*Egypt. J. Chem.* **Vol. 65,** No. SI:13B pp. 1119 - 1128 (2022)

**Egyptian Journal of Chemistry**

**http://ejchem.journals.ekb.eg/**

# Impact of Silver Nanoparticles Mixture with NAA and IBA on Rooting Potential of *Psidium guajava* L. Stem Cuttings

**Abdou Abdallatif[1], Ibrahim Hmmam[1] and Mohamed A. Ali[2]***

[1]*Pomology Department, Faculty of Agriculture, Cairo University, Giza 12613, Egypt*

[2]*Biochemistry Department, Faculty of Agriculture, Cairo University, Giza 12613, Egypt*

**Abstract**

Guava (*Psidium guajava* L.) has been considered a hard-to-root species, where the percentage of rooting and survival of the stem cuttings is low. To enhance the rooting potential of guava stem cuttings, the current study was conducted to investigate the influence of NAA and IBA alone or combined with silver nanoparticles on the rooting of guava semi-hardwood cuttings. Silver nanoparticles were synthesized using the chemical reduction method. UV/VIS spectra and TEM micrographs confirmed the synthesis of silver nanoparticles (AgNPs) with an average particle size ranging d from 14.8 to 32.4 nm, and zeta potential confirmed the stability of AgNPs due to the high negative charge of zeta potential. Semi-hardwood cuttings of guava were subjected to six treatments, including the dipping in separate aqueous solutions of IBA, NAA, IBA-AgNPs, NAA-AgNPs, dipping in IBA followed by dipping in AgNPs solution (IBA+AgNPs), and dipping in NAA followed by dipping in AgNPs solution (NAA+AgNPs). Results indicated that the hormone type significantly affected all rooting parameters; higher measurements of rooting percentage, root number, root length, and root weight were recorded with IBA treatment, whereas NAA recorded significantly lower values. Auxin-AgNPs (IBA-AgNPs and NAA-AgNPs) and the double-dipping treatments improved rooting efficiency compared with the auxin treatments alone. Cutting of NAA-AgNPs treatment developed a lower number of long roots, while IBA-AgNPs recorded higher values of all root parameters. The present results demonstrated the stimulatory effect of auxin and AgNPs treatments on the rooting of guava stem cuttings.

Keywords: *Psidium guajava*, rooting, propagation, stem cuttings, IBA, NAA, silver nanoparticles.

## 1. Introduction

Guava (*Psidium guajava* L., Myrtaceae), the "poor man's fruit" or "apple of the tropics", is a commercially significant fruit crop in tropical and subtropical regions **[1]**. Guava is popular due to its comparatively low price, year-round availability, good taste, and nutritional value as a potential source of carbohydrates, antioxidants, vitamins, polyphenols, and minerals **[2,3]**. Guava cultivation has great economic significance in many countries around the world; the major guava-producing countries are India, Pakistan, Brazil, Egypt, Mexico, and Indonesia **[4]**. Guava is commonly propagated by seeds; subsequently, the produced seedlings do not maintain the genetic purity of the propagated mother plants. Furthermore, seedling trees exhibit vigorous growth and a long juvenile period **[5,6]**. Several methods of clonal propagation of guava have been proposed, such as cutting, layering, budding, grafting, and micropropagation **[7-10]**. In this regard, clonal propagation by grafting did not provide satisfactory results for guava propagation, while the number of plants obtained by air-layering is very low **[11,12]**. Propagation by stem cutting is a simple, rapid, and cost-effective propagation method that ensures the production of true-to-type plants **[13]**. However, all the above-mentioned techniques are still not commercially applicable due to the low rooting potential of guava cuttings. Several methods have been used to improve the rooting of guava cuttings **[7, 13-15]**. Auxin plays an important role in adventitious root formation **[16]** by increasing root primordium initiation **[17]** and promoting starch hydrolysis and sugar mobilization to the cutting base **[18]**. Auxin treatments are frequently used to improve the rooting potential of different fruit tree species **[19,20]**. The effects of IBA and NAA on the rooting of guava cuttings were previously reported **[21,22]**. One of the major challenges in the commercial application of auxin treatment is its sensitivity to

*Corresponding author e-mail: mohamed_soliman@cu.edu.eg (M.A. Ali), ORCID: https://orcid.org/0000-0002-1717-8265

Receive Date: 13 August 2022, Revise Date: 24 August 2022, Accept Date: 25 August 2022

DOI: 10.21608/EJCHEM.2022.155986.6751

environmental conditions; degradation by light and high-temperature results in the loss of biological activity **[23,24]**. Recently, the applications of nanoparticles in agriculture are receiving much attention because of their unique physicochemical properties, *i.e.,* high surface area, higher uptake, and translocation potential in plant tissues **[25-27]**. Nanoparticles can serve as delivery agents of fertilizers, pesticides, and plant growth regulators that target specific organelles in plants **[28,29]**. The use of silver nanoparticles (AgNPs) has been reported for various agricultural and industrial applications **[30,31]**. AgNPs promote root growth and increase root elongation in different plant species **[32-37]**. Moreover, AgNPs exhibited high antimicrobial activity against a broad spectrum of microorganisms **[38]**. AgNPs showed excellent antibacterial, antifungal and antiviral efficacy and had an inhibitory impact against nematode, bacterial, and fungal plant pathogens **[31,39,40]**. Thus, the aim of the current study was to evaluate the effect of synthesized silver nanoparticles combined with IBA and NAA on the rooting of guava semi-hard cuttings.

## 2. Materials and Methods

### *2.1. Chemicals*

Indole-3-butyric acid (IBA) was obtained from Sigma-Aldrich (Basel, *Switzerland)*. 1-naphthalene acetic acid (NAA) was obtained from Caisson Laboratory, Inc. (Smithfield, Utah, USA). Silver nitrate ($AgNO_3$) was obtained from Organik Kimya (Istanbul, Turkey). Sodium borohydride was obtained from CDH-Central Drug House (New Delhi, India) and polyethylene glycol 400 (PEG) was obtained from Alpha Chemika (Maharashtra, India). All solutions were prepared with deionized water unless otherwise stated.

