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Research Article

Biosynthesis of Iron, Copper and Silver Nanoparticles Using *Polygonum Cognatum* and *Tragopogon Porrifolius* Extracts and Evaluation of Their Antimicrobial Potentials

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ABSTRACT

In this study, the biosynthesis of iron (Fe), copper (Cu) and silver (Ag) nanoparticles was aimed using aqueous extracts of *Polygonum cognatum* (*P. cognatum*) and *Tragopogon porrifolius* (*T. porrifolius*). The synthesized nanoparticles were characterized using UV/Vis spectroscopy, fourier transform infrared spectroscopy (FTIR) and dynamic light scattering technique (DLS). The antibacterial activity of the nanoparticles was analyzed against well-known pathogens *Klebsiella pneumonia*, *Pseudomonas aeruginosa*, *Enterococcus faecalis* and *Staphylococcus aureus*. In addition, the anti-fungal activity of the nanoparticles against *Candida albicans* and *Candida utilis* strains was evaluated. The obtained results showed that the synthesized nanoparticles have a moderate antimicrobial effect.

Keywords: *Antimicrobial activity, Metallic nanoparticles, Polygonum cognatum, Tragopogon porrifolius*

Polygonum Cognatum ve *Tragopogon Porrifolius* Ekstraktları Kullanılarak Demir, Bakır ve Gümüş Nanopartiküllerin Biyosentezi ve Antimikrobiyal Potansiyellerinin Değerlendirilmesi

ÖZ

Bu çalışmada, *Polygonum cognatum* (*P. cognatum*) ve *Tragopogon porrifolius*'un (*T. porrifolius*) sulu ekstreleri kullanılarak demir (Fe), bakır (Cu) ve gümüş (Ag) nanopartiküllerin biyosentezi amaçlanmıştır. Sentezlenen NP'ler UV/Vis spektroskopisi, fourier transform kızılötesi spektroskopisi (FTIR) ve dinamik ışık saçılım (DLS) tekniği kullanılarak karakterize edilmiştir. Nanopartiküllerin antibakteriyel aktivitesi, iyi bilinen patojenler *Klebsiella pneumonia*, *Pseudomonas aeruginosa*, *Enterococcus faecalis* ve *Staphylococcus aureus*'e karşı analiz edilmiştir. Ek olarak bu nanopartiküllerin *Candida albicans* ve *Candida utilis* suşlarına karşı antifungal aktivitesi de değerlendirilmiştir. Elde edilen sonuçlar, sentezlenen nanopartiküllerin orta düzeyde bir antimikrobiyal etkiye sahip olduğunu göstermiştir.

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Anahtar Kelimeler: *Antimikrobiyal aktivite, Metalik nanopartiküller, Polygonum cognatum, Tragopogon porrifolius*

I. INTRODUCTION

Nanotechnology offers great potential in the development of new generation drugs in microbial infections in recent years. Metallic nanoparticles (MNPs) have unique optical, chemical, physical, and electrical properties and due to these properties, they have become a new area of interest in nanotechnology [1], [2]. MNPs are versatile nanoparticles with potential use in wound dressings, cell labeling, photo-imaging, sensors, drug delivery, gene delivery, microbial infections, and cancer treatment [3], [4]. Although MNPs have broad therapeutic potential, they have been reported to cause undesirable toxicity. The unwanted toxicity of MNPs can be reduced by coating the nanoparticle surface with biomolecules [5].Therefore, biosynthesis, where biomolecules are used as reducing agents, has become increasingly important. Biosynthesis combines MNPs with the properties of the biomolecule used and reduces the undesirable toxicities of MNPs [6]. The biosynthesis of MNPs can overcome the adverse conditions of physical and chemical methods such as hazardous waste generation, high energy requirements and use of toxic chemicals [2], [7]-[9]. In recent years, researchers have been synthesizing metallic nanoparticles using different metals (such as silver, iron, manganese, gold, zinc, and copper) [10]-[12]. There are many studies using plants, algae, fungi, and bacteria in the synthesis of MNPs [13]-[15]. MNPs exhibit various properties depending on the characteristics of the biological material used in their synthesis [16], [17].MNPs are the most promising inorganic nanoparticles for resistance to conventional antibiotics. These nanoparticles use completely different action mechanisms against antibiotic-resistant bacteria than conventional antibiotics and target multiple biomolecules that are important in the development of resistant strains. MNPs can induce the production of reactive oxygen species. Metal ions can form strong coordination bonds with N, O or S atoms. These atoms are abundant in organic compounds and biomolecules. Because the bonding between metal ions and biomolecules is often nonspecific, MNPs generally exhibit a broad spectrum of activity [18].

Polygonum cognatum (P. cognatum) belongs to the *Polygonaceae* family and is a perennial. It has been used as a diuretic in Turkish folk medicine [19]. *P. cognatum* contains phenolic compounds, polyuronides, vitamin C, saponins and carotenoids. It has been demonstrated by various studies that it has antimicrobial, diuretic, antioxidant, anticancer and antidiabetic activities [19]-[21].

Tragopogon porrifolius (T. porrifolius) belongs to the *Asteraceae* family and is an annual. *T. porrifolius* is consumed in Southern and North America, Central Europe and the United Kingdom [22]. This plant contains essential fatty acids, vitamins, polyphenol and monounsaturated fatty acids [23]. The hepatoprotective, anti-inflammatory, antioxidant, and anticancer activity of *T. porrifolius* has been demonstrated by various studies [22]-[25].

In this study, CuNPs, FeNPs and AgNPs were synthesized using *P. cognatum* and *T. porrifolius*. Physical and chemical characterizations of synthesized nanoparticles were performed. The antimicrobial effects of synthesized nanoparticles on bacteria and fungi were investigated.

MATERIALS AND METHODS

A. EXPERIMENTAL

A.1. Metarials

Silver nitrate ($AgNO_3$) was from Carlo Erbaa. Iron (II) sulfate heptahydrate, Copper (II) sulfate pentahydrate was purchased from Sigma Aldrich. *P. cognatum* and *T. porrifolius* were obtained from Tokat Gaziosmanpaşa University, Faculty of Agriculture, Turkey. Bacterial (*E. faecalis (ATCC 29212)*, *P. aeruginosa* (*ATCC 27853)*, *S. aureus* (*ATCC 25923*) and *K. pneumonia* (*ATCC 15380)*) and fungal

(*C. albicans (ATCC 90819)* and *C. utilis (9950)*) strains were obtained from Tokat Gaziosmanpasa University, Department of Bioengineering and Genetics, Turkey.

