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GC-MS characterization of *polygonatum geminiflorum* depicted by antibacterial efficacy of the biosynthesized silver nanoparticles using its leaf extract

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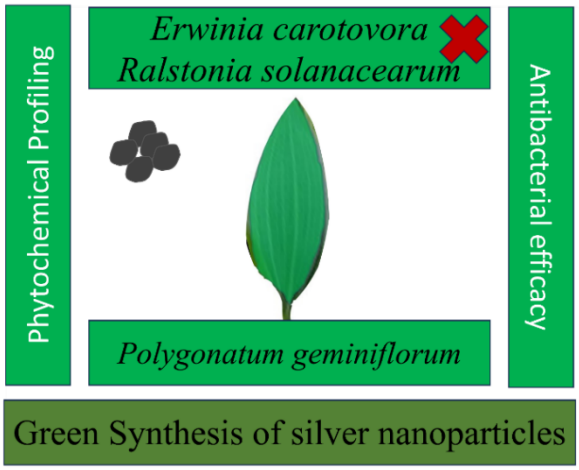
Graphical abstract



Abstract

The biological synthesis of nanomaterials is drawing immense interest because of their non-hazardous nature and enormous antimicrobial application. In the present study, we explored *Polygonatum geminiflorum* Decne for phytochemical profiling and biosynthesis of silver nanoparticles to control soft rot/blackleg and bacterial wilt pathogens of potato through *in vitro* experiment. Phytochemical screening indicated the presence of important secondary chemicals including tannins, glycosides, flavonoids and terpenoids, while, gas chromatography-mass spectrophotometry (GC-MS) study of leaf extract showed the presence of 30 phytochemicals, the most prominent among which included ç-Sitosterol and n-Hexadecanoic acid. The GC–MS qualitative analysis also supported the presence of bioactive compounds responsible for metal reduction processes and synthesized nanoparticles stabilization. *In vitro* study showed that concentration of 100µg/mL of AgNPs and AgNPs-PE efficiently control both *Erwinia carotovora* and *Ralstonia solanacearum*. The outcomes have provided an improved protocol to use prepared AgNPs against the tested pathogens without health hazards.

Keywords: *Ralstonia solanacearum, erwinia carotovora*, GC-MS, silver nanoparticles, potato, pathogens

1. Introduction

Potato (*Solanum tuberosum* L.) is amongst important crops consumed all over the world owing to its easy availability and rich nutrient capacity (Devaux *et al*. 2014). It is also amongst the commonly grown crop in Pakistan as well as around the world which is an excellent source of protein, minerals, vitamins and carbohydrates (Zaheer and Akhtar 2016). This essential vegetable crop is badly affected by plant pathogens, the result being reduced production (Djaya *et al*. 2019). *Erwinia carotovora* is one of the severe pathogens causing soft rot and blackleg diseases in potatoes (Ali *et al*. 2012). Similarly, *Ralstonia solanacearum* is another pathogen causing bacterial wilt of potatoes (Sagar *et al*. 2014). These pathogens have negative impacts and as a result, affecting the crop quality and quantity (Ranjan *et al*. 2016; Oyesola *et al*. 2021).

To control these pathogens, various conventional strategies are used including development of resistant varieties, chemical pesticides and bio-control agents (Cook 1985; Srinon *et al*. 2006). However, due to the pesticide's resistant capability the control of these pathogens is ineffectual (Srinon *et al*. 2006; Sun *et al*. 2012). Therefore, the modern investigators are working to produce novel antimicrobial drugs (León-Buitimea *et al*. 2020).