### *2.2. Preparation of silver nanoparticle*

Silver nanoparticles were synthesized by the chemical reduction method. 1 mM silver nitrate ($AgNO_3$) was prepared by dissolving 0.034 g of $AgNO_3$ in 200 mL of deionized water in an ice bath. The 2 mM sodium borohydride (the reducing agent) was prepared by adding 0.0456 g of $NaBH_4$ to 600 mL of deionized water. The prepared solutions were kept in the freezer for 30 minutes. Polyethylene glycol (PEG 400) was used as stabilizing agent; 0.1% polyethylene glycol was prepared by dissolving 0.2 g of PEG in 200 mL of deionized water. Sodium borohydride solution was placed on a magnetic stirrer on ice bath, and then PEG solution was added. Silver nitrate solution was added to the previous mixture drop by drop using a burette under vigorous stirring until the transparent solution changed to a yellow color. The color change indicates the formation of silver nanoparticles (AgNPs) stabilized by PEG **[41]**.

### *2.3. Preparation of IBA and NAA solution*

The Indole-3-butyricacid (IBA) and naphthalene acetic acid (NAA) solutions were prepared at a concentration of 10 mM by dissolving 1.015 g IBA in 500 mL of 50% aqueous ethanol (v/v) and 0.930 g NAA in 500 mL of 50% aqueous ethanol (v/v); the prepared solutions were stored in a dark bottle kept at 4 °C.

### *2.4. Preparation of IBA-AgNPs and NAA -AgNPs mixture*

The IBA (10 mM) or NAA (10 mM) was mixed to the freshly prepared silver nanoparticles (1 mM); the mixture was subsequently stirred for 30 min. The prepared IBA and NAA-AgNPs mixture were separately stored in a dark bottle kept at 4 °C for further uses.

### *2.5. Characterization of silver nanoparticles*

#### *2.5.1. UV-Vis spectroscopy*

A UV/VIS spectrophotometer (T80, PG Instruments Ltd, UK) was used for the scan spectrum characterization of the synthesized AgNPs at the central lab of the Biochemistry Department, Faculty of Agriculture, Cairo University. The scanning range of the samples was 300–700 nm. Millie-Q water was used as a blank reference.

#### *2.5.2. Transmission Electron Microscopy (TEM)*

A morphological analysis, including the size and shape of the synthesized nanoparticles, was performed using transmission electron microscopy (JEOL JEM-1400, USA) at the Cairo University Research Park (CURP), Faculty of Agriculture, Cairo University. A drop of Millie-Q water, which dissolved synthesized nanoparticles, was placed on a carbon grid (C-grid). The images were obtained at a bias voltage of 40-120 kV.

#### *2.5.3. Particle Size and Zeta Potential*

Particle size and zeta potential of the synthesized nanoparticles were measured by photon correlation spectroscopy and laser Doppler anemometry, respectively, using a Zetasizer® 3000 particulate size description analyzer (Malvern Instruments, UK). The size was measured three times at 25 °C and a 90° scattering angle, and each measurement was recorded for 3 min. The mean hydrodynamic diameter was generated by cumulative analysis. The automatic mode of an aqueous dip cell was used to measure the zeta potential.

### *2.6. Plant material and rooting treatments*

This study was conducted inside the experimental greenhouse (70% shade-net) at the Faculty of Agriculture, Cairo University (031°12'65"E longitude, 30°00'48"N latitude). The plant materials were collected from a 35-year-old guava tree grafted onto seedling rootstocks and growing in the experimental orchard of the Pomology Department, Faculty of Agriculture, Cairo University. These trees were previously selected and characterized as promising guava genotypes by Bakr *et al.* **[42]**. Active growing shoots were collected during June and semi-hard wood cuttings of 15-20 cm in length with a couple of half-leaves at the upper nodes were prepared and subjected to six rooting treatments as follows:

1. Dipping in IBA (10 mM) solution.
2. Dipping in NAA (10 mM) solution.
3. Dipping in IBA-AgNPs mixture.
4. Dipping in NAA-AgNPs mixture.
5. Dipping in IBA followed by dipping in AgNPs solution.
6. Dipping in NAA followed by dipping in AgNPs solution.

Each treatment contained 60 cuttings divided into three replications, 20 cuttings for each. The cuttings were dipped in each rooting treatment for 30 seconds before being planted into plastic trays (15 cm depth) filled with a 1:3 mixture of sand and peatmoss and kept under a mist irrigation system; the cuttings were intermittently misted (15 s misting followed by 10 min pause) from 07:00–18:00 using micro sprinklers, with the mist frequency being automatically regulated. After two months of planting, the measurements of the rooting potential of guava semi-hard wood cutting for each treatment were examined; the measured parameters included rooting percentage, number of roots (roots > 2 mm), root length (cm), number of leaves per each transplant (the remaining leaves plus the newly emerged leaves), and root fresh and dry weight (g).

### *2.7.* Statistical analysis

The experiment was carried out in a randomized complete block design **[43]**. The assumptions of normality were tested by Shapiro-Wilk's test **[44]**; analysis of variance was performed using the R software (version 4.0.5, R Core Team, Vienna, Austria). The mean and standard error (SE) were calculated from three replicates per treatment, and the significant differences between treatments were assessed by means of multiple Duncan range test at significance level of 0.05 **[45]**.

## 3. Results and Discussion

### *3.1. Characterization of the synthesized silver nanoparticles*

#### *3.1.1. UV-Vis spectroscopy*

The UV-VIS spectra and absorption spectrum of the synthesized AgNPs are shown in Fig. (1). AgNPs exhibit a well-defined absorption peak in the scanning range from 300 to 700 nm. The maximum absorbance for the AgNPs sample measured is 410 nm. The characteristic feature of the peak between 400-500 nm in the UV-VIS spectrum validated the synthesis of AgNPs, and it is attributable to plasmon surface resonance (PSR) excitation **[46,47]**. According to Singh *et al.* **[48]**, the UV–Vis peak is between 400 and 435 nm, indicating the synthesis of well-dispersed spherical AgNPs.

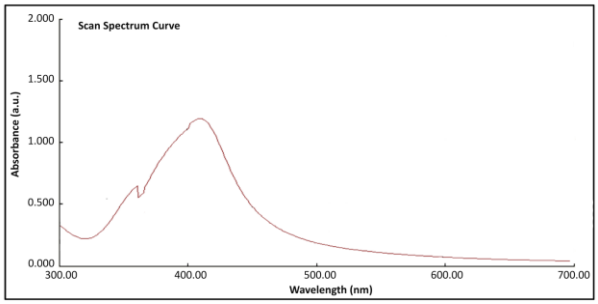


Fig. 1. UV–Vis absorption spectra of synthesized AgNPs

#### *3.1.2. Transmission electron microscopy (TEM)*

TEM analysis was employed to evaluate the shape and size of AgNPs (Fig. 2). The individual AgNPs are mostly dark spherical objects. The average size of silver nanoparticles ranged from 14.8 to 32.4 nm. The TEM image indicated that the AgNPs were spherical in shape and scattered in the solution. The TEM micrograph demonstrated that the mean size of the prepared nanoparticle is comparable to the particle size that has been reported in previous studies **[49,50]**. Therefore, the synthesised AgNPs in our study represented typical nanoparticles in terms of shape and size.