A.2. Preparation of *P. cognatum* and *T. porrifolius* Extracts

P. cognatum (20 g) and *T. porrifolius* (20 g) were weighed. Each of them was taken into 200 mL of distilled water separately and homogenized thoroughly with a blender. They were then mixed for 90 minutes at 60 °C in a shaking incubator at 100 rpm. The obtained homogenates were filtered and and the extracts were obtained by centrifugation at 6 000 rpm for 10 minutes. The extracts (PC-Extract and TP-Extract) were stored at +4 °C before use.

A.3. Synthesis of Metallic Nanoparticles

A.3.1. Synthesis of Silver Nanoparticles (AgNPs)

While all nanoparticles were being synthesized, pre-optimization was made and the optimum extract: metal salt ratio was determined by monitoring the nanoparticle formation with UV-Vis analysis. There is a correlation between the absorption peaks and the particle size of the NPs. Studies have shown that as the particle size decreases, the absorption peaks shift to smaller wavelengths [26]. 10mM silver nitrate was mixed with TP-Extract at a ratio of 2:1 and PC-Extract at a ratio of 1:1 in separate beakers and kept in the microwave for 60 seconds at 475 watts. After the mixture was cooled, it was centrifuged at 20 000 rpm for 10 minutes and the nanoparticles were precipitated. Silver nanoparticles from TP-Extract and PC-Extract (TP-AgNPs and PC-AgNPs, respectively) were washed twice with distilled water and dried at 37 °C.

A.3.2. Synthesis of Copper Nanoparticles (CuNPs)

0.1 M copper sulfate was mixed with TP-Extract at a ratio of 1:4 and PC-Extract at a ratio of 1:5 in beakers and kept in the microwave for 60 seconds at 475 watts. After the mixture was cooled, it was centrifuged at 20 000 rpm for 10 minutes and the nanoparticles were precipitated. Copper nanoparticles from TP-Extract and PC-Extract (TP-CuNPs and PC-CuNPs, respectively) were washed with distilled water and dried at 37 °C.

A.3.3. Synthesis of Iron Nanoparticles (FeNPs)

0.1 M iron sulfate was mixed with TP-Extract at a ratio of 1:4 and PC-Extract at a ratio of 1:5 in beakers and kept in the microwave for 60 seconds at 475 watts. After the mixture was cooled, it was centrifuged at 20 000 rpm for 10 minutes and the nanoparticles were precipitated. Iron nanoparticles from TP-Extract and PC-Extract (TP-FeNPs and PC-FeNPs, respectively) were washed with distilled water and dried at 37 °C.

A.4. Characterization of Metallic Nanoparticles

Characterization of MNPs was performed using a spectrophotometer (DeNoVIX, Wilmington, USA), DLS instrument (HORIBA SZ-100), and FTIR (Jasco FTIR 4700, Germany). The formation of MNPs was characterized by scanning at wavelengths in the 300-800 nm range using spectrophotometer. The presence of extracts used to synthesize MNPs was determined using FTIR. The size and zeta potential of MNPs were measured using the DLS instrument.

A.5. Antimicrobial Activity of Metallic Nanoparticles

The antimicrobial potentials of the synthesized MNPs were determined by minimum inhibitory concentration analysis as detailed in our previous studies [4], [12], [14]. In summary, overnight cultures of *K. pneumonia, P. aeruginosa, E. faecalis, S. aureus, C. albicans,* and *C. utilis* microorganisms were

seeded into 96-well plates according to the 0.5 McFarland standard. 0.5 McFarland turbidity standard equals the density of a bacterial suspension of $1.5x10^8$ colony forming units (CFU mL^{-1}) [27]. Then, the synthesized MNPs were applied to the microorganisms in the concentration range of 1000 µg mL^{-1}-15.625 µg mL^{-1}. The cells were incubated at 37 °C for 24 hours and their absorbance at 600 nm was read using a microplate reader. Percent inhibition and minimum inhibitory concentration (MIC) values of microorganisms exposed to synthesized MNPs were calculated relative to the control groups.

III. RESULTS AND DISCUSSION

A. SYNTHESIS OF MNPS USING PC-EXTRACT AND TP-EXTRACT

MNPs were synthesized with a microwave assistance using aqueous extracts of *P. cognatum* and *T. porrifolius*. Color change in the extracts due to synthesis of AgNPs, CuNPs, and FeNPs was shown in Figure 1. The color change of the extracts was a confirmation that metal ions were reduced and MNPs were formed.

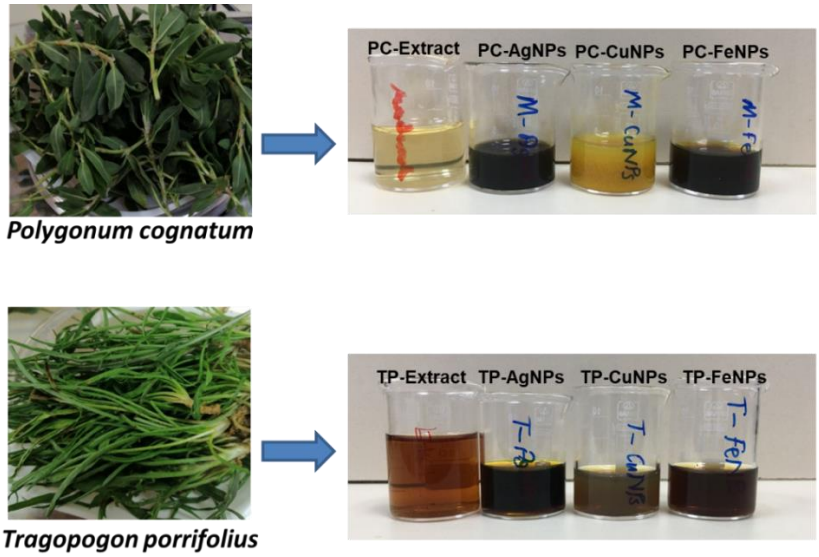
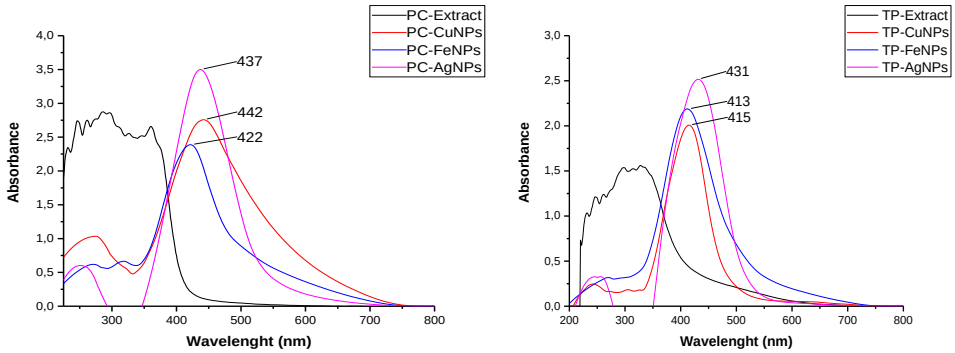


Figure 1. *Synthesis of AgNPs, CuNPs, and FeNPs.*

B. CHARACTERIZATION OF METALLIC NANOPARTICLES

B.1. UV-Visible Spectroscopy

To prove MNPs formation, UV-vis spectroscopy is routinely used in characterization of MNPs [28]. The spectrum of the produced MNPs using PC-Extract and TP-Extract were shown as Figure 2. The maximum absorption peaks of CuNPs, FeNPs, and AgNPs synthesized using PC Extract were analyzed as 437 nm, 442 nm, and 422 nm, respectively, while the maximum absorption peaks of CuNPs, FeNPs, and AgNPs synthesized using TP-Extract were determined as 431 nm, 413 nm, and 415 nm respectively.



