

## Mycosynthesizing and characterizing silver nanoparticles from the mushroom *Inonotus hispidus* (Hymenochaetaceae), and their antibacterial and antifungal activities

Ahmed Saadoon Jaloot<sup>a</sup>, Mustafa Nadhim Owaid<sup>a,b,\*</sup>, Ghassan Adnan Naem<sup>c</sup>, Rasim Farraj Muslim<sup>a</sup>

<sup>a</sup> Department of Environmental Sciences, College of Applied Sciences-Hit, University Of Anbar, Hit, Anbar 31007, Iraq

<sup>b</sup> Department of Heet Education, General Directorate of Education in Anbar, Ministry of Education, Hit, Anbar 31007, Iraq

<sup>c</sup> Department of Biophysics, College of Applied Sciences-Hit, University Of Anbar, Hit, Anbar 31007, Iraq

### ARTICLE INFO

#### Keywords:

AgNPs  
Antimicrobial features  
Green synthesis  
Macrofungi  
Nano drugs

### ABSTRACT

The current research aims to produce silver nanoparticles from isolated *Inonotus hispidus* (Shaggy Bracket fungus) locally from Hit, Iraq. The silver nanoparticles were mycosynthesized from the mushroom extract by adding  $10^{-3}$  M AgNO<sub>3</sub>. The properties of Ag nanoparticles were studied by UV–vis, XRD, FTIR, AFM, and Zetasizer. The color changed from light yellow to brown after 90 min, and the lambda max was at 410–420 nm by the UV–vis spectrum. Zetasizer showed the formation of AgNPs with an average of 69.24 nm. Results of the XRD show that AgNPs have a cube-centered crystalline face. The FTIR showed that active groups belonging to the carbonyl group (C=O) in the form of amides (HN–C=O) in peptides and proteins, as well as the presence of this group in flavonoids, polysaccharides and phenolic compounds. The concentrations 75% and 100% of AgNPs exhibited zone of inhibition 10 mm and 11 mm, respectively against *Escherichia coli*. The lowest zone of inhibition was 7.5 mm toward *Klebsiella* with the concentration of 50%. Also, the higher inhibition percentage against fungi was 12.2% against *Aspergillus niger*, whereas the lower percentage was 6.3% against *Aspergillus flavu*. This paper is considered the first attempt to synthesize AgNPs from *Inonotus hispidus*, which applied against bacteria and fungi.

### 1. Introduction

The formation of green eco-nanoparticles from fungi is significant because it is eco-friendly and non-toxic due to its production from myco-organic materials harmless to nature and the environment (Owaid, 2020). This approach is called myco-nanotechnology, which is used in medicine, industry, and agriculture (Owaid et al., 2019; Owaid and Ibraheem, 2017). Numerous studies on the synthesis of nanoparticles using chemical approaches have indicated using chemicals as reducing agents, and this has toxic effects toward human health when it uses in medicine in addition to the increase of pollutants in nature (Noruzi et al., 2011; Shang et al., 2013). Recently, biologically synthesis of NPs, as an eco-method, is used to apply biomaterials as green reducer agents instead of chemicals to decrease the toxicity (Virkutyte and Varma, 2011) such as extracts of parts of the plant (Al-Bahrani et al., 2018; Noruzi, 2015; Owaid et al., 2019), filaments of algae (Kathiraven et al., 2015), the mycelium and fruiting body of

mushrooms (Owaid, 2019a; Owaid et al., 2015; Rabeea et al., 2020; Dheyab et al., 2020) and truffles (Owaid et al., 2018).

However, using edible and medicinal mushrooms to mycosynthesize nanoparticles is significant because of huge amounts of the produced mycelia, fruiting bodies, and secondary metabolites (Owaid and Ibraheem, 2017). Recently, several works have achieved about the mycosynthesized AgNPs using mushrooms such as *Agaricus bisporus* (Owaid et al., 2020), and *Pleurotus* species, which have activity in biomedicine against pathogenic bacteria, fungi, yeasts, and tumors, in addition to degrading Azo dyes industrially (Owaid, 2019a).

*Inonotus hispidus* is called Shaggy Bracket fungus as a common name that belongs to Hymenochaetaceae family and grows on trees in Germany (Ali et al., 2003), Macedonia (Angelini et al., 2019), and Turkey (Brito et al., 2019). Recent studies reported that the content of polyphenolic compounds, hispolon, hispidin (Suay et al., 2000), *N*-butylbenzenesulfonamide, lauramidopropyl betaine, uplandicine, and 3,5-di-*tert*-butyl-4-hydroxybenzaldehyde compounds (Angelini et al., 2019)

\* Corresponding author at: Department of Environmental Sciences, College of Applied Sciences-Hit, University Of Anbar, Hit, Anbar 31007, Iraq.  
E-mail address: [mustafanowaid@uoanbar.edu.iq](mailto:mustafanowaid@uoanbar.edu.iq) (M.N. Owaid).

obtained from *I. hispidus* could inhibit fungal filaments. Also, this mushrooms recorded therapeutic benefits including antiviral (Ali et al., 2003), anticancer (Zan et al., 2011), antioxidant (Angelini et al., 2019; Zan et al., 2011), antibacterial, anti-candidal and antifungal (Angelini et al., 2019; Pala et al., 2019), antiproliferative activities (Yang et al., 2019) and immunomodulatory effects (Gröndemann et al., 2016).

However, oppositely, only two papers have published about using another species (*I. obliquus*) for mycosynthesizing silver NPs (Nagajyothi et al., 2014) and gold NPs (Lee et al., 2015) which exhibited antibacterial and anticancer activities respectively. Hence, this study is considered the first attempt for synthesizing AgNPs using fresh fruiting bodies of *Inonotus hispidus*, which characterized using UV-vis, FTIR, AFM, ZetaSizer, XRD and tested *in vitro* against different pathogenic fungi and bacteria.

## 2. Materials and methods

### 2.1. Microbial isolates

Three bacterial species, namely *Staphylococcus aureus*, *E. coli*, and *Klebsiella* sp., were obtained from the General Hospital of Heet, Iraq. One yeast *Geotricum* sp. and three fungi namely *Pythium* sp., *Aspergillus flavus*, and *Aspergillus niger* were obtained from the College of Science at University of Baghdad, Iraq.

### 2.2. Mushroom samples

The fresh fruiting bodies of Shaggy Bracket fungus (*I. hispidus*), (Hymenochaetaceae family) were collected from the trunk of live tree *Tamarix articulata* (Tamaricaceae family) from Hit city, Iraq and identified to the species level, according to Buchanan and Ryvarden (1988) and Pala et al. (2011). This mushroom is called Shaggy Bracket fungus as a common name. The samples were cleaned and used in the following tests to prepare silver nanoparticles (AgNPs).