Apart from biotic stresses, several abiotic stresses including salinity, drought, high temperature and heavy metal also affect crop growth and production (Rafique *et al*. 2017; Tariq *et al*., 2021; Hassanisaadi *et al*. 2022; Salam *et al*. 2022). Over the past decades, climate changes and global warming has led to reduction of water resources, ultimately resulting in agricultural land reduction (Sadiq *et al*. 2013; Ali *et al*. 2014; Singh *et al*. 2021). These environmental factors badly reduce the crop production and quality which leads to global food insecurity (Ali *et al*. 2013 and 2022). To overcome this, the development of new techniques need to be developed for improvement of agricultural crops (Singh *et al*. 2021). Green chemistry deals with the production of nanomaterials using bio-reducing agent to overcome toxic and hazardous substances (Salem *et al*. 2021). Green chemistry utilizes natural substances to produce cost effective and non-toxic nanomaterials on large scale (Shah *et al*. 2015). Moreover, it includes the development of significant processes to produce nanomaterials that have positive ecofriendly effects (Yates and Dionysiou 2006; Wong and Karn 2012). Also, these procedures provide efficient solutions to environmental health problems without effecting human health (Khan 2020). Consequently, green chemistry produces safe and ecofriendly nanomaterials that utilize no toxic substances during synthesis process, hence, offering a substitute to other conventional physical and chemical processes (Krishnaswamy and Orsat 2017; Dilbar *et al*. 2023a).

Medicinal plants and fungi have been reported to fulfil the major healthcare needs, and, hence, are utilized in different traditional formulations (Rahman *et al*. 2022a and b; Hussain *et al*. 2023). They provide relatively simple, eco-friendly and safe methods for producing and provide capping layers to stabilize nanomaterial having a particular size and shape (Mata *et al*. 2015). Nanoparticles get synthesized through plants are ecofriendly and shows significant antimicrobial potential against pathogenic microorganisms (Gopinath *et al*. 2017; Handoko *et al*. 2019). Secondary metabolites present in the plants provide favorable routes to produce metallic nanoparticles on large scale (Kuppusamy *et al*. 2016). Biological method of nanoparticles synthesis combined the effects of plant constituents with nanoparticles to provide stability and increase its antimicrobial effects (Choudhury *et al*. 2016). Apart from others, silver nanoparticles were previously studied for different biological activities (Vishwasrao *et al*. 2019; Ahmad *et al*. 2022; Dilbar *et al*. 2023b). Moreover, the low concentrations of silver nanoparticles were found effective against phytopathogens having no toxicity to human health (Jeong *et al*. 2005; Khan *et al*. 2022).

Due to highly medicinal importance of *Polygonatum geminiflorum* (Ullah *et al*. 2021; Sher *et al*. 2022), the present work aimed to investigate phytochemical profile of its leaf extract and its further utilization in biosynthesis of silver nanoparticles. Moreover, the synthesize nanoparticles were studied for the antibacterial effects against potato's pathogens via *in vitro* experiments.

2. Materials and methods

Healthy leaves of P. geminiflorum were collected from Swat District of Khyber Pakhtunkhwa Province, Pakistan. The specimens were submitted to University of Swat herbarium (SWAT) which can be accessed using catalog (accession) number ''SWAT004621''. The healthy leaves were then dried under shade and were used for phytochemicals investigations and silver nanoparticles biosynthesis.

2.1. Preliminary phytochemical analysis

For phytochemicals investigation, 50 g dried leaf material of *P. geminiflorum* was dissolved in in 100 mL aqueous methanol (95%), which was simultaneously incubated for 24 h at 28°C. this was followed by filtration and then evaporation of the solution till the availability of 15 g final mass of crude extract for its further utilization in different phytochemical tests.

2.2. Test for tannins

The crude leaf extract (about 2 mg) was mixed in deionized water (20 mL) and heated in water bath for about 5 min followed by cooling the solution and its subsequent filtration. Then, dropwise addition of ferric chloride (10%) was accomplished to about 1 mL of the filtrate till brownish coloration which indicated the presence of tannins.

2.3. Test for flavonoids

The crude leaf extract (about 1 mg) was mixed in deionized water (10 mL) and heated in water bath for about 5 min followed by cooling the solution and its subsequent filtration. Then, dropwise addition of ferric chloride (20%) was accomplished to about 1 mL of the filtrate till yellow coloration which indicated the presence of flavonoids.

2.4. Test for glycosides

For this purpose, crude leaf extract (about 5 mg) was mixed in deionized water (10 mL) and heated for 15 min at 60 °C which was followed by filtration of the solution. About 5 mL filtrate was then reacted with glacial acetic acid (2 mL) and conc. H_2SO_4 (1 mL), then dropwise ferric chloride was added to it. The appearance of blue ring appearance at the bottom of the flask indicated the existence of glycosides.