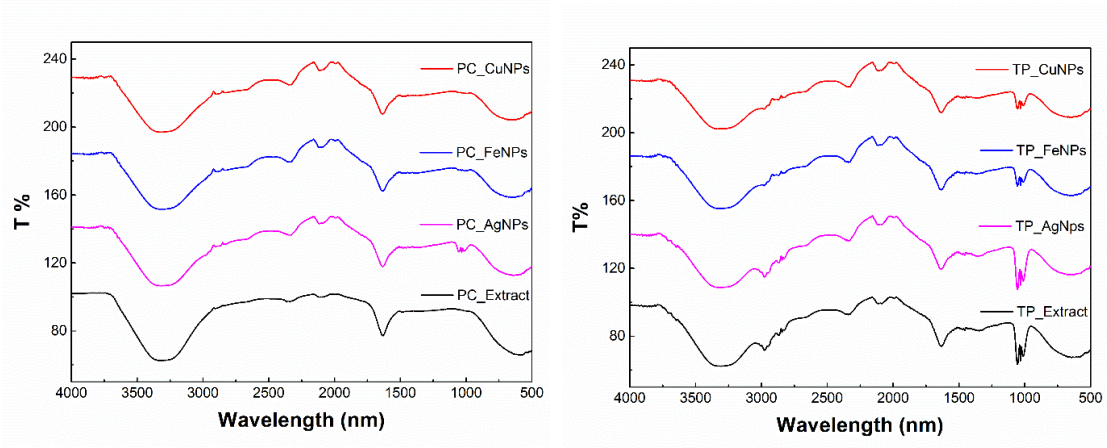
(a) *(b)*

Figure 2. *UV-Vis absorption spectra of the synthesized MNPs using PC-Extract* ***(a)****, and TP-Extract* ***(b).***

The peak formation in the UV-Vis absorption spectra of MNPs may differ in relation to the size and shape of the particle. The UV spectrum wavelengths of MNPs range from 300 to 800 nm, indicating the incidence of various MNPs [10]. When the wavelengths at which the MNPs gave their peaks were compared with the peaks of the crude extracts, it was observed that the peaks were different from each other. Only one sharp peak appeared in the 300 to 800 nm range in all spectra in this study, these peaks can be attributed to the absorption of NPs. This was also evidence for the formation of MNPs.

B.2. FTIR spectroscopy

FTIR spectroscopy curves were used to determine chemical characterization of synthesized MNPs using PC-Extract, and TP-Extract. FTIR spectrums of the synthesized MNPs using PC-Extract (a), and TP-Extract (b) were shown in Figure 3.



(a) *(b)*

Figure 3. *FTIR spectra of the synthesized MNPs using PC-Extract* ***(a)****, and TP-Extract* ***(b).***

The synthesized MNPs using PC-Extract and TP-Extract exhibited a broad absorption band of –OH groups at 3319.86 cm^{-1}. The peak at 2887 cm^{-1} was associated with C–H stretching of aliphatic –CH, –CH2 groups. The absorption peaks at 1634 cm^{-1} and 1031 cm^{-1} were respectively assigned to the -C-O-C- or –C-O- bonds and stretching of -C=C bonds. The presence of functional groups from the plant extract responsible for the reduction of MNPs in the FTIR analysis proved the synthesis of MNPs from PC-extract and TP-extract.

B.3. Dynamic light scattering (DLS)

To determine physical properties like particle size and zeta potential of nanoparticles DLS was preferred. DLS results were shown in Figure 4 and Figure 5.

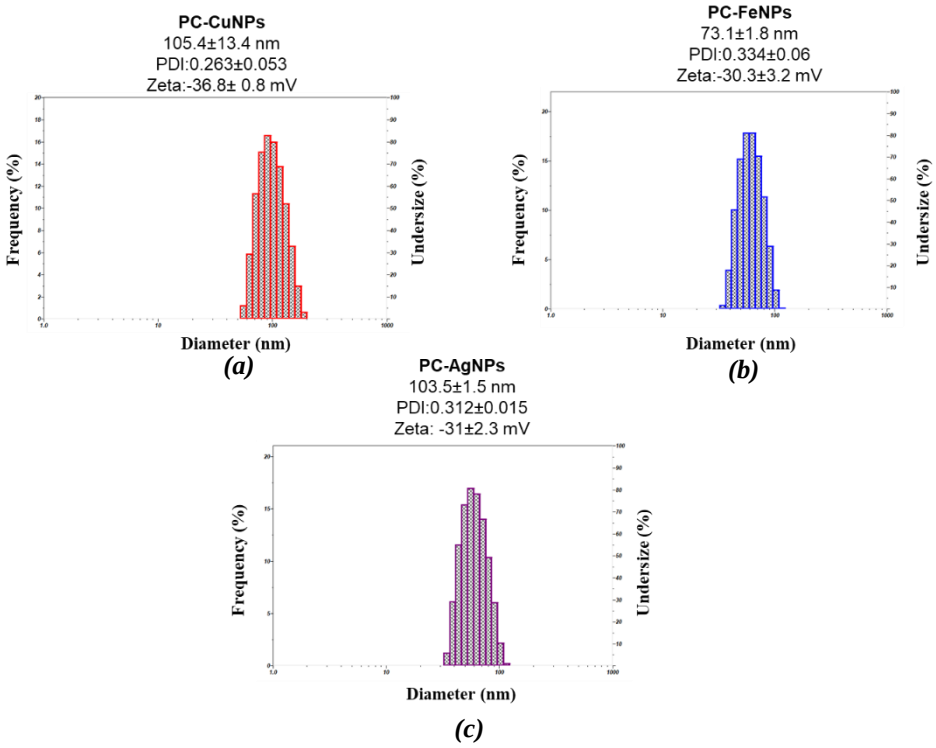


Figure 4. *DLS data of synthesized MNPs using PC-Extract.*

Figure 4 presented DLS data of synthesized CuNPs ***(a)***, FeNPs ***(b)*** and AgNPs ***(c)*** using PC-Extract. It was reported that among the synthesized MNPs using PC-Extract, the smallest particle size was formed using iron salt but zeta potentials of MNPs were similar. On the other hand, the PDI values of the MNPs, which give information about the particle size distribution, were also close to each other.