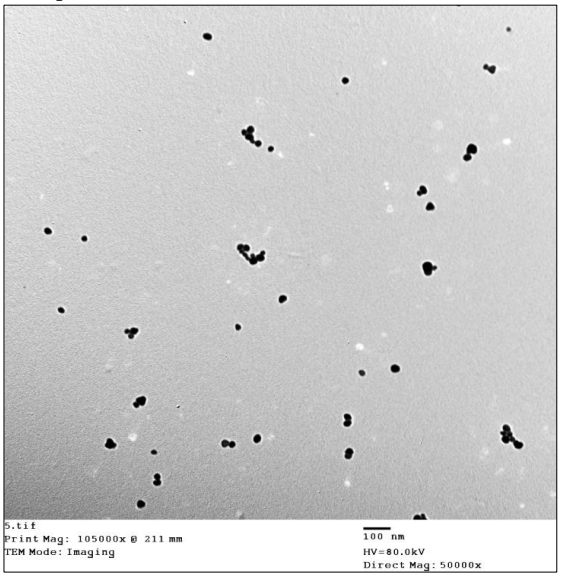


Fig. 2. The transmission electron microscope micrographs of synthesized AgNPs

### *3.1.3. Zeta potential and particle size of AgNPs*

The mean size and size distribution of silver nanoparticle suspension were analyzed using the Zetasizer analysis. The size distribution profile (Fig. 3) represents a typical batch of nanoparticles with a mean diameter of 12.09 nm and a narrow size distribution (polydispersity index < 1). A zeta potential was applied to determine the surface charges of silver nanoparticles. The zeta potential may greatly influence particle stability in suspension through the electrostatic repulsion between particles. Fig. (4) shows that the surfaces of AgNPs have a negative charge of about -31.5 mV. The value of zeta potential is useful for predicting the interactions between particles **[51]**. The high negative charge confirms the repulsion among the particles and thereby increases the stability of the AgNPs **[52]**. The negative charge confirms the role of PEG as stabilizing agent in preventing the aggregation of the AgNPs due to electrostatic repulsion among the negative charges **[53]**.

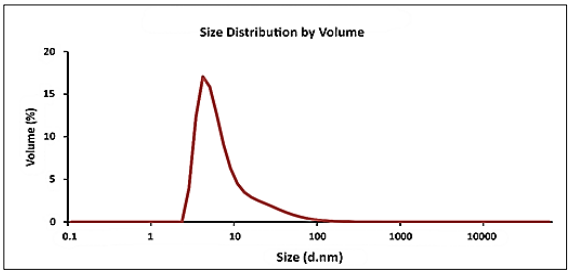


Fig. 3. Size distribution by intensity of AgNPs.

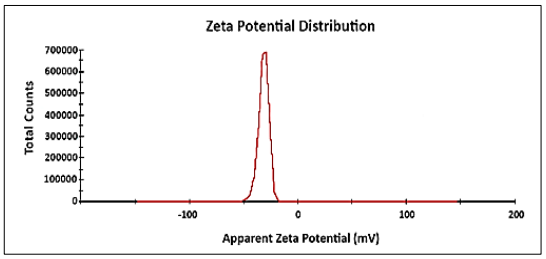


Fig. 4. Zeta potential distribution of AgNPs

## *3.2. The effect of silver nanoparticles on rooting potential*

### *3.2.1. Rooting percentage*

According to the data illustrated in Fig. (5), auxin and AgNPs treatments had a significant effect ($P \leq 0.05$) on the rooting percentage of guava cuttings. IBA recorded a significantly higher rooting percentage (26.67%) compared with NAA (5.34%). Concerning the effect of AgNPs, the mixture of auxin and AgNPs (auxin-AgNPs) and double-dipping (auxin+AgNPs) treatments improved the rooting potential of guava cuttings; NAA-AgNPs increased rooting percentage by 9.79 times (52.33%) while double-dipping treatments recorded 5.91 times increase in rooting percentage compared with NAA (5.33%). Both the BA-AgNPs and double-dipping (IBA+AgNPs) treatments statistically recorded similar results (31.6 and 33.3%, respectively).

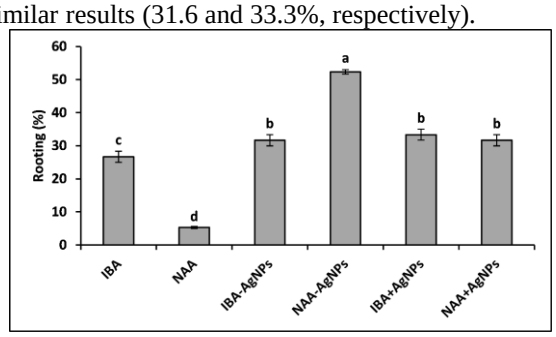


Fig. 5. Effect of silver NPs and hormonal treatments on the rooting percentage of guava stem cutting. Means with different letters between treatments are significantly different at $P \leq 0.05$. Vertical bars represent standard error (± SE).

### *3.2.2. Root number and length*

The presented data in Fig. (6) indicated that the number of roots developed per cutting was significantly affected by auxin and silver nanoparticle treatments. NAA recorded the lowest root number (2 roots), followed by the NAA-AgNPs treatment (5 roots), while the dipping in IBA-AgNPs and the double-dipping with IBA then AgNPs (IBA+AgNPs) recorded significantly ($P \leq 0.05$) the highest root number (18 roots), followed by the treatment with IBA (13.5 roots). Also, root lengths were significantly affected by different rooting treatments (Fig. 7); IBA recorded a higher root length (8.67 cm) compared with NAA (2 cm), IBA+AgNPs (8.11 cm), and NAA+AgNPs (8.56 cm), while IBA-AgNPs and NAA-AgNPs mixture recorded the highest root lengths (12.89 and 12.78 cm, respectively).