2.5. Test for terpenoids

For this purpose, crude leaf extract (about 1 mg) was mixed in deionized water (5 mL), heated till boiling, followed by cooling and filtration of the solution. About 2 mL filtrate was then reacted with chloroform (2 mL) and conc. H_2SO_4 (1 mL) till the appearance of reddish brown coloration which indicated the presence of terpenoids.

2.6. GC-MS analysis

The leaf extract was subjected to GS-MS analysis carried out at Department of Chemistry, University of Peshawar. For extract preparation, 25 g of powdered leaf was dissolved in 200 mL methanol and kept in shaking incubator. After, 24 h the solution was filtered and 50 mL methanol was added to the extract and placed on shaking. After 24 h, the solution was filtered again and 50 mL methanol was added the extract followed by shaking incubation for 24 h. The solution was filtered and the

obtained extract was analyzed using Thermo Scientific (GC-MS) DSQ instrument. NIST/EPA/NIH mass spectral library was used for the identification of compounds. The GC-MS data were obtained on Elite-I mode with HP-5 column of 30 m × 0.25 mm × 0.25 µm and 70 eV energy. The Helium gas as a carrier at 1 mL/min flow rate was used and 230C of injector temperature. Initially the temperature was set up to 110 ^{o}C, which remains for 2 min and it was raised up to 200 ^{o}C which held for 10 min. The temperature was further increased to 280 ^{o}C which hold up for 9 min. The temperature for injecting and detecting was maintained respectively at 250 ^{o}C and 280 ^{o}C, while, it was retained at 200 ^{o}C for ion source. The MS of compounds in the sample extract was attained by electron ionization 70 eV energy and scan mode detector was run by 45-450 amu. A 0.5 seconds of scan interval was maintained at total running time of 27 min.

2.7. Biosynthesis and characterization of silver nanoparticles

Biosynthesis of silver nanoparticles has previously discussed in Ahmad *et al*. (2022). Briefly, 4 mM $AgNO_3$ solution was mixed with 5 mg mL^{-1} aqueous leaf extract in equal volume. The obtained solution was stirred and exposed to sunlight to notice change in coloration in reaction mixture. The solution after 15 min were placed in room temperature for 24 h. After 24 h, centrifugation of solution was accomplished at 14,000 rpm for 10 min. The pelleting and washing process was repeated multiple times to eradicate unreacted substances or impurities. This process produced pure washed nanoparticles which were used for further analysis.

The biosynthesized silver nanoparticles were previously analyzed using UV-Visible spectrophotometric and Fourier transmission infrared spectroscopy (FTIR), Transmission electron microscopy (TEM) and X-ray diffraction (XRD) characterizations (Ahmad *et al*. 2022).

2.8. In vitro antibacterial bioassay

The antibacterial activities against *R. solanacearum* and *E. carotovora* were performed for PE (plant extract), AgNPs (Green synthesized washed silver nanoparticles) and AgNPs-PE (plant-extract-coated silver nanoparticles). Briefly, a 96-well microtiter plate with change in concentrations (15.62, 31.25, 62.5, 125, 250, 500 and 1000 µg/mL) of PE, AgNPs and AgNPs-PE was utilized. Pure bacterial cultures were refreshed and incubated at 28 °C for 36 h, followed by its inoculation to nutrient broth and were grown overnight at 28 °C to adjust the OD (600) = 1. Next, each well of the microtiter plate was poured with 150 µL of each treatment (AgNPs, PE and AgNPs-PE) and equal volume of bacterial suspension was added to it. The OD of the prepared microtiter plate was measured at 0 h and placed in shaking incubator (200 rpm) at 28 °C. The experiment was replicated three times and control treatments contained bacterial suspension without AgNPs, PE and AgNPs-PE. After 24 h, the optical density (OD) was recorded and the growth inhibition pattern was calculated by measuring the OD at 0 h and 24 h time points using the following formula:

$$\text{Antibacterial activity} = \frac{\text{Control} - \text{Treatment}}{\text{Control}} \times 100 \quad (1)$$

3. Results

3.1. Phytochemical analysis

The methanolic leaf extract of P. geminiflorum has shown the existence of prominent phytochemicals which included flavonoids, glycosides, terpenoids and tannins that are presented in Table 1.