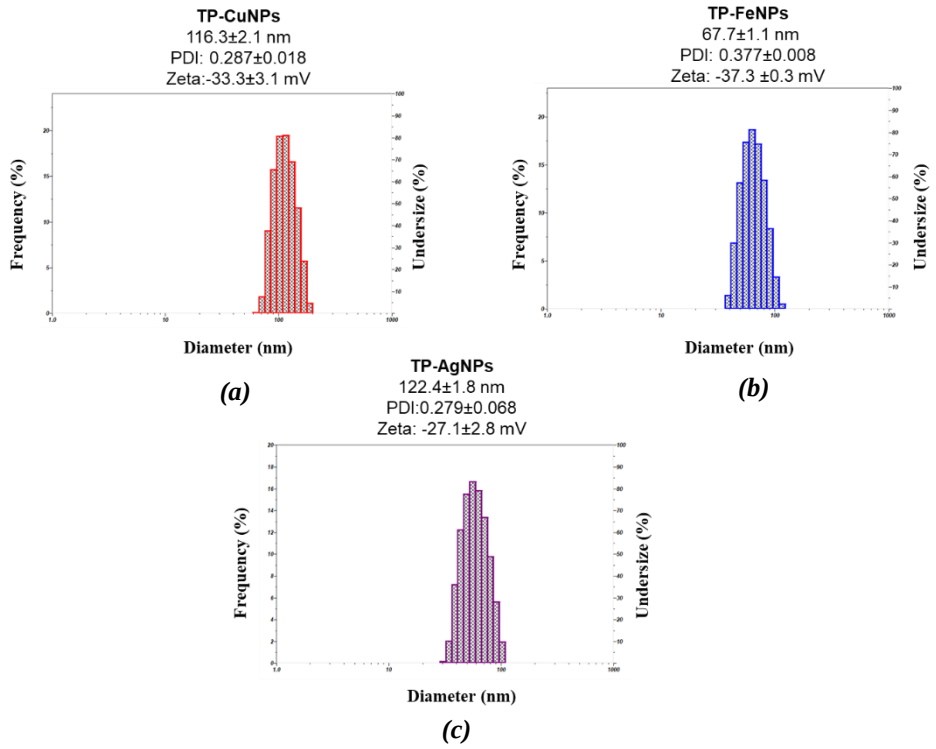


Figure 5. *DLS data of synthesized MNPs using TP-Extract.*

Figure 5 presented DLS data of synthesized CuNPs ***(a)***, FeNPs ***(b)*** and AgNPs ***(c)*** using TP-Extract. Particle sizes of AgNPs and CuNPs were similar while FeNPs had the smallest particle size among the synthesized all MNPs using PC-Extract and TP- Extract. However, zeta potentials and PDI values were also similar.

B.4. Antibacterial Activity of Biosynthesized MNPs Using PC-Extract and TP-Extract

MNPs, which are synthesized using various natural products, and metal salts, are known to exhibit antibacterial activities against bacteria strain [11]. In this study, MNPs were produced using PC-Extract and TP- Extract and their antibacterial activities were tested against *S. aureus*, *E. faecalis*, *P. aeruginosa*, and *K. pneumonia* bacteria strain. To determine the antibacterial activity of produced all MNPs, the minimum inhibition concentration method was used. Measurements were evaluated as the percentage of inhibition versus concentrations.

Inhibition curves of produced MNPs using PC-Extract were indicated in Figure 6. The inhibition value of the crude PC-Extract shown in Figure 6 (d) was less than fifty percent at the maximum concentration. Also, CuNPs, FeNPs, and AgNPs did not affect *K. pneumonia* and *E. faecalis*. However, CuNPs, FeNPs, and AgNPs showed more toxic effects on *S. aureus* and *P. aeruginosa* than PC-Extract, and higher inhibition values were obtained at the same concentration. It was tested that AgNPs were the most effective on *S. aureus*, while CuNPs were the most effective on *P. aeruginosa*.