### 2.3. Extraction of the natural compounds in *Inonotus hispidus*

The fresh fruiting bodies of *I. hispidus* were extracted in D.W. (distilled water) Only 40 g of this mushroom were cut and extracted in 200 mL D.W and heated on the hotplate magnetic stirrer for 15 min until boiling for 10 min. Then, the watery extract was filtered using gauze, refiltered by Whatman filter paper No. 1, and centrifuged using a centrifuge at 4000 rpm. The aqueous extract was stored at 4 °C until use to form AgNPs (Owaid et al., 2019).

### 2.4. Mycosynthesizing silver nanoparticles

To synthesizing Ag nanoparticles, 30 mL of the *I. hispidus* extract was mixed with 100 mL of 1 mM silver nitrate (AgNO<sub>3</sub>, purchased from AFCO for Metal, China, purity of 99.9%) and heated at 60 °C for 60 min until changing the mixture color from colorless to bright brown color, which is evidence to form silver NPs in the solution.

### 2.5. Characterization of mycosynthesized Ag nanoparticles

The characteristics of the mycosynthesized silver nanoparticles (AgNPs) were studied using the change in color, UV-vis spectrum, FTIR, XRD, AFM, and Zetasizer. The average grain size of AgNPs was determined by the X-ray diffraction peak using the equation of Debye-Scherrer (Abdelrahim et al., 2017):

$$D = k\lambda/\beta \cos \theta \quad (1)$$

### 2.6. Antimicrobial efficacy

The antimicrobial efficacy of the mycosynthesized Ag nanoparticles from *I. hispidus* against bacteria (three species) and yeast (one species) was achieved using the well diffusion method on plates of Muller Hinton Agar. Three concentrations of AgNPs 50%, 75%, and 100% were separately introduced into the bore wells on the agar using sterile dropping pipette. Three concentrations of the mushroom extract alone were tested as a control in this study. The inoculated plates were kept at 4 °C for 30 min to let for the diffusion of solutions of the extract and AgNPs in plates before incubation at 37 °C for 18 h. The zone of inhibition in the incubated plates was measured by a ruler (Owaid et al., 2018).

### 2.7. Antifungal activity

The antifungal efficacy of the AgNPs and the extract of mushroom were determined using the well diffusion method, as mentioned by Owaid et al. (Owaid et al., 2017) with some modifications. Three concentrations of AgNPs (50%, 75%, and 100%) and three concentrations of the extract (50%, 75%, and 100%) were placed in the plate of Potato Dextrose Agar and then left 30 min to spread solutions in the medium. They were inoculated by the disc of old seven-day pathogenic fungi centrally, incubated at 28 °C to enable monitoring of their development; then the diameter of mycelial growth was measured. The three fungi isolates were inoculated without the mushroom extract and AgNPs in the medium as control. The inhibition percentage was calculated by Eq. (2) (Owaid, 2017):

$$\text{Inhibition (\%)} = [(M_1 - M_2)/M_1] \times 100 \quad (2)$$

where  $M_1$ : the diameter of the control plate,  $M_2$ : the diameter of the treatment plate.

### 2.8. Statistical analysis

The design of this work was applied with triplicates using SAS software. The effect of two factors, including the mushroom extract and its AgNPs were subjected at  $p < 0.01$ . The statistical analysis was completed by one-way analysis of variance using CRD (Completely Randomized Design).

## 3. Results and discussion

The optical vision of the formation of silver nanoparticles appears in Fig. 1. The first unmistakable sign to the mycosynthesis of AgNPs is changing of color of the mixture from colorless to yellowish-brown as shown in Fig. 1. The change of color was seen after 15 min of starting the reaction at 60 °C on the magnetic stirrer hot plate. The mycosynthesized AgNPs from the *I. hispidus* extract exhibited an absorbance reached 0.499 a.u at lambda max of 360 nm then the absorbance increased with increasing time of interaction. After 30 min, the absorbance recorded 0.630 a.u. at a wavelength of 380 nm. The color of reaction became darker with the time and the absorbance increased to 0.919 a.u. and 1.131 a.u. at wavelength of 410 nm after 45 min and 60 min respectively, because of excitation of surface plasmon resonance (SPR) in colloidal AgNPs (Shankar et al., 2003), and that was confirmed using UV-vis spectra as in Fig. 1. These results agree with the results many recent studies which tested the natural myco-materials from mushrooms in the formation of AgNPs (Al-Bahrani et al., 2017; Owaid et al., 2020, 2015).

Fig. 2 illustrates two images of atomic force microscope (AFM) including the lateral (2D) and three-dimensional (3D) images that exhibit surface roughness of the mycosynthesized AgNPs at size images 1922 nm × 1626 nm. Surface roughness analysis exhibits various parameters like amplitude, hybrid and functional parameters.

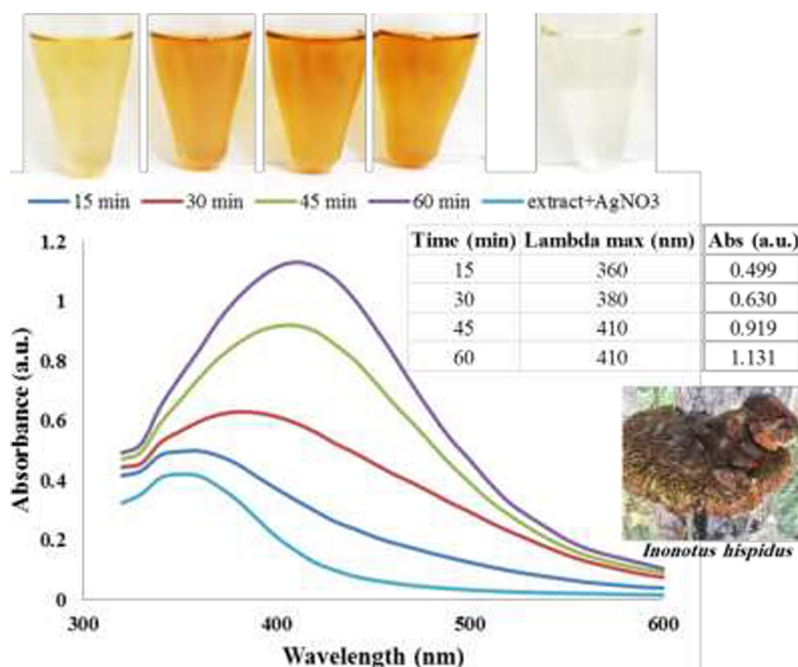


Fig. 1. Visual and UV-vis analyses of the synthesized AgNPs from *I. hispidus*.