Table 1. Qualitative phytochemicals screening of P. geminiflorum leaf extract

S. No	Phytochemicals	Result	Indication
1	Glycosides	Present	Appearance of blue ring
2	Flavonoids	Present	Yellowish color
3	Tannins	Present	Brownish color
4	Terpenoids	Present	Reddish brown color

3.2. GC-MS analysis

GC-MS characterization of leaf extract showed the presence of thirty important plant compounds which indicated the importance of this high valued medicinal plant (figure 1). The compounds were identified on the basis of retention times, peak percent area, molecular formula and molecular weight (table 2). The first compound detected at retention time of 7.28 were Cyclohexanecarboxylic acid, 2-hydroxy-, ethyl ester having 0.57 percent area. Other compounds detected were, Hydroquinone, Phenol, 2-(6-methyl-5,6-dihydrothiazolo[2,3-c][1,2,4]triazol-3-yl)-, Cyclohexane, 1,4-dimethyl-2-octadecyl-, Benzene, (1-pentylheptyl)-, Pentacosane, 13-phenyl-, n-Hexadecanoic acid, acetate, Oleic Acid, Azuleno[4,5-b]furan-2(3H)-one,9a-[(acetyloxy)methyl]decahydro-6a,9-dihydroxy-6-methyl-3-methylene-, [3aS-(3aà,6á,6aà,9á,9aá,9bà)], 9,12,15-Octadecatrienoic acid, 1-Methyl-4-phenyl-3,4-dihydroisoquinoline and ç-Sitosterol. The compounds identified in the GC-MS study are biologically important and can be utilized for various biological activities.

3.3. Characterization of the biosynthesized AgNPs

The characterization of the biosynthesized silver nanoparticles was previously performed, and the details were discussed in Ahmad *et al*. (2022). Briefly, the appearance of brown color was observed after mixing $AgNO_3$ with plant extract with direct exposure for about 15 min to sunlight. The color was turned to brown due silver ion reduction is a general characteristic of AgNPs synthesis. The UV-Visible spectrophotometric characterization recorded after 24 h and the highest absorption surface plasmon resonance peak at 440 nm. The reaction mixture was observed for many days however no significant change in the absorbance spectrum was observed after 48 h. The FTIR spectroscopy of the biosynthesized AgNPs identified various functional groups in the leaf extract and on the surface of silver nanoparticles. The detection of these groups may have resulted due to presence of plant secondary metabolites which may have led to formation of stable AgNPs. Size and shape morphology of the prepared silver nanoparticles was observed through different

magnification lenses using transmission electron microscopy (system JEOL JEM-101). XRD pattern analysis of the prepared silver nanoparticles was accomplished using system, JDX-3432, JEOL, Japan following Debye–Scherrer equation. Both TEM and XRD analysis showed the formation of crystalline nature particles having a size in the range of 8-34 nm.

Table 2. List of compounds detected during the GC-MS analysis of the leaf extract

