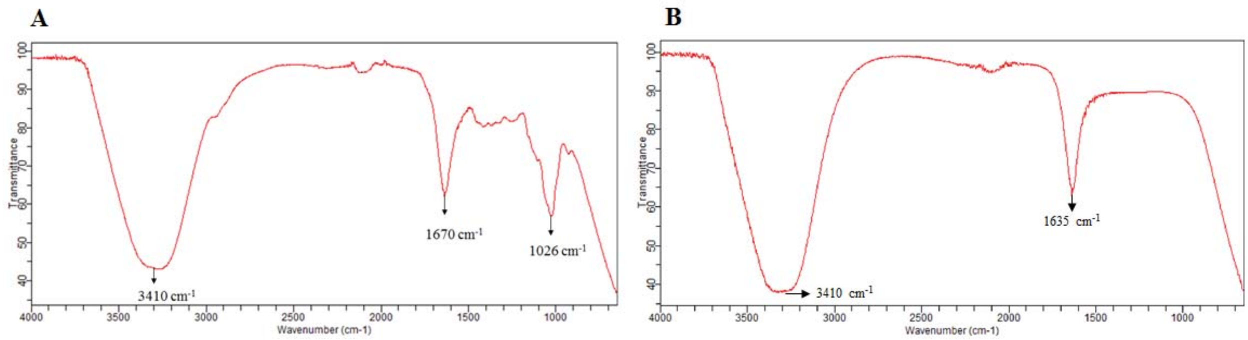

FIGURE 1. **A** FT-IR spectrums of aqueous garlic (*Allium sativum*) extract; **B**. FT-IR spectrums of green synthesized sulfur nanoparticles (gSNP)

XRD Analysis

Analysis of the X-ray diffraction (XRD) pattern of the gSNP using *Allium sativum*, Figure 2 and Table 1 show that gSNP were well crystallized. The diffraction peaks of gSNP correspond well with the standard sulfur diffraction pattern according to JCPDS.

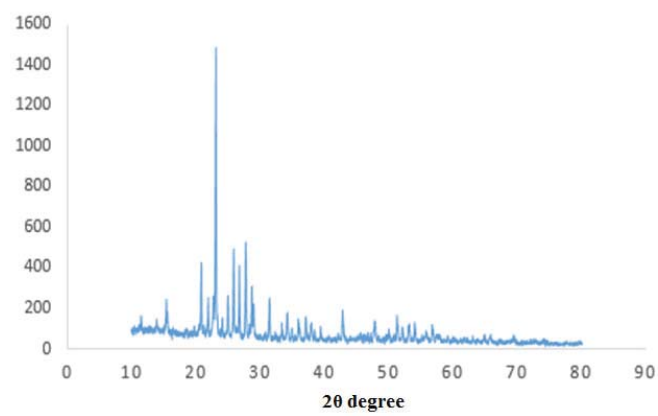

FIGURE 2. XRD analysis of synthesized sulfur nanoparticles (gSNP).

The size of the gSNP using aqueous garlic extract was 54.73 nm, which correspond well with the previous obtained results of 55.61 nm [14].

TABLE 1. XRD analysis of synthesized sulfur nanoparticles (gSNP).

2θ(º)		**FWHM(º)**	**Intensity**	**phase**
gSNP	**JCPDS[a]**			
23.0877	20.9630	0.14420	939	S

SEM-EDX Analysis

The SEM analysis has shown that the surface morphology structure of gSNP has a spherical particle form, which is in agreements with the reports by [11, 14] (Figure 3A). The EDX spectrum shows that the sulfur atoms were detected at 2,307 keV (Figure 3B).