Amplitude parameters have measured such as roughness average, root mean square, and ten-point height reach to 3.05 nm, 3.58 nm and 15.8 nm respectively. Hybrid parameters have measured such as root mean square slope, and mean summit curvature which reach to  $0.27 \text{ nm}^{-1}$ ,  $-0.03 \text{ nm}^{-1}$  while surface area ratio records 3.36. Functional parameters have measured like reduced summit height, core roughness depth, and reduced valley depth records 1.3 nm, 11.6 nm, and 1.52 nm, respectively. This is an indicator to form silver nanoparticles in a small size reaches to 85.00 nm in around  $\leq 90\%$  of its NPs. The average of the mycosynthesized silver nanoparticles is 69.24 nm in this study as in Fig. 3 which shows histogram of granularity Cumulation Distribution (Zetasizer) of AgNPs. On the other hand, the smallest size is 55.00 nm with a lower Cumulation reaches to 15.46% while the nanoparticles have 125 nm as a bigger size shows Cumulation of 100%. These findings agree with the result of (Owaid et al., 2020) especially for Cumulation of silver nanoparticles (Fig. 3).

Peaks of XRD as in Fig. 4 located at  $2\theta$  of  $46.72^\circ$  could be due to the (2 0 0) crystallographic plane of the face-centered cubic silver crystals which agree with values of (Reference Code 01-089-3697) (Abod et al., 2017; Ghareib et al., 2016). The peak of (2 0 0) has the highest intensity, which indicates that (2 0 0) is the preferred orientation. The

present result exhibited that the mean crystallite size of AgNPs is 45.23 nm. The pattern of XRD illustrated that the formed AgNP in the present study was crystalline.

Fig. 5 exhibits FTIR spectra of extracts of the mushroom (Fig. 5A), and biosynthesized Ag nanoparticles (Fig. 5B). This analysis is useful for determining the functional groups in the myco-materials and for expecting the composition of chemical materials in this fungal extract which bonded or coated the silver nanoparticle. The absorption band at  $1048 \text{ cm}^{-1}$  is due to the single bond (C–C) in groups of methylene ( $-\text{CH}_2$ ) and the methyl groups ( $-\text{CH}_3$ ) (Nakamoto, 2009; Silverstein et al., 2005) which found in the composition of amino acids, peptides, proteins, solid fatty acids and fats. The presence of the band  $3242 \text{ cm}^{-1}$  declares the presence of stretched vibration of the group  $-\text{OH}$  in the mushroom extract thus the existence of phenolic compounds as the Hispidin compound (*E*)-6-(3,4-dihydroxystyryl)-4-hydroxy-2H-pyran-2-one, (3*Z*,5*E*)-6-(3,4-dihydroxyphenyl)-4-hydroxyhexa-3,5-dien-2-one (Nasser et al., 1996), (*E*)-7-(3,4-dihydroxystyryl)-3-hydroxy-3-methyl-3,4-dihydrooxepine-2,5-dione and (*E*)-4-(3,4-dihydroxyphenyl)but-3-en-2-one (Zan et al., 2011) as in Fig. 6. This absorption may also be due to the phenolic compound: (4*S*,5*S*)-4-hydroxy-3,5-dimethoxycyclohex-2-enone (Yang et al., 2019).

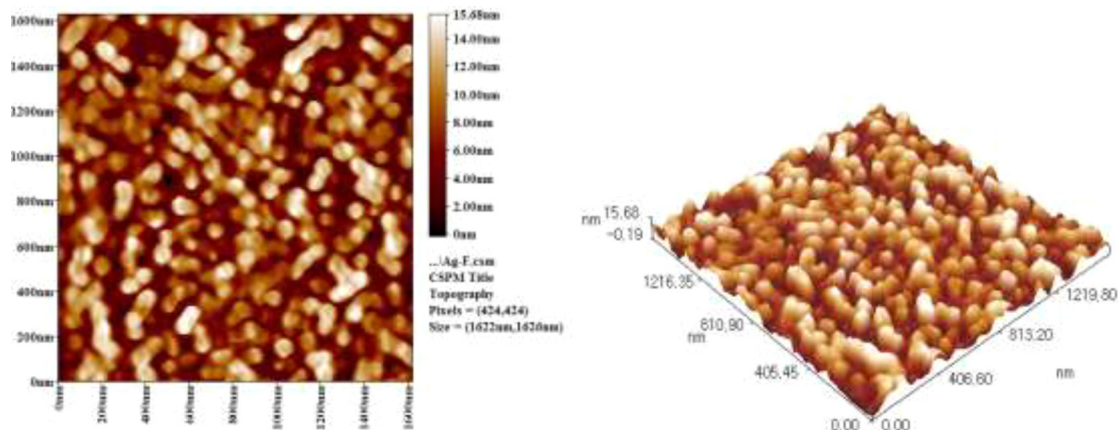


Fig. 2. AFM of AgNPs, lateral (2D) and three dimensional (3D).

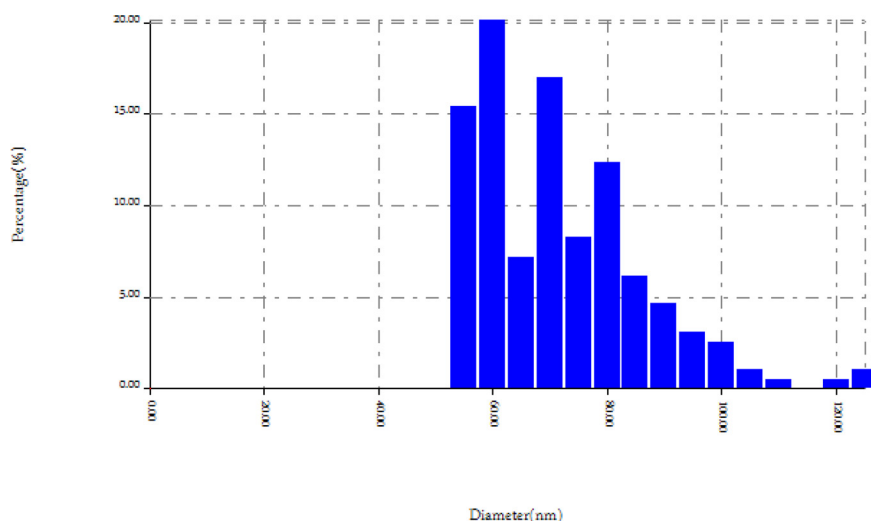


Fig. 3. Histogram of granularity cumulation distribution of AgNPs.