S.No.	Area %	RT	Compound	Formula	Molecular weight
1	0.57	7.28	Cyclohexanecarboxylic acid, 2-hydroxy-, ethyl ester	$C_9H_{16}O_3$	172
2	0.96	8.62	Benzofuran, 2,3-dihydro-	C_8H_8O	120
3	0.76	8.97	1-Naphthalenol, 1,2,3,4-tetrahydro-, acetate	$C_{12}H_{14}O_2$	190
4	0.39	9.34	4-(4-Methoxy-6-methyl-5,6,7,8-tetrahydro- [1,3] dioxolo[4,5-g] isoquinolin-5-yl)-5-propyl-2,4-dihydro-pyrazol-3-one	$C_{18}H_{23}N_3O_4$	345
5	1.7	10.23	Hydroquinone	$C_6H_6O_2$	110
6	0.27	12.19	Azuleno[4,5-b] furan-2(3H)-one, 9a-[(acetyloxy)methyl] decahydro-6a,9-dihydroxy-6-methyl-3-methylene-, [3aS-(3aà,6á,6aà,9á,9aá,9bà)]-	$C_{17}H_{24}O_6$	324
7	0.23	12.47	4-(4-Methoxy-6-methyl-5,6,7,8-tetrahydro- [1,3] dioxolo[4,5-g] isoquinolin-5-yl)-5-propyl-2,4-dihydro-pyrazol-3-one	$C_{18}H_{23}N_3O_4$	345
8	2.64	13.25	Phenol, 2-(6-methyl-5,6-dihydrothiazolo[2,3-c] [1,2,4] triazol-3-yl)-	$C_{11}H_{11}N_3OS$	233
9	0.47	14.33	Propanoic acid	$C_{19}H_{26}O_6$	350
10	0.19	14.9	Phenmethylcynide, à, à-dimethyl-2-methoxy-6-nitro	$C_{11}H_{12}N_2O_3$	220
11	1.38	15.65	Cyclohexane, 1,4-dimethyl-2-octadecyl-	$C_{26}H_{52}$	364
12	1.75	16.2	Benzene, (1-pentylheptyl)-	$C_{18}H_{30}$	246
13	0.46	16.73	Hexanoic acid, 5-methylene-6-(5-methoxycarbonyl-1-cyclohexenyl)-, methyl ester	$C_{16}H_{24}O_4$	280
14	0.88	17.18	Benzene, (1-methylnonadecyl)-	$C_{26}H_{46}$	358
15	1.96	17.54	Pentacosane, 13-phenyl-	$C_{31}H_{56}$	428
16	0.13	18.24	Pregn-5-ene-3,8,11,12,14,20-hexol, (3á,11à,12á,14á)-	$C_{21}H_{34}O_6$	382
17	0.22	18.48	Cyclopropanebutanoic acid	$C_{25}H_{42}O_2$	374
18	6.06	18.97	n-Hexadecanoic acid	$C_{16}H_{32}O_2$	256
19	1.52	19.89	E-9-Methyl-8-tridecen-2-ol, acetate	$C_{16}H_{30}O_2$	254
20	10.61	20.23	Oleic Acid	$C_{18}H_{34}O_2$	282
21	0.73	21.25	4-Hexyl-1-(7-methoxycarbonylheptyl) bicyclo [4.4.0] deca-2,5,7-triene	$C_{25}H_{40}O_2$	372
22	0.89	21.54	1-Heptatriacotanol	$C_{37}H_{76}O$	536
23	1.2	22.39	9,12,15-Octadecatrienoic acid, 2,3-bis[(trimethylsilyl)oxy] propyl ester, (Z, Z, Z)-	$C_{27}H_{52}O_4Si_2$	496
24	3.44	23.21	Azuleno[4,5-b] furan-2(3H)-one	$C_{17}H_{24}O_6$	324
25	1.16	23.76	9,12,15-Octadecatrienoic acid	$C_{27}H_{52}O_4Si_2$	496
26	0.34	0.34	Acetamide, N-[2-(3-ethyl-1-methyl-9H-carbazol-2-yl) ethyl]-N-methyl	$C_{20}H_{24}N_2O$	308
27	2.77	24.59	1-Methyl-4-phenyl-3,4-dihydroisoquinoline	$C_{16}H_{15}N$	221
28	0.48	25	Isoquinoline	$C_{16}H_{23}NO_2$	261
29	0.21	25.24	Ethanol,2-(1,2,3,4-tetrahydro-6,7-dimethoxy-1-methylisoquinolin-2-yl)-1-(3-nitrophenyl)-	$C_{20}H_{24}N_2O_5$	372
30	55.64	26.16	ç-Sitosterol	$C_{29}H_{50}O$	414

3.4. Antibacterial bioassay of the synthesized AgNPs

Both AgNPs and AgNPs-PE (1000 µg/mL) intensely inhibited the cell growth of both *E. carotovora* and *R. solanacearum*, while PE exhibited significant inhibition. The control treatment in the experiment did not inhibit the cell growth of both *E. carotovora* and *R. solanacearum* (Figure 2).