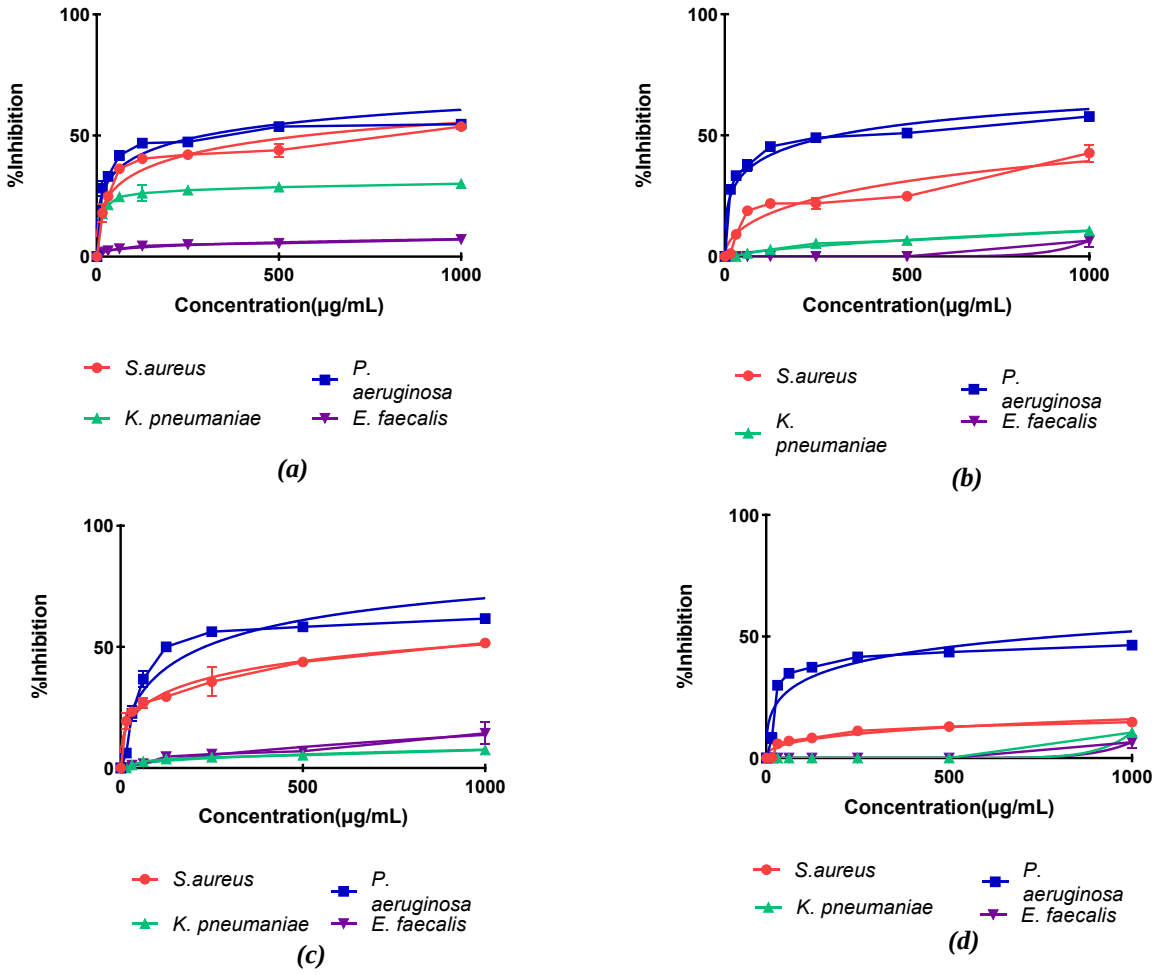


Figure 6*. Growth inhibition curves of pathogenic bacteria strains exposed to PC-Extract and produced MNPs using this extract.*

Figure 7 presented the inhibition curves of produced MNPs using TP-Extract. MNPs and TP-Extract did not show the antibacterial effect on *K. pneumonia*, also only CuNPs inhibited *E. faecalis*. Antibacterial activity, percent inhibition values of MNPs and TP-Extract on *P. aeruginosa* were very close to each other and it was determined that the synthesized MNPs did not provide any increase in inhibition value. However, the produced AgNPs were the most effective on *S. aureus* and increased in inhibition value. All of the synthesized MNPs and both extracts had no antibacterial effect on *K. pneumoniae* but all MNPs and both extracts inhibited *P. aureginosa*. Inhibition data of over fifty percent indicate that MNPs may completely inhibit strains at a concentration above 1000 µg mL^{-1}. Eruygur et al. showed that *P. cognatum* and *T. porrifolius* ethanol extracts inhibited *P. aureginosa* and *S. aureus* but had no antimicrobial effect on *E. faecalis* and *K. pneumoniae* [22]. In addition to, Yıldırım et al. revealed that ether and ethanol extracts of *P. cognatum* exhibited antimicrobial activity against *S. aureus* and *Bacillus subtilis* [20].

Pekdemir et al. synthesized magnetic Fe_3O_4 nanoparticles and processed these nanoparticles with the ethanolic extract of *P. cognatum*. The ethanolic extract of *P. cognatum* had a moderate antimicrobial effect against *S. aureus*, *Escherichia coli*, *K. pneumoniae*, *C. albicans*, and *Bacillus megaterium* while iron nanoparticles treated with *P. cognatum* did not show antimicrobial effect. The iron magnetic nanoparticles destroyed the antimicrobial effect of the plant extract [29].

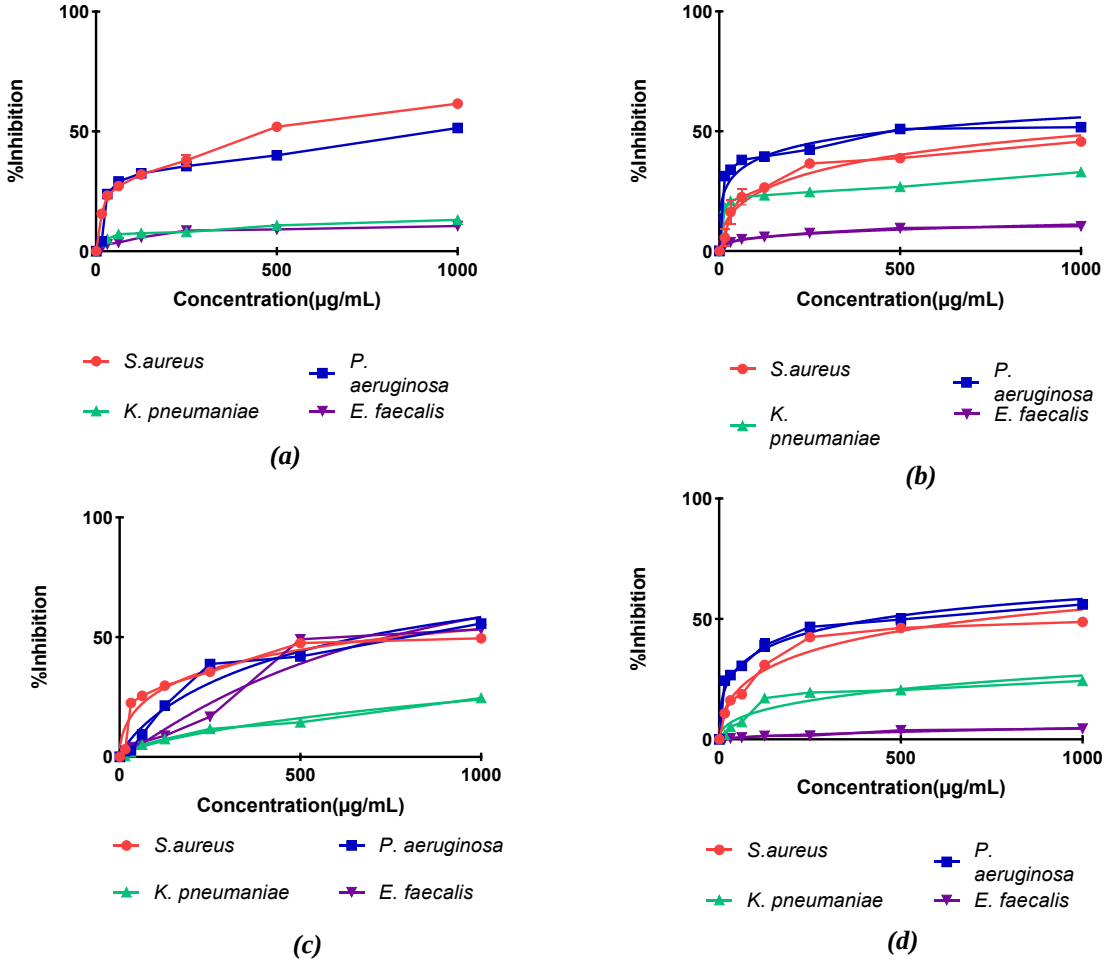


Figure 7. *Growth inhibition curves of pathogenic bacteria strains exposed to TP-Extract and produced MNPs using this extract.*

B.5. Antifungal Activity of Biosynthesized MNPs Using PC-Extract and TP- Extract

Antifungal activities of the synthesized MNPs using PC-Extract and TP-Extract were tested on *C. utilis* and *C. albicans* as another biological activity. *C. utilis* and *C. albicans* are known as significant human fungal pathogens [30], [31]. Antifungal activity was determined using same method, minimum inhibitory method indicated as Figure 8. PC-Extract, FeNPs, and CuNPs did not inhibit *C. albicans*, but AgNPs were exhibited in increased in inhibition value of *C. albicans* when compared to crude PC-Extract. Yıldırım et al. showed that ether, water and ethanol extracts of *P. cognatum* had no antifungal effect against *C. albicans* [20].