The broad absorption band confined between 2400 and 3600  $\text{cm}^{-1}$  indicates the existence of the group of carboxyl ( $-\text{COOH}$ ) in acidic compounds such as Hispidin (Fig. 6) and carboxyfluorescein (Gründemann et al., 2016). This group also belongs to the saturated fatty acids found in the extract, especially palmitic acid and arachidic acid as well as unsaturated fatty acids (linoleic acid and oleic acid) (Olennikov et al., 2014). Moreover, the band at 1559  $\text{cm}^{-1}$  may be due to the  $\text{C}=\text{N}$  groups in the harmaline and harmine compounds (Benarous et al., 2015). The carbonyl group appeared at 1669  $\text{cm}^{-1}$  due to the interference and the extract components appeared with a small band.

As shown in Fig. 5B, FTIR of the biosynthesized AgNPs from the mushroom, it shows that bands more clearly and the nanoparticles of silver atoms are formed. The clear results of the bands in better form prove that and the small particles sizes leads to increase the surface area exposed to the infrared. That leads to a better arrangement for the functional groups inside the empty orbitals of the silver atoms as well as the possibility of these functional groups in electron-rich AgNPs to participate electrons with empty orbitals in the outer shell of silver. The carbonyl group appeared at 1671  $\text{cm}^{-1}$  due to the interference with the Ag nanoparticles band which appeared with a small band. The band of 1647  $\text{cm}^{-1}$  supports the existence of Ag nanoparticles in this study (Owaid et al., 2020). Thus, we can expect the arrangement of the

organic compounds of *I. hispidus* like Hispidin as a phenolic compound around Ag atoms as in Fig. 7.

Table 1 exhibited the zone of inhibition of microbes in Muller-Hinton Agar. Three concentrations of biosynthesized AgNPs (50%, 75% and 100%) were investigated toward three pathogenic bacteria (*S. aureus*, *E. coli*, and *Klebsiella* sp.) and one yeast isolate (*Geotricum* sp.). The mushroom extract had not any positive influence against all pathogens because of the low concentration of extract, while *S. aureus* and *Geotricum* sp. were resistance for AgNPs. Furthermore, *E. coli*, and *Klebsiella* sp. showed sensitivity which inhibited by the synthesized AgNPs. The higher zone of inhibition was recorded 11 mm against *E. coli* by the concentration 100% followed 10 mm and 8.5 mm by the concentrations 75% and 50%, respectively. However, the concentration of 100% of AgNPs recorded a zone of inhibition reached 8 mm toward *Klebsiella* sp. In comparison, the concentration of 75% of AgNPs recorded the lowest zone of inhibition (7.5 mm) against the previous bacteria.

On the other hand, the sensitivity of 10% both the extract of mushroom and the mycosynthesized AgNPs separately was achieved in Potato Dextrose Agar. The sensitivity was investigated against three pathogenic fungi, including *Pythium* sp., *Aspergillus niger*, and *Aspergillus flavus*. The mushroom extract had not any influence against all fungal pathogens as in Fig. 8. Also, AgNPs did not show any inhibition against

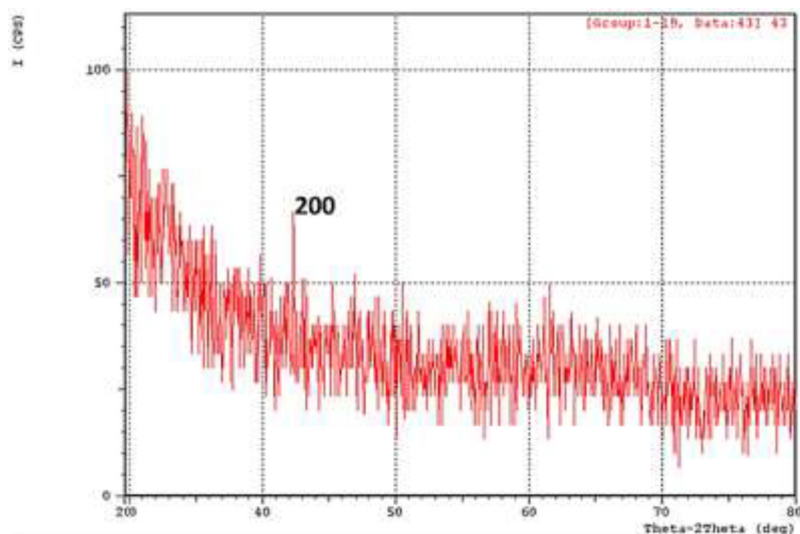


Fig. 4. XRD of the prepared AgNPs.



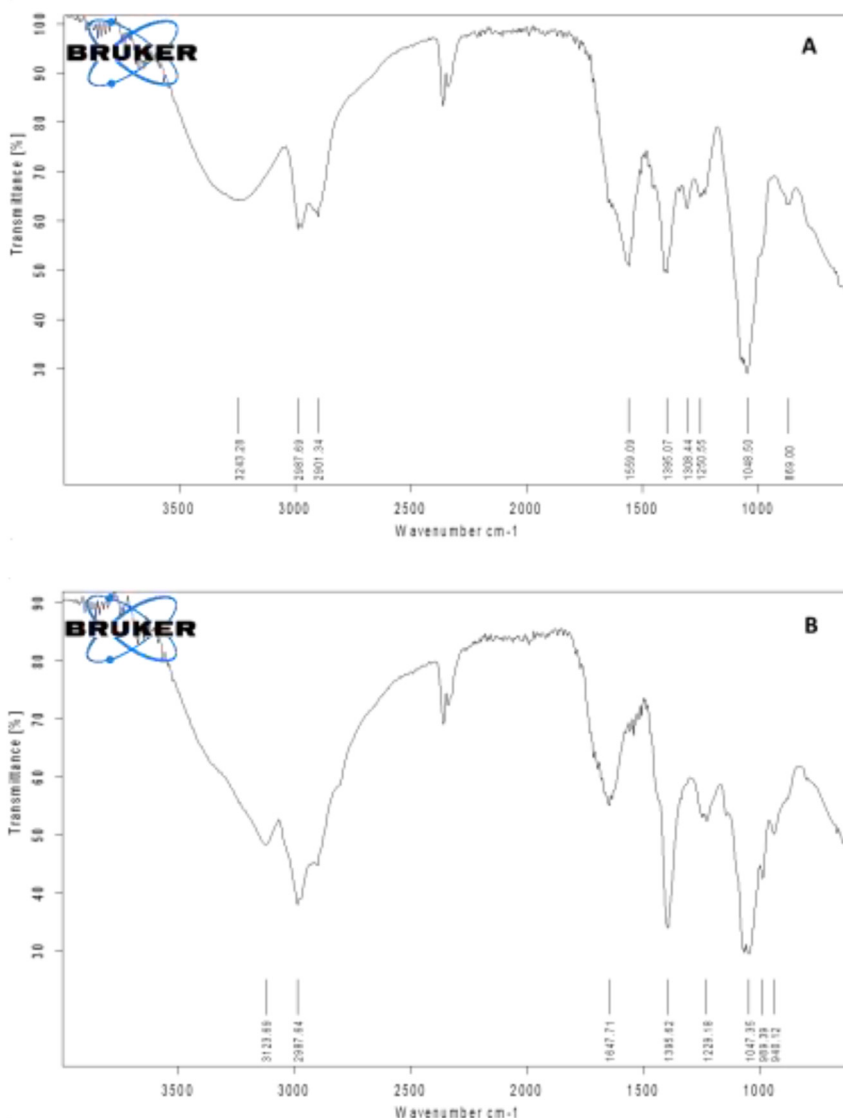


Fig. 5. FTIR spectra of the mushroom extract (A) and the mycosynthesized Ag nanoparticles (B).

the fungus *Pythium* sp. However, the higher inhibition percentage was 12.2% against *A. niger*, whereas the lower inhibition percentage was 6.3% against *A. flavus*.