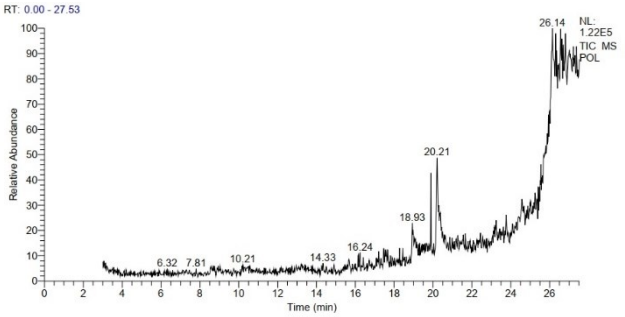


Figure 1. GC-MS analysis of the leaf extracr of P. geminiflorum

Observation regarding *E. carotovora* in microtitier plate experiment revealed 98 % inhibition by AgNPs-PE and 94 % by AgNPs (both at concentration of 1000 µg/mL). The PE (1000 µg/mL) treatment showed an optimum inhibition (58%) (Figure 2). The AgNPs-PE (1000 µg/mL) and AgNPs (1000 µg/mL) inhibited the growth of *R. solanacearum* by 97% and 95% respectively (Figure 2).

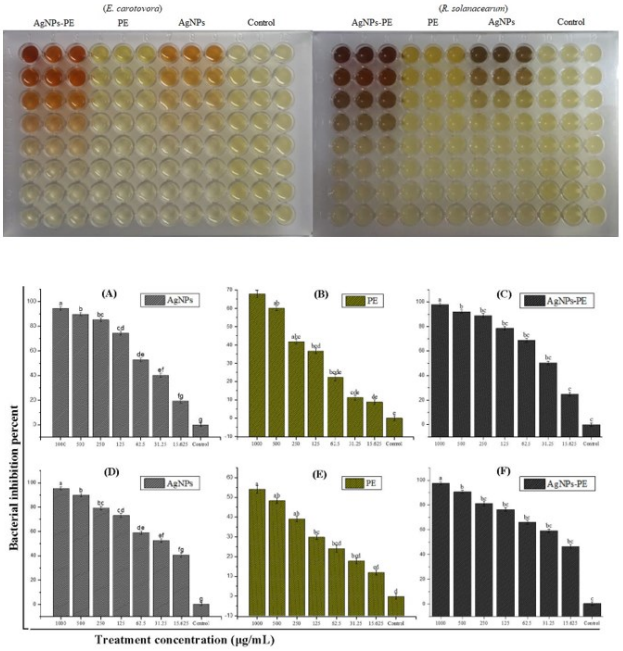


Figure 2. Growth Inhibition pattern of of E. carotovora (A to C) and R. solanacearum (D to F). Different letters are showing statistically significant differences at p< 0.05 for the same concentration

4. Discussion

Disease resistant capability of pathogenic microbes is becoming a challenging threat because of its diverse harmful effects (Peterson and Kaur 2018). These pathogens are causing serious diseases to important crops and declining their production (Nazarov *et al*. 2020). As a result of this major problem, the world is moving towards the food insecurity (Al-Ani and Furtado 2020). For instance, potato is one of the important widely grown vegetable crop which is badly affected by various phytopathogens (Wilson 2014; Oyesola *et al*. 2021). *E. carotovora* and *R. solanacearum* are the known pathogens causing soft rot and bacterial wilt in potato crop respectively (Tsror 1999; Ranjan *et al*. 2016). These pathogens directly attack on potato crop and lessening its production and nutrient capacity (Guchi 2015). To control these diseases several strategies have been developed including novel nano bio-control agents (Elmer and White 2018; Arif *et al*. 2022). Biosynthesized AgNPs are non-hazardous in nature and has been used as an alternative antimicrobial agent (Rosman *et al*. 2020). The plant secondary metabolites combine with silver metal to increase its antimicrobial effects (Marslin *et al*. 2018). Moreover, at low concentration AgNPs are more effective against hazardous pathogens and have no toxicity effects (Gurunathan *et al*. 2014).