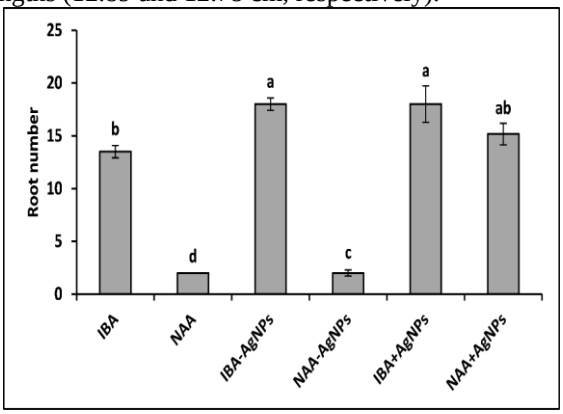


Fig. 6. Effect of silver NPs and hormonal treatments on the root number of guava stem cutting. Means with different letters between treatments are significantly different at $P \leq 0.05$. Vertical bars represent standard error (± SE).

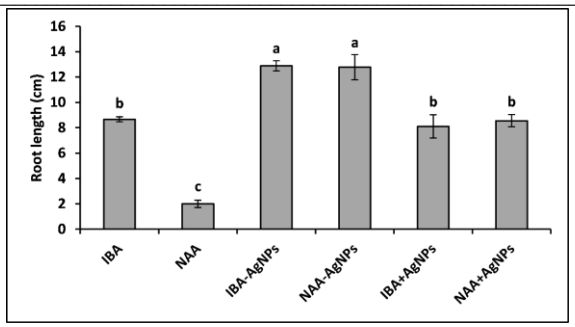


Fig. 7. Effect of silver NPs and hormonal treatments on the root length of guava stem cutting. Means with different letters between treatments are significantly different at $P \leq 0.05$. Vertical bars represent standard error (± SE).

### *3.2.3. Roots weight*

The effect of hormonal and AgNPs treatments on root fresh weight (FW) and dry weight (DW) per transplant is shown in Fig. (8 and 9). The treatment of guava cuttings with IBA significantly enhanced root fresh (0.53 g) and dry weight (0.38 g) compared to the NAA treatment, which recorded the lowest values (0.20 and 0.10 g, respectively). Generally, AgNPs significantly improved the root fresh and dry weight, IBA-AgNPs had significantly higher root fresh and dry weight compared with NAA-AgNPs, while there were non-significant differences between dipping treatments with IBA+AgNPs or NAA+AgNPs treatments on root FW. In other words, IBA-AgNPs had the highest root FW value (0.81 g), and IBA-AgNPs and IBA+AgNPs had the highest root DW values (0.54 and 0.48 g, respectively), compared to all other experimental treatments.

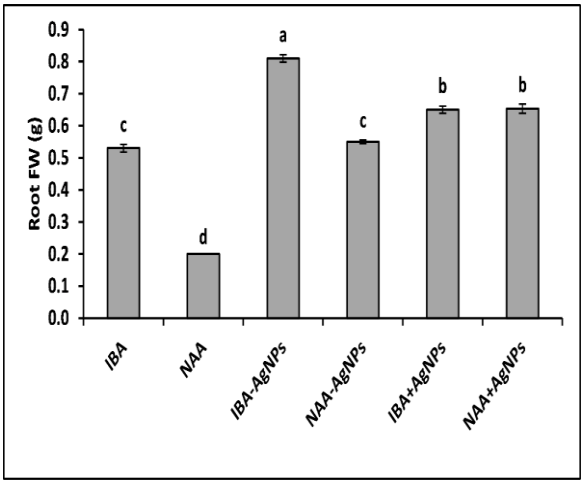


Fig. 8. Effect of silver NPs and hormonal treatments on the root fresh weight of guava stem cutting. Means with different letters between treatments are significantly different at $P \leq 0.05$. Vertical bars represent standard error (± SE).

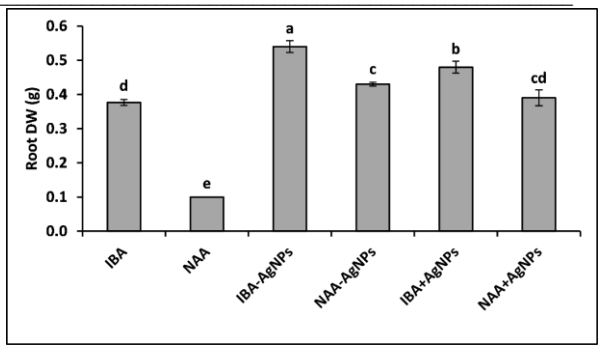


Fig. 9. Effect of silver NPs and hormonal treatments on the root dry weight of guava stem cutting. Means with different letters between treatments are significantly different at $P \leq 0.05$. Vertical bars represent standard error (± SE).

### *3.2.4. Leaves number*

Results in Fig. (10), indicate that there are slight differences between the examined rooting treatments in leaf number per cutting. The highest leaf number was recorded for IBA treatment. NAA-treated cuttings showed complete defoliation at the end of the rooting experiment. The NAA+AgNPs treatment recorded the highest leaf number (4 leaves) compared with all other treatments. Moreover, there were no significant differences between the IBA-AgNPs and IBA+AgNPs treatments.

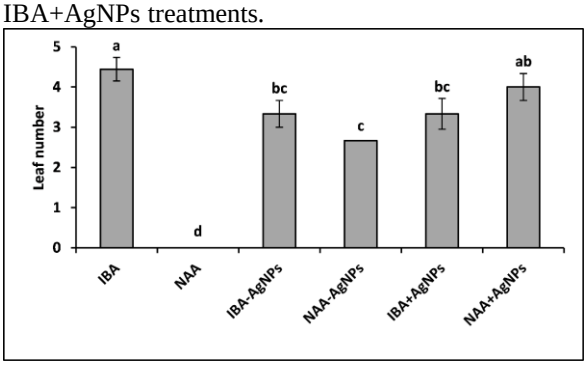


Fig. 10. Effect of silver NPs and hormonal treatments on the leaf number of guava stem cutting. Means with different letters between treatments are significantly different at $P \leq 0.05$. Vertical bars represent standard error (± SE).