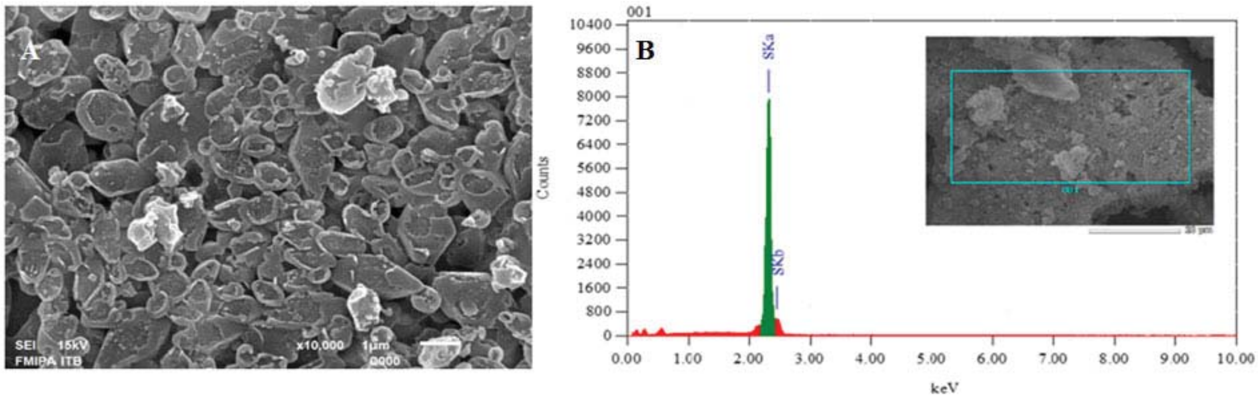

FIGURE 3. A. SEM images of gSNP 10000 magnification; B. EDX spectrum of synthesized sulfur nanoparticles (gSNP).

Fungicidal Activity of gSNP

The fungicidal activity of gSNP using aqueous garlic extract was evaluated to determine the activity of sulfur nanoparticles against pathogenic fungi of *Colletotrichum capsici*. The fungicidal activity of gSNP was determined using the disc diffusion method after 24 and 48 hours of incubation. The antifungal activity of gSNP is presented in Figure 4, while the inhibition zone of *Colletotrichum capsici* is listed in Table 2.

TABLE 2. Diameters inhibition zone of gSNP on *Colletotrichum capsici*

Treatment	**Inhibition Zone (mm)**	
	24 h	**48 h**
Control (negative control)	0.0	0.0
AGE (positive control)	18.0	18.7
Dithiane-M45 (positive control)	23.0	24.0
Nystatin (positive control)	22.7	23.7
gSNP 600 ppm	10.0	13.0
gSNP 800 ppm	13.3	14.0

The antifungal activity of gSNP increases with concentration and time of exposure. The zone of inhibition of gSNP increases from 10 mm to 13 mm after exposure for 48 hours and it increase to 13.3 mm when the concentration was increased to 800 ppm.

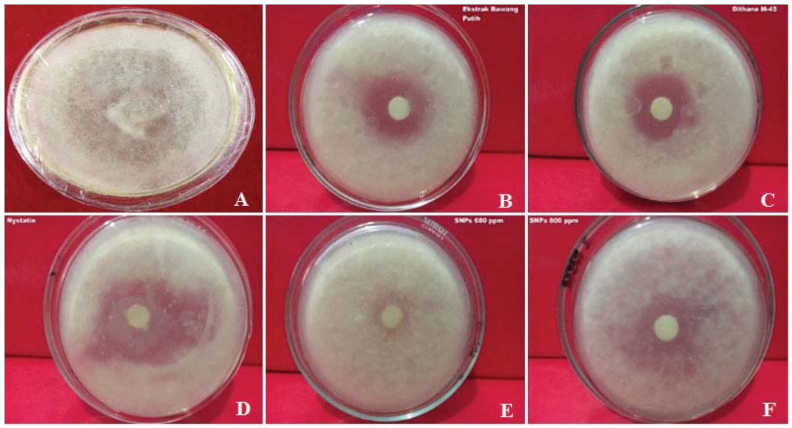

FIGURE 4. The fungicidal activity of gSNP on *Colletotrichum capsici*. A: control; B: aqueous garlic extract; C: dithiane-M45 (fungicide as positive control); D. nystatin (antifungal medication as positive control); E. gSNP 600 ppm; F. gSNP 800 ppm.

Effect of Sulfur Nanoparticles (gSNP) on Growth of Chili Plant

Chili (*C. annum* L.) is an herbaceous plant and belonging to the Solanaceae family. Chili is commonly cultivated in the tropical, and sub-tropical regions, including Indonesia [24]. Sulfur is an essential mineral for plant growth and function. In plants, sulfur is very important in the formation of plant proteins. Deficiency of sulfur can cause serious plant problems and loss of vitality [25]. The quality and yield of fresh *C. annum* L. are influenced by vegetative organs (leaf area, leaf chlorophyll, shoot dry weight and root dry weight) [26] and reproductive properties (flowers, seeds, and fruits) [27]. In this preliminary study, we investigated the effects of gSNP on the growth parameters of chili plants. The results show that the foliar application of gSNP on chili plants improved all parameters positively such as height of plant, number of leaves, length of root, dry-weight of root and gross-weight of root. The plant applied with 400 ppm gSNP showed the highest value for height (19.9 cm), number of leaves (36.1), length of root (31.7 cm), weight of root wet and dry (5 and 0.6 g respectively). Samples treated with gSNP (400 and 200 ppm) showed better growth parameters as compared to untreated plants and positive control (IAA) (Figure 5).