The role of the mycosynthesized silver nanoparticles is remarkable and potent against human pathogenic bacteria and fungi compared with the mushroom extract alone. The large surface area of the

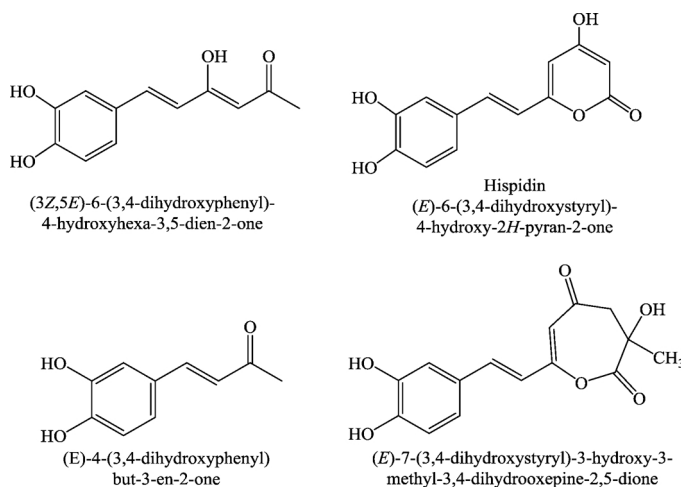


Fig. 6. Structures of the expected phenolic compounds in the extract of *I. hispidus* mushroom.

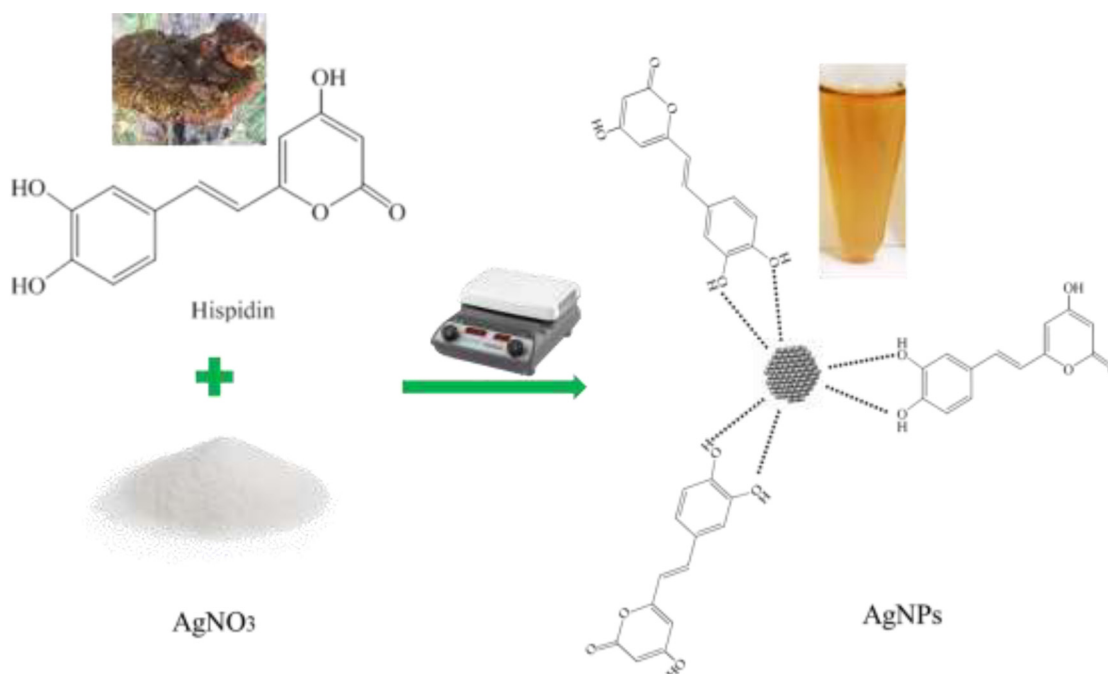


Fig. 7. The schematic diagram of the formation of AgNPs and the expected arrangement of organic compounds around silver atoms.

Table 1

The zone of inhibition of microbes by the mycosynthesized AgNPs (mm).

Pathogenic microbes	The mycosynthesized AgNPs		
	50%	75%	100%
<i>Staphylococcus aureus</i>	0	0	0
<i>Escherichia coli</i>	8.5	10	11
<i>Klebsiella sp.</i>	0	7.5	8
<i>Geotricum sp.</i>	0	0	0

mycosynthesized AgNP provides more surface contact with the microbe (Logeswari et al., 2015) and makes particular interactions with its cell membrane (Morones et al., 2005). The Ag nanoparticles contact and adhere to the cellular wall and membrane of microbes. Thus some of the ions of silver release and adhere (Brunner et al., 2006) to the sulfur-containing proteins on the cell membrane (Klasen, 2000). These interactions at the membrane cause several morphological and structural changes in the cell wall like pores and pits formation. Through these pores, some of the cellular components release into the extracellular fluid because of osmotic differences. Whereas the AgNP pass to the

inside of the microbe and interacts with RNA and DNA thus they will inhibit the replication of proteins in it which leads to the death of the microbe (Brunner et al., 2006; Feng et al., 2000).

Moreover, these interactions have been bonded to the suppression of enzymes (Yamanaka et al., 2005), and stopped the protein translation (Velusamy et al., 2016) that relate with the capability of producing ATP in microbes. Also, the affinity of the AgNP to the thiol group of proteins are causing the degradation of proteins and unfolding the protein chain (Soliman et al., 2018). Nevertheless, AgNPs have ability firstly to induce new several intracellular components like proteins as stress responses (Markowska et al., 2014), which released during membrane disruption after exposure to AgNPs in *Candida albicans*.