Guava is traditionally propagated by seeds, which is the simplest propagation method. At the same time, seedling trees are undesirable in commercial orchards; they represent a great variability in productivity and fruit quality. In addition, seedling trees have a long juvenility period **[54]**. In contrast, vegetative propagation produces uniform trees with a short juvenile period compared to seedling plants **[1]**. Propagation by cuttings has significant advantages, such as obtaining true-to-type trees and being easy to implement; it insures production of nursery plants in one growing season **[4]**. Exogenous auxin application is commonly used to stimulate adventitious root

formation of stem-cuttings **[19,20]**. The effects of IBA and NAA on rooting of guava cuttings were previously reported **[21,22]**. In the present study, IBA significantly enhanced rooting parameters compared with NAA; the obtained results are in agreement with previous research which confirms that IBA is efficient in inducing root formation of guava cuttings. IBA significantly exhibited higher rooting percentage, number of roots per cutting, and root length **[7,13,21,55]**. The efficiency of IBA compared to NAA may be due to the slow and continuous release of IAA from IBA **[56,57]**; enhanced translocation of carbohydrates to the base of cuttings **[6]**; also an increase in indole-3-acetyl-aspartic acid (IAAsp) was reported in avocado micro-cuttings treated with IBA which stimulates normal growth of root meristemoids **[58]**. The obtained results confirm the inferior effect of NAA in root induction; as NAA is very stable, which may block the development of root meristemoids **[59]**, root elongation is very sensitive to auxin and the higher concentration may inhibit rooting **[60]**. IBA promotes root elongation by influencing the synthesis of enzymes involved in cell enlargement **[61,62]**. Roots developed with NAA treated cutting were shorter in length; and leaf abscission was noticed in NAA, whereas IBA produced longer roots. Similar results were previously reported by Ali *et al.* **[63]**. The higher value of root fresh and dry weight in IBA treated cutting may be attributed to the higher number of roots and root length; the effect of AgNPs in promoting root growth, increasing root elongation and fresh and dry biomass of roots was previously reported in different plant species including rice **[32,36]**, *Arabidopsis* **[35]**, *Populas tremula* **[34]**, *Eruca sativa* **[33]** and strawberry **[37]**. Our results demonstrated that AgNPs had a positive effect on the rooting parameters of guava cutting; the effect of AgNPs on root induction and root growth may be due to the effect of AgNPs on blocking ethylene signaling **[64]**. $Ag^{+}$ inhibits ethylene action upon binding to ethylene receptors through replacement of the $Cu^{+2}$ ions by $Ag^{+}$ which blocks ethylene receptors and inhibits the ethylene action **[65]**. The higher concentrations of auxin inhibit rooting elongation by increasing ethylene biosynthesis **[60]**. Our results showed that AgNPs significantly improved rooting parameters of NAA treated cutting; this may be due to the effect of AgNPs as ethylene inhibitors **[64-66]**. De Klerk and Hanecakova **[67]** indicated that the high NAA concentrations increase ethylene synthesis in mung bean cuttings, which inhibits rooting. Moreover, the application of silver thiosulfate (inhibitor of ethylene action) or aminoethoxyvinylglycine (inhibitor of ethylene synthesis) promotes rooting even at high NAA concentrations. Also, AgNPs increase *Arabidopsis* root elongation and increase number of cells in the root apical meristem **[68]**. Moreover, AgNPs inhibit defoliation of plant leaves through inhibiting the activity of ethylene gas **[69]**. Furthermore, AgNPs treatments can indirectly influence the rooting potential of stem cuttings through inhibition of soil-borne pathogens. The high moisture during rooting of stem cuttings under mist irrigation often promotes the spread of soil-borne pathogens, which usually attack the stem cutting base **[70]**. In this regard, silver nanoparticles (AgNPs) exhibited highly antimicrobial activity against plant pathogens **[39,40]**.

## 4. Conclusion

According to the obtained results, auxin type significantly affected all rooting parameters; higher rooting percentage, root number, root length, and root weight were recorded with IBA treatment. Interestingly, silver nanoparticles mixed with auxin treatments significantly improved rooting potential and recorded higher values of rooting parameters compared with the auxin treatments alone. The present results demonstrated the stimulatory effect of auxin and AgNPs mixture on the rooting of guava stem cuttings. Therefore, it could be a novel method for commercial vegetative propagation.

## 5. Conflicts of interest

There are no conflicts to declare.

## 6. References

[1] Singh, G., Gupta, S., Mishra, R., and Singh, A. (2007). Technique for rapid multiplication of Guava (*Psidium guajava* L.). *Acta Horticulturae*, 735, 177-183. https://doi.org/10.17660/ActaHortic.2007.735.22

[2] Hassimotto N. M., Genovese M. I., and Lajolo F. M. (2005) Antioxidant activity of dietary fruits, vegetables and commercial frozen fruit pulps. *Journal of Agricultural and Food Chemistry*, 53: 2928–2935. https://doi.org/10.1021/jf047894h

[3] Geidam Y. A., Ambali A. G., and Onyeyili P. A. (2007). Phytochemical screening and antibacterial properties of organic solvent fractions of *Psidium guajava* aqueous leaf extracts. *International Journal of Pharmacology*, 3, 68-73

[4] Pereira, F. M. P., Usman, M., Mayer, N. A., Nachtigal, J. C., Maphanga, O. R., and Willemse, S. (2017). Advances in guava propagation. *Revista Brasileira de Fruticultura*, 39(4), 1-24. https://doi.org/10.1590/0100-29452017358

[5] Ali, N., Mulwa, R. M. S., Mortan, M. A., and Skirvin, R. M. (2003) Micropropagation of guava (*Psidium guajava* L.). *Journal of Horticultural*

*Science and Biotechnology*, 78(5): 739-741. https://doi.org/10.1080/14620316.2003.11511692

[6] Hartmann, H. T., Kester, D. E., Davies F. T., and Geneve R.L. (2014). Hartmann and Kester's Plant Propagation: principles and Practices, 8th ed. Prentice-Hall, London.

[7] Abdullah, A. T. M., Hossain, M. A., and Bhuiyan, M. K. (2006). Clonal propagation of guava (*Psidium guajava* Linn.) by stem cutting from mature stock plants. *Journal of Forest Research*, 17, 301–304. https://doi.org/10.1007/s11676-006-0069-2

[8] Chandra R., and Mishra M. (2007) Biotechnological interventions for improvement of Guava (*Psidium guajava* L). *Acta Horticulturae*, 735, 117-125. DOI:10.17660/ActaHortic.2007.735.15

[9] Rymbai, H., and Reddy, G. S. (2010). Effect of IBA, time of layering and rooting media on air-layers and plantlets survival under different growing nursery conditions in guava. *Indian Journal of Horticulture*, 67(4), 99-104.