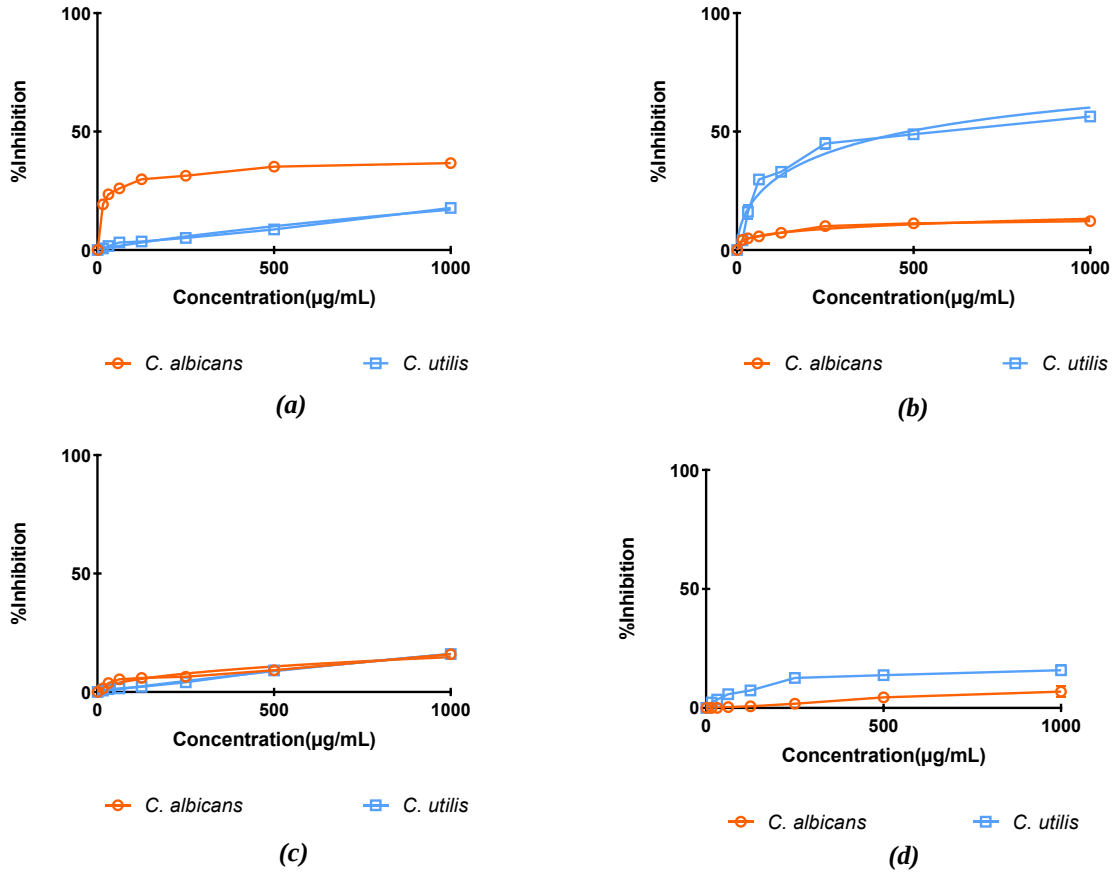


Figure 8. *Growth inhibition curves of human fungal pathogen strains exposed to PC-Extract and produced MNPs using this extract.*

Among the synthesized MNPs using PC-Extract, FeNPs were the most effective on *C. utilis* and its inhibition value was over fifty percent at a concentration 1000 µg mL^{-1}. Except for the effect of PC-FeNPs on *C. utilis*, MNPs produced from PC-Extract did not generally exhibit antifungal activity.