In this study, the prepared leaf-extract-coated AgNPs was used as a stable antimicrobial drug against *E. carotovora* and *R. solanacearum*. Our findings regarding phytochemical indicated the presence of important secondary chemicals including flavonoids, tannins, glycosides and terpenoids. The GC-MS analysis of the extract revealed the existence of 30 plant important compounds which were important in respect of different biological properties. Among these compounds, the highest percent were detected for ç-Sitosterol which is a member of the phytosterols class and which is playing its role as an antioxidant and anticholesteremic drug (Zozio *et al*., 2014; Zhu *et al*., 2022). Further, oleic acid and n-Hexadecanoic acid were also detected in the leaf extract which is an important fatty acid and helps to combat free radical, strengthen the immune system and showed anti-inflammatory effects (Joel and Bhimba, 2010). Previously, AgNPs have been shown to exhibit antibacterial properties through rapid release of Ag^+ ions targeting depolarization of the plasma membrane and inhibition of respiratory and metabolic activities (Bondarenko *et al*. 2018). These compounds may be considered to be key agents for increasing the medicinal applications of the plant. Previously we have reported the silver nanoparticles from the studied plant and were found for the presence of bioactive phenolic compounds (Ahmad *et al*., 2022). Also, these compounds were found to increase the antimicrobial efficiency of the prepared nanoparticles (Kavaz *et al*., 2018; de Carvalho Bernardo *et al*. 2022). Thus, the present analysis showed 30 important secondary compounds that may also enhancing the antimicrobial application of the prepared nanoparticles. Our phytochemicals results were completely corresponding to the previous studies (Sonam *et al*. 2017; Kavaz *et al*., 2018; Fauzi *et al*. 2020; Lalam *et al*. 2020). Previously, researchers found stable AgNPs synthesis by plants because of the presence of important secondary constituents (Arif *et al*. 2022). Moreover, the leaf extract increases the antimicrobial potential of AgNPs and were effective against several pathogens (Anandalakshmi *et al*. 2016; Jinu *et al*. 2017). Therefore, plant-based biosynthesis of AgNPs is more effective because of the combined effects of Ag metal and plant extract (Anand *et al*. 2022; Dilbar *et al*., 2023b).

Results regarding *in vitro* antibacterial activities revealed that both AgNPs and AgNPs-PE in high concentration (1000 µg/mL) were promising which effectively inhibited the cell growth of both *R. solanacearum* and *E. carotocora* by exhibiting strong inhibitory effects against the tested pathogens. Plant extract alone effectively control the cell growth of the pathogens in comparison to control. Our results regarding antibacterial activity were completely in agreement with those previously reported (Wypij *et al*. 2021; Arif *et al*. 2022).

Previous studies showed the effectiveness of plant-based nanoparticles synthesis and its use to control the growth of plant pathogens (Jinu *et al*. 2017; Khan *et al*. 2022 Mohammadzadeh *et al*. 2022). Several plants have previously been used for the antimicrobial silver nanoparticles synthesis (Borase *et al*. 2014; Ahmad *et al*. 2016). Therefore, due to its high medicinal importance we have use *P. geminiflorum* as source agent for the efficient synthesis of silver nanoparticles. Further, silver nanoparticles were effectively synthesized with potential of controlling the cell growth of potato's pathogens.

5. Conclusion

In this study, we used the leaf extract of *P. geminiflorum* for phytochemical screening and antimicrobial silver nanoparticles synthesis. The phytochemical screening revealed the presence of 30 important secondary chemicals in the leaf extract. The biosynthesis process resulted with significant synthesis of silver nanoparticles. Moreover, the antimicrobial activity of AgNPs efficiently controls the growth of two pathogens of potato crop during *in vitro* experiment. Therefore, the result of *in vitro* experiment needs to be improved further for in planta antimicrobial applications. Similarly, the management of abiotic stresses is also one of the main concerns because of its severe effects on the plant growth. These stresses declining the production rate of important crops and leading to food insecurity worldwide. Further *in vivo* studies need to be conducted to investigate the mode of action and effects of synthesized nanoparticles on plant growth for better management of biotic and abiotic stresses.

Conflict of interest

We declare no conflict of interest.

Data availability statement

The generated data is present in the manuscript.

Acknowledgment

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