[10] Elezaby, A. A., and Abdallatif, A. M. (2017). *In vitro* propagation of guava (*Psidium guajava* L.): Effect of antioxidants, nutrient media and growth regulators. *Journal of Horticultural Science and Ornamental Plants*, 9(3), 144-149. DOI: 10.5829/idosi.jhsop.2017.144.149

[11] Singh, A. (1986). Fruit Physiology and Production. Kalyani, New Delhi, pp. 328-3

[12] Rai, M. K., Asthana, P., Jaiswal, V. S., and Jaiswal, U. (2010). Biotechnological advances in guava (*Psidium guajava* L.): recent developments and prospects for further research. *Trees*, 24(1), 1-12. DOI 10.1007/s00468-009-0384-2

[13] Kareem A., Jaskani M. J., Fatima B., and Sadia B. (2013). Clonal multiplication of guava through softwood cuttings under mist conditions. Pakistan Journal of Agricultural Science, 50(1):23-27.

[14] Pandey S. K., and Bisen A. (2010). Effect of mechanical treatments on rooting in cuttings of guava, lemon and pomegranate. *Journal of Horticulture and Forestry*, 2(5):95-98. https://doi.org/10.5897/JHF.9000031

[15] El-Sharony, T. F., Rashedy, A .A., Abd Allatif, A. M., and Hassan, S. A .M. (2018). Stimulating rooting of guava cuttings by chemical and physical treatments. *Acta Horticulturae*, 1216, 91-98. DOI: 10.17660/ActaHortic.2018.1216.12

[16] Pacurar, D. I., Perrone, I., and Bellini, C. (2014). Auxin is a central player in the hormone cross-talks that control adventitious rooting. *Physiologia Plantarum*, 151(1), 83-96. https://doi.org/10.1111/ppl.12171

[17] Fogaça, C. M., and Fett-Neto, A. G. (2005). Role of auxin and its modulators in the adventitious rooting of *Eucalyptus* species differing in recalcitrance. *Plant Growth Regulation*, 45(1), 1-10. https://doi.org/10.1007/s10725-004-6547-7

[18] Das, P., Basak, U. C., and Das, A. B. (1997). Metabolic changes during rooting in pre-girdled stem cuttings and air-layers of Heritiera. *Botanical Bulletin of Academia Sinica*, 38.

[19] Tchoundjeu, Z., Avana, M. L., Leakey, R. R. B., Simons, A. J., Assah, E., Duguma, B., and Bell, J. M. (2002). Vegetative propagation of *Prunus africana*: effects of rooting medium, auxin concentrations and leaf area. *Agroforestry systems*, 54(3), 183-192. https://doi.org/10.1023/A:1016049004139

[20] Tworkoski, T., and Takeda, F. (2007). Rooting response of shoot cuttings from three peach growth habits. *Scientia Horticulturae*, 115(1), 98-100. https://doi.org/10.1016/j.scienta.2007.08.004

[21] Rahman, N., Nabi, T. G., and Taslim, J. (2004). Effect of different growth regulators and types of cuttings on rooting of guava (*Psidium guajava* L.). *Quarterly Science Vision*, *91*(2), 3-4.

[22] Gautam, N. N., Singh, K., Singh, B., Seal, S., Goel, A., and Goel, V. L. (2010). Studies on Clonal Multiplication of Guava ('Psidium guajava' L.) through Cutting under Controlled Conditions. Australian *Journal of Crop Science*, 4(9), 666-669.

[23] Sorin, C., Bussell, J. D., Camus, I., Ljung, K., Kowalczyk, M., Geiss, G., and Bellini, C. (2005). Auxin and light control of adventitious rooting in Arabidopsis require ARGONAUTE1. *The Plant Cell*, 17(5), 1343-1359. https://doi.org/10.1105/tpc.105.031625

[24] Casanova-Sáez, R., and Voß, U. (2019). Auxin metabolism controls developmental decisions in land plants. *Trends in Plant Science*, 24(8), 741-754. https://doi.org/10.1016/j.tplants.2019.05.006

[25] Prasad, R., Bhattacharyya, A., and Nguyen, Q. D. (2017). Nanotechnology in sustainable agriculture: recent

developments, challenges, and perspectives. *Frontiers in microbiology*, 8, 1014. https://doi.org/10.3389/fmicb.2017.01014.

[26] Darwesh, O., and Matter, I. (2021). Challenges of Nanotechnology Applications in Addressing Environmental Pollution. *Egyptian Journal of Chemistry*. 65(2), 275-285. DOI: 10.21608/ejchem.2021.86072.4172

[27] Ali, M. A., Abdel-Moein, N. M., Owis, A. S., Ahmed, S. E., and Hanafy, E. A. (2022). Preparation, characterization, antioxidant and antimicrobial activities of lignin and eco-friendly lignin nanoparticles from Egyptian cotton stalks. *Egyptian Journal of Chemistry*, *65*(1), 703-716. DOI: 10.21608/ejchem.2021.86987.4221

[28] Torney F., Trewyn B. G., Lin V. S.-Y., and Wang K. (2007). Mesoporous silica nanoparticles deliver DNA and chemicals into plants. *Nature Nanotechnology*, 2, 295–300. https://doi.org/10.1038/nnano.2007.108

[29] Siddiqui M. H., Al-Whaibi M. H., Firoz M., and Al-Khaishany M. Y. (2015) Role of nanoparticles in plants. In: M. H. Siddiqui, M. H. Al-Whaibi and F. Mohammad (eds) Nanotechnology and plant sciences. Springer International Publishing Switzerland pp. 19-35. DOI 10.1007/978-3-319-14502-0_2

[30] Gupta N., Upadhyaya C. P., Singh A., Abd-Elsalam K. A., and Prasad R. (2018a) Applications of Silver Nanoparticles in Plant Protection. In: Abd-Elsalam K., Prasad R. (eds) Nanobiotechnology Applications in Plant Protection. Nanotechnology in the Life Sciences. Springer, Cham, pp. 247-265 https://doi.org/10.1007/978-3-319-91161-8_9

[31] Abo-El-Yazid, Z. H., Ahmed, O. K., El-Tholoth, M., and Ali, M. A. Green synthesized silver nanoparticles using *Cyperus rotundus* L. extract as a potential antiviral agent against infectious laryngotracheitis and infectious bronchitis viruses in chickens. *Chemical and Biological Technologies in Agriculture*, 9, 55 (2022). https://doi.org/10.1186/s40538-022-00325-z