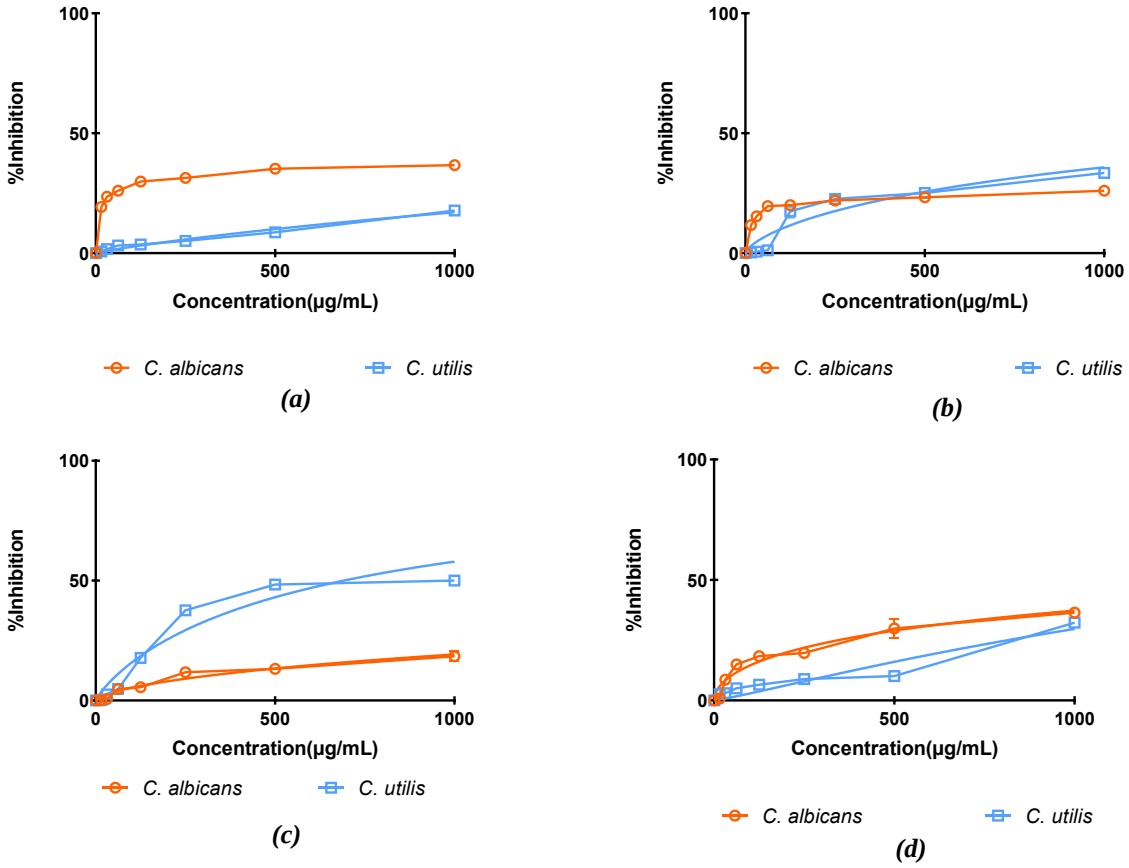


Figure 9. *Growth inhibition curves of human fungal pathogen strain exposed to TP-Extract and produced MNPs using this extract.*

Antifungal activities of the synthesized MNPs using TP-Extract were shown in Figure 9. CuNPs were exhibited antifungal activity on *C. utilis* at maximum concentration. AgNPs, FeNPs and TP-Extract had no antifungal activities at the applied concentrations. Eruygur et al. revealed that *T. porrifolius* ethanol extract did not exhibit antifungal activity against *C. albicans* [22].

Plants contain a wide variety of active biomolecules. Almost all parts of plants can be used in the synthesis of metallic NPs. The phenols and flavonoids of the plant exhibit specific chemical properties and can synthesize nanoparticles by reducing metallic salts. The fact that these biomolecules contain carboxyl and hydroxyl groups allows them to bind to the metal. Medicinal plants have many natural and active ingredients, and these ingredients have certain health benefits. Such medicinal plants are not only used as reducing and stabilizing agents, but also assist in the development of NPs with fascinating biological properties [32]. The proteins, enzymes, organic acids and vitamins contained in the extract are responsible for the reduction and stabilization of NPs. It is widely known in the literature that there is a correlation between the biological properties of the plant and the final properties of the NPs synthesized using these plants [33]. In our study, there is a relationship between the antimicrobial activity of plant extracts and the antimicrobial activity of metallic nanoparticles synthesized from these plants.

IV. CONCLUSION

MNPs are rapidly progressing to become a new generation drug in the fight against microbial infections due to their antimicrobial properties [1], [2]. The use of plant extracts to make MNPs is simple, environmentally friendly, easily scaled up, inexpensive, convenient, and safe. It is a particularly viable

strategy for making NPs that should be free of toxic contaminants as required in biomedical and therapeutic applications. The plant extract-based synthesis of NPs is higher in yield than other physical and chemical methods [2], [7], [9].

In this study, MNPs were synthesized from two plants with antimicrobial properties as well as other therapeutic potentials such as antioxidant and anticancer. These nanoparticles have been characterized physically and chemically. The antimicrobial properties of the synthesized nanoparticles were evaluated. It was revealed that the obtained nanoparticles have a moderate antimicrobial effect.

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V. REFERENCES

[1] H. Barabadi, M. Najafi, H. Samadian, A. Azarnezhad, H. Vahidi, M. A. Mahjoub, M. Koohiyan, and A. Ahmadi, "A Systematic review of the genotoxicity and antigenotoxicity of biologically synthesized metallic nanomaterials: are green nanoparticles safe enough for clinical marketing?," Medicina (Kaunas), vol. 55, no. 8, 2019.

[2] M. Ovais, A. T. Khalil, A. Raza, M. A. Khan, I. Ahmad, N. U. Islam, M. Saravanan, M. F. Ubaid, M. Ali, and Z. K. Shinwari, "Green synthesis of silver nanoparticles via plant extracts: beginning a new era in cancer theranostics," Nanomedicine (Lond), vol. 11, no. 23, pp. 3157-3177, 2016.

[3] H. Barabadi, A. Alizadeh, M. Ovais, A. Ahmadi, Z. K. Shinwari, and M. Saravanan, "Efficacy of green nanoparticles against cancerous and normal cell lines: a systematic review and meta-analysis," IET Nanobiotechnol, vol. 12, no. 4, pp. 377-391, 2018.

[4] Ö. Kaplan, N. Gökşen Tosun, A. Özgür, S. Erden Tayhan, S. Bilgin, İ. Türkekul, and İ. Gökçe, "Microwave-assisted green synthesis of silver nanoparticles using crude extracts of Boletus edulis and Coriolus versicolor: Characterization, anticancer, antimicrobial and wound healing activities," Journal of Drug Delivery Science and Technology, vol. 64, pp. 102641, 2021.

[5] J. Xiong, X. D. Wu, and Q. J. Xue, "Biomolecule-assisted synthesis of highly stable dispersions of water-soluble copper nanoparticles," J Colloid Interface Sci, vol. 390, no. 1, pp. 41-6, 2013.

[6] D. Zhang, X. L. Ma, Y. Gu, H. Huang, and G. W. Zhang, "Green Synthesis of Metallic Nanoparticles and Their Potential Applications to Treat Cancer," Front Chem, vol. 8, pp. 799, 2020.

[7] A. Ahsan, M. A. Farooq, A. Ahsan Bajwa, and A. Parveen, "Green Synthesis of Silver Nanoparticles Using Parthenium Hysterophorus: Optimization, Characterization and In Vitro Therapeutic Evaluation," Molecules, vol. 25, no. 15, 2020.

[8] E. Alphandery, "Natural metallic nanoparticles for application in nano-oncology," Int J Mol Sci, vol. 21, no. 12, 2020.

[9] L. Carson, S. Bandara, M. Joseph, T. Green, T. Grady, G. Osuji, A. Weerasooriya, P. Ampim, and S. Woldesenbet, "Green synthesis of silver nanoparticles with antimicrobial properties using phyla dulcis plant extract," Foodborne Pathog Dis, vol. 17, no. 8, pp. 504-511, 2020.

[10] P. K. Thakur, and V. Verma, "A Review on Green Synthesis, Characterization and Anticancer Application of Metallic Nanoparticles," Appl Biochem Biotechnol, vol. 193, no. 7, pp. 2357-2378, 2021.

[11] A. Singh, P. K. Gautam, A. Verma, V. Singh, P. M. Shivapriya, S. Shivalkar, A. K. Sahoo, and S. K. Samanta, "Green synthesis of metallic nanoparticles as effective alternatives to treat antibiotics resistant bacterial infections: A review," Biotechnol Rep (Amst), vol. 25, pp. e00427, 2020.