The AgNPs had an important impact against pathogenic fungi such as *Colletotrichum sp.* and *Bipolaris sorokiniana* (Lamsal et al., 2011). Moreover, AgNPs decrease the mycotoxin production of *A. niger* and *A. flavus* and reduce mold cytotoxicity. Also, AgNPs effect on the organic acid production of *A. niger* (Pietrzak et al., 2016). The reason for this state is probably due to the high intensity at which the solution can saturate and adhere to the hypha of fungi and control on the fungal diseases (Lamsal et al., 2011). Also, the toxicity of AgNPs may be due to the direct interaction and disruption of bio-macromolecules or

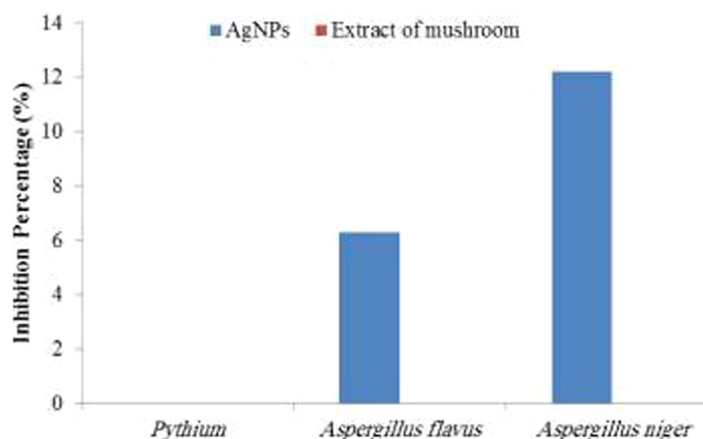


Fig. 8. Inhibition percentages of pathogenic fungi by the 10% of AgNPs colloid and the extract of mushroom.

production of reactive oxygen species (ROS) (Brunner et al., 2006). Many researchers reported that the mycosynthesized AgNPs had antibacterial and antifungal activities (Owaid, 2019a). The results of this study agree with the finding of (Owaid, 2019b).

#### 4. Conclusion

The work aims to mycosynthesize silver nanoparticles from isolated *Inonotus hispidus* (Shaggy Bracket fungus). The color changed from light yellow to brown after 90 min, and the lambda max was at 410–420 nm by the UV–vis spectrum. Zetasizer showed the formation of AgNPs with an average of 69.24 nm. Results of the XRD show that AgNPs have a cube-centered crystalline face. The FTIR showed that active groups belonging to the carbonyl group (C=O) in the form of amides (HN–C=O) in peptides and proteins, as well as the presence of this group in flavonoids, polysaccharides and phenolic compounds. The concentration of 100% of AgNPs exhibited zone of inhibition 11 mm against *E. coli*. While the higher inhibition percentage against fungi was 12.2% against *Aspergillus niger*. This paper is considered the first attempt to synthesize AgNPs from *I. hispidus*, which applied against bacteria and fungi.

#### Authorship statement

M.N. Owaid, A.S. Jaloot: substantial contribution to conception and design.

M.N. Owaid, A.S. Jaloot: substantial contribution to acquisition of data.

R.F. Muslim, G.A. Naeem: substantial contribution to analysis and interpretation of data.

M.N. Owaid, R.F. Muslim: drafting the article.

R.F. Muslim: critically revising the article for important intellectual content.

M.N. Owaid: final approval of the version to be published.

#### Declaration of Competing Interest

None.

#### Acknowledgments

The authors thank Department of Environmental Sciences, College of Applied Sciences-Hit at University Of Anbar for achieving this Research Project No. 5 on 15 Oct 2018 and the UOA Bridging Grant No. 5/15.10.2018 for funding this study.

#### References

- Abdelrahim, K., Younis, S., Mohamed, A., Salmeen, K., Mustafa, A.E.M.A., Moussa, S., 2017. Extracellular biosynthesis of silver nanoparticles using *Rhizopus stolonifer*. Saudi J. Biol. Sci. 24, 208–216. <https://doi.org/10.1016/j.sjbs.2016.02.025>.
- Abod, H.A., Bander, I., Zain-Al-Abddein, S.S., 2017. The effect of silver nanoparticles prepared using *Aspergillus niger* in some pathogenic bacteria *Aspergillus niger*. Kirkuk Univ J /Scientific Stud 12, 1–16.
- Al-Bahrani, R., Raman, J., Lakshmanan, H., Hassan, A.A., Sabaratnam, V., 2017. Green synthesis of silver nanoparticles using tree oyster mushroom *Pleurotus ostreatus* and its inhibitory activity against pathogenic bacteria. Mater. Lett. 186, 21–25. <https://doi.org/10.1016/j.matlet.2016.09.069>.
- Al-Bahrani, R.M., Muayad, S., Majeed, A., Owaid, M.N., 2018. Phyto-fabrication, characteristics and anti-candidal effects of silver nanoparticles from leaves of *Ziziphus mauritiana* Lam. Acta Pharm. Sci. 56, 85–92. <https://doi.org/10.23893/1307-2080.APS.05620>.
- Ali, N.A.A., Mothana, R.A.A., Lesnau, A., Pilgrim, H., Lindequist, U., 2003. Antiviral activity of *Inonotus hispidus*. Fitoterapia 74, 483–485. [https://doi.org/10.1016/S0367-326X\(03\)00119-9](https://doi.org/10.1016/S0367-326X(03)00119-9).
- Angelini, P., Girometta, C., Tirillini, B., Moretti, S., Covino, S., Cipriani, M., D'Ellena, E., Angeles, G., Federici, E., Savino, E., Cruciani, G., Venanzoni, R., 2019. A comparative study of the antimicrobial and antioxidant activities of *Inonotus hispidus* fruit and their mycelia extracts. Int. J. Food Prop. 22, 768–783. <https://doi.org/10.1080/10942912.2019.1609497>.
- Benarous, K., Bombarda, I., Iriepa, I., Moraleda, I., Gaetan, H., Linani, A., Tahri, D., Sebba,