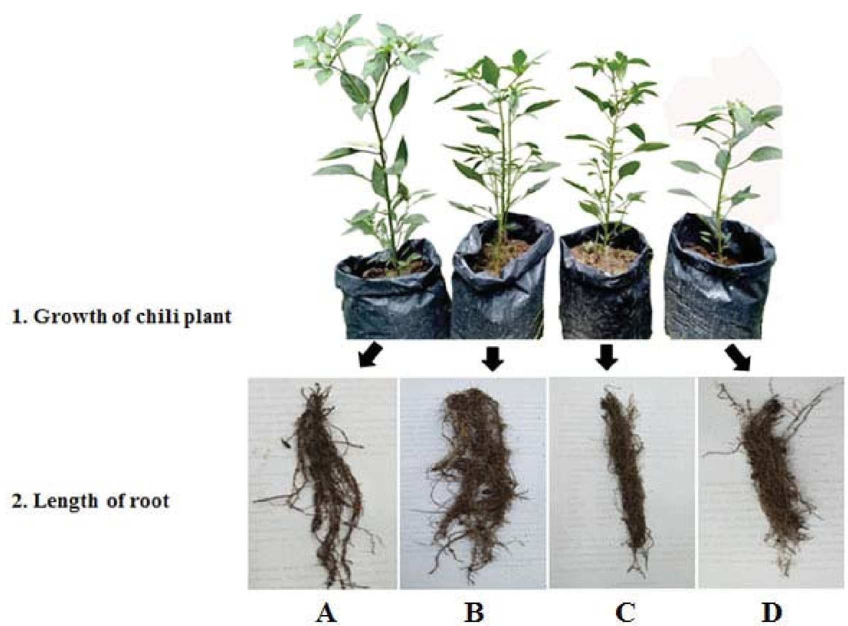

FIGURE 5. Response of synthesized sulfur nanoparticles (SNPs) on length of root of chili plant by foliar spray applications. A 400 ppm of SNPs; B. 200 ppm of SNPs; C. Growth hormone (IAA) as positive control; and D. Untreated as negative control.

Healthier and greener leaves of chili plants were observed in the plants treated with 200 and 400 gSNP, especially with 400 ppm (Figure 5). These results were probably due to the interaction of sulfur nanoparticles by leaves with organic compounds of chili tissues forming organosulfur compounds, which help to build the chlorophyll content of the chili leaves. In addition, under atmosphere these organosulfur compounds decompose and release hydrogen sulfide H_2S, which acts as antifungal agent [23]. On the other hand, sulfur nanoparticles suppress crop disease, and subsequently enhance the *Capsicum annum* growth. This enhancement suggests that sulfur nanoparticles have the potential to reduce pathogenic organisms in the soil and increase nutritional value by producing organosulfur compounds in plant [5] and also enhance sulfur uptake through sulfate transporters from the soil [4]. Therefore, this preliminary study implies that green synthesized sulfur nanoparticles (gSNP) have strong impact on the growth of the *Capsicum annum* plant.