[12] N. Gökşen Tosun, and Ö. Kaplan, "Optimization of the Green Synthesis of Silver Nanoparticle with Box-Behnken Design: Using Aloe Vera Plant Extract as a Reduction Agent," SAUJS, vol. 25, no. 3, pp. 774-787, 2021.

[13] B. Uzair, A. Liaqat, H. Iqbal, B. Menaa, A. Razzaq, G. Thiripuranathar, N. Fatima Rana, and F. Menaa, "Green and cost-effective synthesis of metallic nanoparticles by algae: safe methods for translational medicine," Bioengineering (Basel), vol. 7, no. 4, 2020.

[14] Ö. Kaplan, and N. Gökşen Tosun, "Biosynthesis of silver nanoparticles from Teucrioside and investigation of its antibacterial activity," Cumhuriyet Sci. J., vol. 42, no. 1, pp. 60-67, 2021.

[15] B. M. Abdallah, and E. M. Ali, "Green synthesis of silver nanoparticles using the lotus lalambensis aqueous leaf extract and their anti-candidal activity against oral candidiasis," ACS Omega, vol. 6, no. 12, pp. 8151-8162, 2021.

[16] A. Wibowo, G. U. N. Tajalla, M. A. Marsudi, G. Cooper, L. Asri, F. Liu, H. Ardy, and P. Bartolo, "Green synthesis of silver nanoparticles using extract of cilembu sweet potatoes (ipomoea batatas l var. rancing) as potential filler for 3d printed electroactive and anti-infection scaffolds," Molecules, vol. 26, no. 7, 2021.

[17] A. Salayova, Z. Bedlovicova, N. Daneu, M. Balaz, Z. Lukacova Bujnakova, L. Balazova, and L. Tkacikova, "Green synthesis of silver nanoparticles with antibacterial activity using various medicinal plant extracts: morphology and antibacterial efficacy," Nanomaterials (Basel), vol. 11, no. 4, 2021.

[18] E. Sánchez-López, D. Gomes, G. Esteruelas, L. Bonilla, A. L. Lopez-Machado, R. Galindo, A. Cano, M. Espina, M. Ettcheto, A. Camins, A. M. Silva, A. Durazzo, A. Santini, M. L. Garcia, and E. B. Souto, "Metal-Based Nanoparticles as Antimicrobial Agents: An Overview," Nanomaterials (Basel), vol. 10, no. 2, 2020.

[19] F. T. Guragac Dereli, M. Ilhan, E. Kozan, and E. Kupeli Akkol, "Effective eradication of pinworms (Syphacia obvelata and Aspiculuris tetraptera) with Polygonum cognatum Meissn," Exp Parasitol, vol. 196, pp. 63-67, 2019.

[20] A. Yıldırım, A. Mavi, and A. Kara, "Antioxidant and antimicrobial activities of Polygonum cognatum Meissn extracts," J. Sci. Food Agric., vol. 83, no. 1, pp. 64-69, 2003.

[21] M. Pehlivan, H. Karahan Çöven, B. Çerçi, A. Eldem, T. Öz, and N. Savlak, "The cytotoxic effect of Polygonium cognatum and chemotherapeutic effect of doxorubicin on glioblastoma cells," Eur J Ther, vol. 27, no. 1, pp. 50-54, 2021.

[22] N. Eruygur, E. Ucar, M. Atas, M. Ergul, M. Ergul, and F. Sozmen, "Determination of biological activity of Tragopogon porrifolius and Polygonum cognatum consumed intensively by people in Sivas," Toxicol Rep, vol. 7, pp. 59-66, 2020.

[23] C. Formisano, D. Rigano, F. Senatore, M. Bruno, and S. Rosselli, “Volatile constituents of the aerial parts of white salsify (Tragopogon porrifolius L., Asteraceae),” Nat Prod Res, vol. 24, no. 7, pp. 663-8, 2010.

[24] C. Zidorn, U. Lohwasser, S. Pschorr, D. Salvenmoser, K. H. Ongania, E. P. Ellmerer, A. Borner, and H. Stuppner, “Bibenzyls and dihydroisocoumarins from white salsify (Tragopogon porrifolius subsp. porrifolius),” Phytochemistry, vol. 66, no. 14, pp. 1691-7, 2005.

[25] C. Tenkerian, M. El-Sibai, C. F. Daher, and M. Mroueh, “Hepatoprotective, Antioxidant, and Anticancer Effects of the Tragopogon porrifolius Methanolic Extract,” Evid Based Complement Alternat Med, vol. 2015, pp. 161720, 2015.

[26] B. Plackal Adimuriyil George, N. Kumar, H. Abrahamse, and S. S. Ray, “Apoptotic efficacy of multifaceted biosynthesized silver nanoparticles on human adenocarcinoma cells,” Sci Rep, vol. 8, no. 1, pp. 14368, 2018.

[27] A. Zapata, and S. Ramirez-Arcos, “A comparative study of McFarland turbidity standards and the Densimat photometer to determine bacterial cell density,” Curr Microbiol, vol. 70, no. 6, pp. 907-9, 2015.

[28] J. Grand, B. Auguie, and E. C. Le Ru, “Correction to combined extinction and absorption uv-visible spectroscopy as a method for revealing shape imperfections of metallic nanoparticles,” Anal Chem, vol. 92, no. 5, pp. 4164, 2020.

[29] M. E. Pekdemir, S. Pekdemir, Ş. İnci, S. Kırnağ, and M. Çiftci, “Thermal, magnetic properties and antimicrobial effects of magnetic iron oxide nanoparticles treated with Polygonum cognatum,” Iran J Sci Technol Trans Sci vol. 45, pp. 1579–1586, 2021.

[30] F. L. Mayer, D. Wilson, and B. Hube, “Candida albicans pathogenicity mechanisms,” Virulence, vol. 4, no. 2, pp. 119-28, 2013.

[31] G. Scoppettuolo, C. Donato, E. De Carolis, A. Vella, L. Vaccaro, A. La Greca, and M. Fantoni, “Candida utilis catheter-related bloodstream infection,” Med Mycol Case Rep, vol. 6, pp. 70-2, 2014.

[32] S. S. Salem, and A. Fouda, “Green Synthesis of Metallic Nanoparticles and Their Prospective Biotechnological Applications: an Overview,” Biol Trace Elem Res, vol. 199, no. 1, pp. 344-370, 2021.

[33] F. Moradi, S. Sedaghat, O. Moradi, and S. A. Salmanabadi, “Review on green nano-biosynthesis of silver nanoparticles and their biological activities: with an emphasis on medicinal plants,” Inorganic and nano-metal chemistry vol. 51, no. 1, pp. 133-142.