- M., Youfi, M., 2015. Harmaline and hispidin from *Peganum harmala* and *Inonotus hispidus* with binding affinity to *Candida rugosa* lipase: in silico and in vitro studies. Bioorg. Chem. 62, 1–7. <https://doi.org/10.1016/j.bioorg.2015.06.005>.
- Brito, I., Antunes, P.M., Carvalho, M., De Tuinen, D., Van Goss, M.J., 2019. Investigation of polyphenol composition, biological activities and detoxification properties of some medicinal mushrooms from Turkey. Turk. J. Pharm. Sci. 16, 155–160. <https://doi.org/10.4193/Rhino13.217>.
- Brunner, T.J., Wick, P., Manser, P., Spohn, P., Grass, R.N., Limbach, L.K., Bruinink, A., Stark, W.J., 2006. In vitro cytotoxicity of oxide nanoparticles: comparison to asbestos, silica, and the effect of particle solubility. Environ. Sci. Technol. 40, 4374–4381. <https://doi.org/10.1021/es052069i>.
- Buchanan, P., Ryvarden, L., 1988. Type studies in the Polyporaceae 18. Species described by G.H. Cunningham. Mycotaxon 31, 1–38.
- Dheyab, M.A., Owaid, M.N., Rabeea, M.A., Abdul Aziz, A., Jameel, M.S., 2020. Mycosynthesis of gold nanoparticles by the Portabella mushroom extract, Agaricaceae, and their efficacy for decolorization of Azo dye. Environ. Nanotechnol. Monit. Manag. 100312. <https://doi.org/10.1016/j.enmm.2020.100312>.
- Feng, Q.L., Wu, J., Chen, G.Q., Cui, F.Z., Kim, T.N., Kim, J.O., 2000. A mechanistic study of the antibacterial effect of silver ions on *Escherichia coli* and *Staphylococcus aureus*. J. Biomed. Mater. Res. 52, 662–668. [https://doi.org/10.1002/1097-4636\(20001215\)52:4<662::AID-JBM10>3.0.CO;2-3](https://doi.org/10.1002/1097-4636(20001215)52:4<662::AID-JBM10>3.0.CO;2-3).
- Ghareib, M., Abu, M., Mostafa, M., Abdallah, W.E., 2016. Rapid extracellular biosynthesis of silver nanoparticles by *Cunninghamella phaeospora* culture supernatant. Iran. J. Pharm. Res. 15, 915–924.
- Gröndemann, C., Arnhold, M., Meier, S., Bäcker, C., Garcia-Käuffer, M., Grunewald, F., Steinborn, C., Klemd, A.M., Wille, R., Huber, R., Lindequist, U., 2016. Effects of *Inonotus hispidus* extracts and compounds on human immunocompetent cells. Planta Med. 82, 1359–1367. <https://doi.org/10.1055/s-0042-111693>.
- Gröndemann, C., Arnhold, M., Meier, S., Bäcker, C., Garcia-Käuffer, M., Grunewald, F., Steinborn, C., Huber, R., Lindequist, U., 2016. Influence of *Inonotus hispidus* on function of human immune cells. Eur. J. Integr. Med. 8, 54. <https://doi.org/10.1016/j.eujim.2016.08.128>.
- Kathiraven, T., Sundaramanickam, A., Shanmugam, N., Balasubramanian, T., 2015. Green synthesis of silver nanoparticles using marine algae *Caulerpa racemosa* and their antibacterial activity against some human pathogens. Appl. Nanosci. 5, 499–504. <https://doi.org/10.1007/s13204-014-0341-2>.
- Klasen, H.J., 2000. Historical review of the use of silver in the treatment of burns. I. Early uses. Burns 26, 117–130.
- Lamsal, K., Kim, S.W., Jung, J.H., Kim, Y.S., Kim, K.S., Lee, Y.S., 2011. Application of silver nanoparticles for the control of *Colletotrichum* species in vitro and pepper anthracnose disease in field. Mycobiology 39, 194–199. <https://doi.org/10.5941/MYCO.2011.39.3.194>.
- Lee, K.D., Nagajyothi, P.C., Sreekanth, T.V.M., Park, S., 2015. Eco-friendly synthesis of gold nanoparticles (AuNPs) using *Inonotus obliquus* and their antibacterial, antioxidant and cytotoxic activities. J. Ind. Eng. Chem. 26, 67–72. <https://doi.org/10.1016/j.jiec.2014.11.016>.
- Logeswari, P., Silambarasan, S., Abraham, J., 2015. Synthesis of silver nanoparticles using plants extract and analysis of their antimicrobial property. J. Saudi Chem. Soc. 19, 311–317. <https://doi.org/10.1016/j.jscs.2012.04.007>.
- Markowska, K., Grudniak, A., Krawczyk, K., Wrobeland, I., Kl, W., 2014. Modulation of antibiotic resistance and induction of a stress response in *Pseudomonas aeruginosa* by silver nanoparticles. J. Med. Microbiol. 63, 849–854.
- Morones, J., Elechiguerra, J., Camacho, A., Holt, K., Kouri, J., Ramirez, J., Yacaman, M., 2005. The bactericidal effect of silver nanoparticles. Nanotechnology 16, 2346–2353.
- Nagajyothi, P.C., Sreekanth, T.V.M., Lee II, J., Lee, K.D., 2014. Mycosynthesis: antibacterial, antioxidant and antiproliferative activities of silver nanoparticles synthesized from *Inonotus obliquus* (Chaga mushroom) extract. J. Photochem. Photobiol. B, Biol. 130, 299–304. <https://doi.org/10.1016/j.jphotochem.2013.11.022>.
- Nakamoto, K., 2009. Infrared and Raman Spectra of Inorganic and Coordination Compounds Part A: Theory and Applications in Inorganic Chemistry, 6th ed. A John Wiley and Sons, Inc.
- Nasser, A.A., Jansen, A.R., Pilgrim, H., Liberra, K., Lindequist, U., 1996. Hispolon, a yellow pigment from *Inonotus hispidus*. Phytochemistry 41, 927–929. [https://doi.org/10.1016/0031-9422\(95\)00717-2](https://doi.org/10.1016/0031-9422(95)00717-2).
- Noruzi, M., 2015. Biosynthesis of gold nanoparticles using plant extracts. Bioprocess Biosyst. Eng. 38, 1–14.
- Noruzi, M., Zare, D., Khoshnevisan, K., Davoodi, D., 2011. Rapid green synthesis of gold nanoparticles using *Rosa hybrida* petal extract at room temperature. Spectrochim. Acta Part A Mol. Biomol. Spectrosc. 79, 1461–1465.
- Oleinnikov, D.N., Agafonova, S.V., Penzina, T.A., Borovskii, G.B., 2014. Fatty acid composition of fourteen wood-decaying basidiomycete species growing in permafrost conditions. Rec. Nat. Prod. 8, 184–188.
- Owaid, M.N., 2017. Antagonistic role of hypha and cell-free culture filtrates of medicinal mushrooms to *Verticillium* sp. and *Pythium* sp. Fungal pathogens. Curr. Res. Environ. Appl. Mycol. J. Fungal Biol. 7, 94–102. <https://doi.org/10.5943/cream/7/2/6>.
- Owaid, M.N., 2019a. Green synthesis of silver nanoparticles by *Pleurotus* (oyster mushroom) and their bioactivity : review. Environ. Nanotechnol. Monit. Manag. 12, 100256. <https://doi.org/10.1016/j.enmm.2019.100256>.
- Owaid, M.N., 2019b. Silver nanoparticles as unique nano-drugs. In: Grumezescu, A.M., Grumezescu, V. (Eds.), Materials for Biomedical Engineering: Bioactive Materials, Properties and Applications. Elsevier, pp. 545–580.
- Owaid, M.N., 2020. Biomedical applications of nanoparticles synthesized from mushrooms. In: Patra, J., Fraceto, L., Das, G., C.E (Eds.), Green Nanoparticles. Nanotechnology in the Life Sciences. Springer Nature Switzerland AG, pp. 289–303. [https://doi.org/10.1007/978-3-030-39246-8\\_14](https://doi.org/10.1007/978-3-030-39246-8_14).
- Owaid, M.N., Ibraheem, I.J., 2017. Mycosynthesis of nanoparticles using edible and