[32] Mirzajani, F., Askari, H., Hamzelou, S., Farzaneh, M., and Ghassempour, A. (2013). Effect of silver nanoparticles on Oryza sativa L. and its rhizosphere bacteria. *Ecotoxicology and Environmental Safety*, 88, 48-54. https://doi.org/10.1016/j.ecoenv.2012.10.018

[33] Vannini, C., Domingo, G., Onelli, E., Prinsi, B., Marsoni, M., Espen, L., and Bracale, M. (2013). Morphological and proteomic responses of *Eruca sativa* exposed to silver nanoparticles or silver nitrate. *PloS one*, 8(7), e68752. https://doi.org/10.1371/journal.pone.0068752

[34] Wang, J., Koo, Y., Alexander, A., Yang, Y., Westerhof, S., Zhang, Q., and Alvarez, P. J. (2013). Phytostimulation of poplars and *Arabidopsis* exposed to silver nanoparticles and Ag+ at sublethal concentrations. *Environmental Science and Technology*, 47(10), 5442-5449. https://doi.org/10.1021/es4004334

[35] Syu Y. Y., Hung J. H., Chen J. C., and Chuang H. W. (2014). Impacts of size and shape of silver nanoparticles on *Arabidopsis* plant growth and gene expression. Plant Physiol. Biochem., 83, 57–64. https://doi.org/10.1016/j.plaphy.2014.07.010

[36] Gupta, S. D., Agarwal, A., and Pradhan, S. (2018b). Phytostimulatory effect of silver nanoparticles (AgNPs) on rice seedling growth: An insight from antioxidative enzyme activities and gene expression patterns. *Ecotoxicology and environmental safety*, *161*, 624-633. https://doi.org/10.1016/j.ecoenv.2018.06.023

[37] Tung, H. T., Thuong, T. T., Luan, V. Q., Hien, V. T., Hieu, T., Nam, N. B., ... and Nhut, D. T. (2021). Silver nanoparticles improved explant disinfection, *in vitro* growth, runner formation and limited ethylene accumulation during micropropagation of strawberry (*Fragaria*× *ananassa*). *Plant Cell, Tissue and Organ Culture (PCTOC)*, 145(2), 393-403. https://doi.org/10.1007/s11240-021-02015-4

[38] Bruna, T., Maldonado-Bravo, F., Jara, P., and Caro, N. (2021). Silver nanoparticles and their antibacterial applications. *International Journal of Molecular Sciences*, 22(13), 7202. https://doi.org/10.3390/ijms22137202

[39] Gogoi, B., Kumar, R., Upadhyay, J., and Borah, D. (2020). Facile biogenic synthesis of silver nanoparticles (AgNPs) by Citrus grandis (L.) Osbeck fruit extract with

excellent antimicrobial potential against plant pathogens. *SN Applied Sciences*, 2(10), 1-7., 1723 (2020). https://doi.org/10.1007/s42452-020-03529-w

[40] Khan, M., Khan, A. U., Bogdanchikova, N., and Garibo, D. (2021). Antibacterial and antifungal studies of biosynthesized silver nanoparticles against plant parasitic nematode *Meloidogyne incognita*, Plant pathogens *Ralstonia solanacearum* and *Fusarium oxysporum*. *Molecules*, 26(9), 2462. 2462; https://doi.org/10.3390/molecules26092462

[41] Motol, R. K. C., Espineli, C. A., Tapit, C. M. V., and Tiangco, C. E. (2020). Synthesis and characterization of silver nanoparticles as a potential sensor for volatile organosulfides for visual detection of postharvest storage in garlic. In IOP Conference Series: Materials Science and Engineering, 778, 012002. doi:10.1088/1757-899X/778/1/012002

[42] Bakr, E.I.M., A.T.M. Salem, A. E. Killany and G. M. Hasseb (1987) A study on growth and fruiting of some guava hybrids. Zagazig Journal of Agric, Res., 14 (1): 42-70.

[43] Snedecor W.B., and Cochran G.W. (1967) Statistical Methods 6th Ed. Iowa State College. Press Amer. Low, U.S.A.

[44] Shapiro S.S., Wilk M.B. (1965) An analysis of variance test for normality (complete samples). *Biometrika*, 52(3/4): 591-611. https://doi.org/10.2307/2333709

[45] Duncan D.B., (1955) Multiple Range and Multiple F-Tests. *Biometrics*,111-42. DOI: 10.2307/3001478

[46] Shousha, W. G., Aboulthana, W. M., Salama, A. H., Saleh, M. H., and Essawy, E. A. (2019). Evaluation of the biological activity of Moringa oleifera leaves extract after incorporating silver nanoparticles, *in vitro* study. *Bulletin of the National Research Centre*, 43, 212. https://doi.org/10.1186/s42269-019-0221-8

[47] Solaiman, M. A., Ali, M. A., Abdel-Moein, N. M., and Mahmoud, E. A. (2020). Synthesis of Ag-NPs developed by green-chemically method and evaluation of antioxidant activities and anti-inflammatory of synthesized nanoparticles against LPS-induced NO in RAW 264.7 macrophages. *Biocatalysis and Agricultural Biotechnology*, 29, 101832.

[48] Singh, A., Sharma, B., and Deswal, R. (2018). Green silver nanoparticles from novel Brassicaceae cultivars with enhanced antimicrobial potential than earlier reported Brassicaceae members. *Journal of Trace Elements in Medicine and Biology*, 47, 1-11. https://doi.org/10.1016/j.jtemb.2018.01.001

[49] Huq, M. (2020). Green synthesis of silver nanoparticles using *Pseudoduganella eburnea* MAHUQ-39 and their antimicrobial mechanisms investigation against drug resistant human pathogens. *International Journal of Molecular Sciences*, 21(4), 1510. https://doi.org/10.3390/ijms21041510

[50] Hmmam, I., Zaid, N. M., Mamdouh, B., Abdallatif, A., Abd-Elfattah, M., and Ali, M. (2021). Storage Behavior of "Seddik" mango fruit coated with CMC and guar gum-based silver nanoparticles. *Horticulturae*, *7*(3), 44. https://doi.org/10.3390/horticulturae7030044

[51] Wangoo, N., Kaushal, J., Bhasin, K. K., Mehta, S. K., and Suri, C. R. (2010). Zeta potential based colorimetric immunoassay for the direct detection of diabetic marker HbA1c using gold nanoprobes. *Chemical Communications*, 46(31), 5755-5757. 10.1039/C0CC00224K