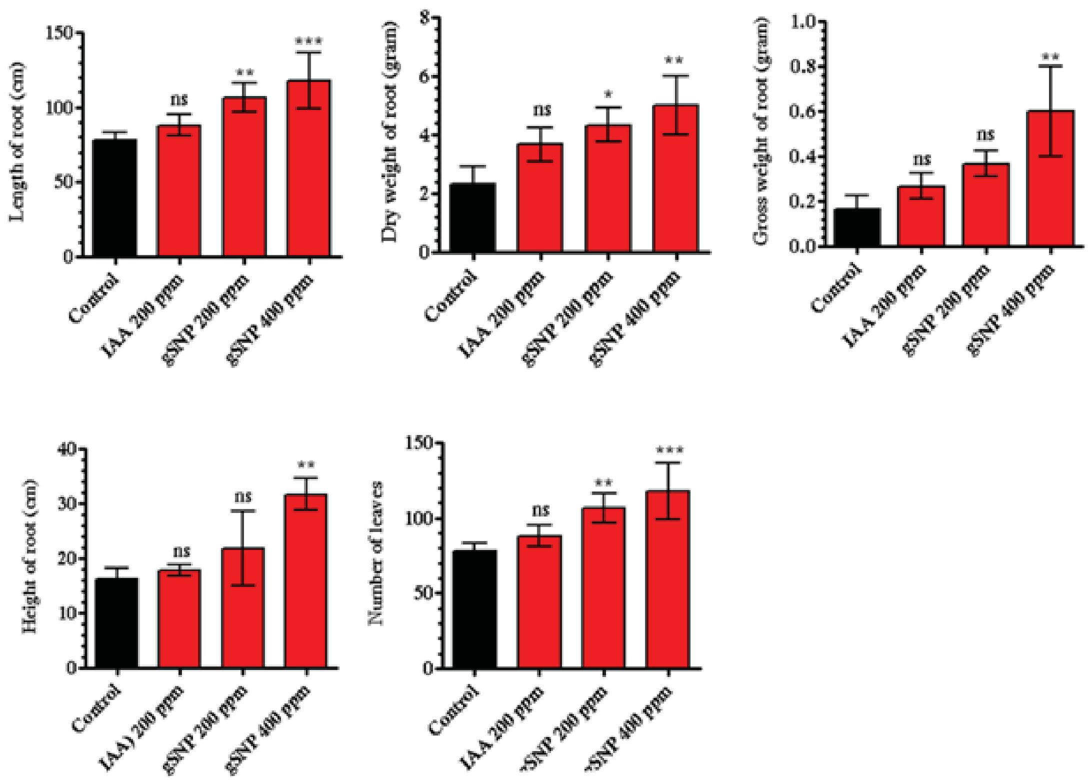

FIGURE 6. Response of gSNP on growth of chili plant (*Capsicum annum L)* by foliar spray applications. Graph A. Height of plant; . Graph B. Number of leaves; . Graph C. Length of root; . Graph D. Dry-weight of root and Graph E. Gross-weight of root. Significances are expressed to the control. Data presented as viability % ± SD. Significances: ns $p \geq 0.05$, * $p < 0.05$, ** $p < 0.01$ and *** $p < 0.001$.

CONCLUSIONS

Green synthesis of gSNP exhibited high purity of sulfur nanoparticles, uniform shape, and are highly crystalline with the average crystal size of about 54.73 nm. There was also better activity against pathogenic *Colletotrichum capsici* upon treatment with gSNP. Growth parameters also showed better results in chili plant treated with gSNP as compared to the growth hormone IAA. These observations may suggest that gSNP could play an important role in the enhancement of sulfur uptake which is beneficial for plant growth.

ACKNOWLEDGEMENTS

The authors thank Universitas Syiah Kuala for the research facilities to conduct this research.