- medicinal mushrooms. *Eur. J. Nanomed.* 9, 5–23. <https://doi.org/10.1515/ejnm-2016-0016>.
- Owaid, M.N., Raman, J., Lakshmanan, H., Al-Saeedi, S.S.S., Sabaratnam, V., Ali, I.A., 2015. Mycosynthesis of silver nanoparticles by *Pleurotus cornucopiae* var. *Citrinopileatus* and its inhibitory effects against *Candida* sp. *Mater. Lett.* 153, 186–190. <https://doi.org/10.1016/j.matlet.2015.04.023>.
- Owaid, M.N., Al-Saeedi, S.S.S., Al-Assaffii, I.A.A., 2017. Antifungal activity of cultivated oyster mushrooms on various agro-wastes. *Summa Phytopathol.* 43, 9–13. <https://doi.org/10.1590/0100-5405/2069>.
- Owaid, M.N., Muslim, R.F., Hamad, H.A., 2018. Mycosynthesis of silver nanoparticles using *terminia* sp. Desert truffle, Pezizaceae, and their antibacterial activity. *Jordan J. Biol. Sci.* 11, 401–405.
- Owaid, M.N., Ayesh, M., Abdul, A., Jameel, M.S., Ali, M., 2019. Mushroom-assisted synthesis of triangle gold nanoparticles using the aqueous extract of fresh *Lentinula edodes* (shiitake), Omphalotaceae. *Environ. Nanotechnol. Monit. Manag.* 12, 100270. <https://doi.org/10.1016/j.enmm.2019.100270>.
- Owaid, M.N., Naem, G.A., Muslim, R.F., Oleiwi, R.S., 2020. Synthesis, characterization and antitumor efficacy of silver nanoparticle from *Agaricus bisporus* pileus, Basidiomycota. *Walailak J. Sci. Technol.* 17, 75–87.
- Pala, S.A., Wani, A.H., Bhat, M.Y., 2011. Six hitherto unreported basidiomycetic macrofungi from Kashmir Himalayas. *Nusant Biosci.* 3, 92–97. <https://doi.org/10.13057/nusbiosci/n030206>.
- Pala, S.A., Wani, A.H., Ganai, B.A., 2019. Antimicrobial potential of some wild macro-mycetes collected from Kashmir Himalayas. *Plant Sci. Today* 6, 137–146. <https://doi.org/10.14719/pst.2019.6.2.503>.
- Pietrzak, K., Glińska, S., Gapińska, M., Ruman, T., Nowak, A., Aydin, E., Gutarowska, B., 2016. Silver nanoparticles: a mechanism of action on moulds. *Metallomics* 8, 1294–1302. <https://doi.org/10.1039/c6mt00161k>.
- Rabeea, M.A., Owaid, M.N., Aziz, A.A., Jameel, M.S., Dheyab, M.A., 2020. Mycosynthesis of gold nanoparticles using the extract of *Flammulina velutipes*, Physalacriaceae, and their efficacy for decolorization of methylene blue. *J. Environ. Chem. Eng.* 103841. <https://doi.org/10.1016/j.jece.2020.103841>.
- Shang, Y., Min, C., Hu, J., Wang, T., Liu, H., Hu, Y., 2013. Synthesis of gold nanoparticles by reduction of HAuCl<sub>4</sub> under UV irradiation. *Solid State Sci* 15, 17–23.
- Shankar, S.S., Ahmad, A., Sastry, M., 2003. *Geranium* leaf assisted biosynthesis of silver nanoparticles. *Biotechnol. Prog.* 19, 1627–1631. <https://doi.org/10.1021/bp034070w>.
- Silverstein, R., Webster, F., Kiemle, D., 2005. *Spectrometric Identification of Organic Compounds*, 7th ed. John Wiley and sons, Inc., London, UK.
- Soliman, H., Elsayed, A., Dyaa, A., 2018. Antimicrobial activity of silver nanoparticles biosynthesised by *Rhodotorula* sp. Strain ATL72. *Egypt J. Basic Appl. Sci.* 5, 228–233. <https://doi.org/10.1016/j.ejbas.2018.05.005>.
- Suay, I., Arenal, F., Asensio, F., Basilio, A., Cabello, M., Díez, M., García, J., Del Val, A., Gorrochategui, J., Hernández, P.P.F., 2000. Screening of basidiomycetes for antimicrobial activities. *Antonie Van Leeuwenhoek* 78, 129–140.
- Velusamy, P., Kumar, G.V., Jeyanthi, V., Das, J., Pachaiappan, R., 2016. Bio-inspired green nanoparticles: synthesis, mechanism, and antibacterial application. *Toxicol. Res.* 32, 95–102. <https://doi.org/10.5487/TR.2016.32.2.095>.
- Virkutyte, J., Varma, R.S., 2011. Green synthesis of metal nanoparticles: biodegradable polymers and enzymes in stabilization and surface functionalization. *Chem. Sci.* 2, 837–846.
- Yamanaka, M., Hara, K., Kudo, J., 2005. Bactericidal actions of a silver ion solution on *Escherichia coli*, studied by energy-filtering transmission Electron microscopy and proteomic analysis. *Appl. Environ. Microbiol.* 71, 7589–7593. <https://doi.org/10.1128/AEM.71.11.7589>.
- Yang, S., Bao, H., Wang, H., Lia, Q., 2019. Anti-tumour effect and pharmacokinetics of an active ingredient isolated from *Inonotus hispidus*. *Biol. Pharm. Bull.* 42, 10–17. <https://doi.org/10.1248/bpb.b18-00343>.
- Zan, L.F., Qin, J.C., Zhang, Y.M., Yao, Y.H., Bao, H.Y., Li, X., 2011. Antioxidant hispidin derivatives from medicinal mushroom *Inonotus hispidus*. *Chem. Pharm. Bull.* 59, 770–772. <https://doi.org/10.1248/cpb.59.770>.