[52] Rao, Y. S., Kotakadi, V. S., Prasad, T. N. V. K. V., Reddy, A. V., and Gopal, D. S. (2013). Green synthesis and spectral characterization of silver nanoparticles from Lakshmi tulasi (*Ocimum sanctum*) leaf extract. Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy, 103, 156-159. https://doi.org/10.1016/j.saa.2012.11.028

[53] Ajitha, B., Reddy, Y. A. K., Reddy, P. S., Jeon, H. J., and Ahn, C. W. (2016). Role of capping agents in controlling silver nanoparticles size, antibacterial activity and potential application as optical hydrogen peroxide sensor. *RSC advances*, 6(42), 36171-36179. 10.1039/C6RA03766F

[54] Pereira, F. (1990). Factors affecting guava production and quality with special reference to São Paulo, Brazil. *Acta Horticulturae*, 275, 103-110 https://doi.org/10.17660/ActaHortic.1990.275.8

[55] Shahzad, U., Kareem, A., Altaf, K., Zaman, S., Ditta, A., and Yousafi, Q. (2019). Effects of auxin and media additives on the clonal propagation of guava cuttings (*Psidium guajava* L.) Var. Chinese Gola. *Agricultural Science and Food Research*, 10, 265. doi: 10.35259/2593-9173.19.10.265.

[56] Epstein, E. and J. L. Muller. (1993). Indole-3-butyric acid in plants: Occurrence, synthesis, metabolism and transport. *Physiologia Plantarum*, 88(2): 382-389. https://doi.org/10.1111/j.1399-3054.1993.tb05513.x

[57] Krieken, W. M., Breteler, H., Visser, M. H., and Mavridou, D. (1993). The role of the conversion of IBA into IAA on root regeneration in apple: Introduction of a test system. *Plant Cell Reports*, 12(4): 203-206. https://doi.org/10.1007/BF00237054

[58] Garcia. M. L. G., Romero, C. S., Munoz, A. B., Heredia, A., and Alfaro, F. P. (1994). Levels of endogenous indole-3-acetic acid and indole-3-butyric acid during adventitous rooting in Avocado micrcuttings. *Journal of Experimental Botany*, 45(6): 865-870. https://doi.org/10.1111/j.1399-3054.1991.tb02953.x

[59] Hausman, J. F. (1993). Changes in peroxidase activity, auxin level and ethylene production during root formation by poplar shoots raised *in vitro*. *Plant Growth Regulation*, 13(3): 263-268.https://doi.org/10.1007/BF00024847

[60] Kollmeier M., Felle H. H., and Horst W. J. (2000). Genotypical differences in aluminum resistance of maize are expressed in the distal part of the transition zone. Is reduced basipetal auxin flow involved in inhibition of root elongation by aluminum? *Plant Physiology*, 122:945–956. https://doi.org/10.1104/pp.122.3.945

[61] Wada, S., Tanimoto, E., and Masuda Y. (1998). Cell elongation and metabolic turnover of the cell wall as affected by auxin and cell wall degrading enzymes. *Plant and Cell Physiology*, 9(2): 369-376. https://doi.org/10.1093/oxfordjournals.pcp.a079353

[62] Muller, J. L. (2000). Indole-3-butyric acid in plant growth and development. *Plant Growth Regulation*, 32:219–230, 2000 DOI : 10.1023/A:1010746806891

[63] Ali, A., Ahmad, T., Abbasi, N. A., and Hafiz, I. A. (2009). Effect of different concentrations of auxins on *in vitro* rooting of olive cultivar 'Moraiolo'. *Pakistan Journal of Botany*, *41*(3), 1223-1231

[64] Ha, N. T. M., Do, C. M., Hoang, T. T., Dai Ngo, N., and Nhut, D. T. (2020). The effect of cobalt and silver nanoparticles on overcoming leaf abscission and enhanced growth of rose (Rosa hybrida L.'Baby Love') plantlets cultured *in vitro*. *Plant Cell, Tissue and Organ Culture (PCTOC)*, 1-13. https://doi.org/10.1007/s11240-020-01796-4

[65] Zhao, X. C., Qu, X., Mathews, D. E., and Schaller, G. E. (2002). Effect of ethylene pathway mutations upon expression of the ethylene receptor ETR1 from Arabidopsis. *Plant Physiology*, 130(4), 1983-1991. DOI: https://doi.org/10.1104/pp.011635

[66] Hegazi, E. S. S., Yousef, A., Abdallatif, A. M., Mahmoud, T. S., and Mostafa, M. K. M. (2021). Effect of Silver Nanoparticles, Medium Composition and Growth Regulators on in vitro propagation of Picual Olive Cultivar. *Egyptian Journal of Chemistry*, *64*(12), 6961-6969. DOI:10.21608/ejchem.2021.78774.3853

[67] De Klerk, G. J., and Hanecakova, J. (2008). Ethylene and rooting of mung bean cuttings. The role of auxin induced ethylene synthesis and phase-dependent effects. *Plant Growth Regulation*, 56(2), 203-209. https://doi.org/10.1007/s10725-008-9301-8

[68] Wang, L., Sun, J., Lin, L., Fu, Y., Alenius, H., Lindsey, K., and Chen, C. (2020). Silver nanoparticles regulate *Arabidopsis* root growth by concentration-dependent modification of reactive oxygen species accumulation and cell division. *Ecotoxicology and Environmental Safety*, 190, 110072. https://doi.org/10.1016/j.ecoenv.2019.110072

[69] Karuppanapandian, T, Wang, H. W., Prabakaran, N., Jeyalakshmi, K., Kwon, M., Manoharan, K., and Kim, W. (2011). 2,4-Dichlorophenoxyacetic acid-induced leaf senescence in mung bean (Vigna radiata L. Wilczek) and senescence inhibition by co-treatment with silver nanoparticles. *Plant Physiology* and *Biochemistry*, 49(2):168–177. https://doi.org/10.1016/j.plaph y.2010.11.007

[70] Aleandri, M. P., Chilosi, G., Bruni, N., Tomassini, A., Vettraino, A. M., and Vannini, A. (2015). Use of nursery potting mixes amended with local *Trichoderma* strains with multiple complementary mechanisms to control soil-borne diseases. *Crop Protection*, 67, 269-278. https://doi.org/10.1016/j.cropro.2014.10.023