REFERENCES

1. B. Park, Current and Future Applications of Nanotechnology, 1-18, (2016).
2. S. Shankar, R. Pangeni, J. Woo, and J. W. Rhim, Materials Science & Engineering C **92**, 508–17 (2017).
3. K. Khairan, R. Idroes, M. Bahi, K. H. Schaefer, T. Schneider, and C. Jacob. Jurnal Fakultas Kedokteran Hewan **9** (1), 1–5 (2015).
4. T. Gigolashvili, and S. Kopriva, "Transporters in Plant Sulfur Metabolism," Front. Plant Sci. 1–16 (2014).
5. M. Suleiman, M. Al-Masri, A. Ali, D. Aref D, A. Hussein, I. Saadeddin, I. Warad, "Synthesis of nano-sized sulfur nanoparticles and their antibacterialactivities," J. Mater. Environ. Sci. **6**, 513–518 (2015).
6. V. Makarov, V. A J Love, O. V. Sinitsyna, S. S. Makarova, and I. V. Yaminsky, "Green Nanotechnologies : Synthesis of Metal Nanoparticles Using Plants," Acta Natura 6 (**20**), 35–44 (2014).
7. P. Palikar, and R. Mahendra, "Bio-Inspired Synthesis of Sulphur Nanoparticles Using Leaf Extract of Four Medicinal Plants with Special Reference to Their Antibacterial Activity," IET Nanobiotechnology, **12**, 1, 25-31 (2018).
8. J. G. Parsons, E. Gomez, H. E. Troiani, P. Santiago, and M. Jose Yacaman, "Formation and Growth of Au Nanoparticles inside Live Alfalafa Plants," Nano Letters **2**(4), 397-401 (2002).
9. S. S. Shankar, R. Akhilesh, A. Ahmad, and M. Sastry, "Rapid Synthesis of Au , Ag , and Bimetallic Au Core-Ag Shell Nanoparticles Using Neem (*Azadirachta Indica*) Leaf Broth," J Colloid Interface Sci. **275**(2), 496-502 (2004).
10. S. Maensiri, P. Laokul, J. Klinkaewnarong, S. Phokha, and S. Seraphin, "Indium Oxide (In_2O_3) Nanoparticles Using Aloe Vera Plant Extract : Synthesis and Optical Properties," Materials Science and Engineering **10** (3), 161–65 (2008).
11. M. Salem, S. A. Luma, A. M. Awwad, " Green synthesis of sulfur nanoparticles using *Punica granatum* peelsand the effects on the growth of tomato by foliar spray applications, "Environmental Nanotechnology, Monitoring & Management **6**, 83–87 (2016).
12. V. Ravichandran, S. Vasanthi, S. Shalini, S. Adnan, A. Shah, M. Tripathy, and N. Paliwal, "Results in Physics Photocatalytic Activity of *Parkia speciosa* Leaves Extract Mediated Silver Nanoparticles," Results in Physics **15**, (2019).
13. S. Bhatnagar, T. Kobori, D. Ganesh, and K. Ogawa, "Biosynthesis of Silver Nanoparticles Mediated by Extracellular Pigment from *Talaromyces purpurogenus* and Their Biomedical Applications," Nanomaterials (Basel) **9**(7), 1042 (2019).
14. K. Khairan, Z. Riaz, and Z. Jalil, "Green Synthesis Of Sulphur Nanoparticles Using Aqueous Garlic Extract (*Allium sativum*), "Rasayan Journal Chemistry, **12** (1): 50–57 (2019).
15. Y. Z. Hamada, "Metal Ions In Biological Systems," Electronic Journal of Biology, S2, 1-1 (2016).
16. S. Papu, S. Jaivir, S. Sweta, and B. R. Singh, "Medicinal Values of Garlic (*Allium sativum* L.) in Human Life : An Overview," Greener Journal of Agricultural Sciences (2014).
17. T. Schneider, A. Baldauf, L. A. Ba, V. Jamier, K. Khairan, M. B. Sarakbi, and N. Reum, "Selective Antimicrobial Activity Associated with Sulfur Nanoparticles," J Biomed Nanotechnol **7**(3), 395-405 (2011).
18. C. Jacob, G. I. Giles, N. M. Giles, and H. Sies, "Sulfur and Selenium : The Role of Oxidation State in Protein Structure and Function, "Angewandte, 4742–58 (2003).
19. A. Rigon, B. Leonardi, D. Miron, E. Schapoval, J. Rodrigues, De Oliveira, and G. Gosmann, "Antioxidant and Anti-Inflammatory Properties of *Capsicum baccatum* : From Traditional Use to Scientific Approach, " Journal of Ethnopharmacology **139** (1), 228–33 (2012).
20. P. P. Than, H. Prihastuti, and S. Phoulivong, "Chilli Anthracnose Disease Caused by Colletotrichum Species, " **9** (10), 764–78, J Zhejiang Univ Sci B, **9**(10), 764-78 (2008).
21. S. Alam, "Synthesis, Antibacterial and Antifungal Activity of Some Derivatives 2-phenyl-chromen-4-one," J. Chem. Sci. **116**, 325-331 (2004).

22. R. G. Chaudhuri, and S. J. Paria, "Synthesis of Sulfur Nanoparticles in Aqueoussurfactant Solutions," J. Colloid Interface Sci. **343**, 439–446 (2010).
23. N. M. Salem, L. S. Albanna, A. O. Abdeen, Q. I. Ibrahim QI, A. M. Awwad, "Sulfurnanoparticles Improves Root and Shoot Growth of Tomato,"J. Agri. Sci. **8**, 179–185 (2016).
24. T. M. Olatunji, and A. J. Afolayan, "The Suitability of Chili Pepper (*Capsicum Annuum* L .) for Alleviating Human Micronutrient Dietary Deficiencies : A Review," Food Sci Nutr. **6**(8), 2239-51 (2018).
25. H. Liu, Y. Zhang, S. Zheng, Z. Weng, J. Ma, Y. Li, X. Xie, and W. Zheng, "Biochemical and Biophysical Research Communications Detention of Copper by Sulfur Nanoparticles Inhibits the Proliferation of A375 Malignant Melanoma and MCF-7 Breast Cancer Cells, "Biochemical and Biophysical Research Communications **477** (4), 1031–37 (2016).
26. A. B. Oliveira, S. De Maria, De Mendonça, and M. S. Renata, "Anatomy of Vegetative Organs of *Scutellaria agrestis*, a Medicinal Plant Cultivated by Riverine Populations of the Brazilian Amazon, " Revista Brasileira de Farmacognosia-Brazilian Journal of Pharmacognosy **23** (3), 386–97 (2013).
27. L. Moravcová and J. Pergl, "Getting the Right Traits : Reproductive and Dispersal Characteristics Predict the Invasiveness of Herbaceous Plant Species," Plos One **23**, 1–16 (2015).