



PREPARATION OF SILVER NANOPARTICLES FROM YEAST USING SABOURAUD DEXTROSE BROTH AND MEASUREMENT OF ANTIBACTERIAL EFFECT

Karrar Nadhim*, Aseel Mustafa and Nibras Nazar Mahmood

Physics Department, Mustansiriyah University, Iraq

Abstract

The method of preparing AgNPS in this paper is a simple and different from other methods. Where we used *Saccharomyces cerevisiae* and Sabouraud Dextrose broth (SDB) medium and mix it with AgNO_3 solution, the interaction between these solutions has produced AgNPS as a result of oxidative reduction in solution the size of crystal appears (15nm) in XRD and peak absorption (470nm), SEM images showed that AgNPS were different shapes (clusters, semispherical), AgNPS was used as an antibacterial on two types of pathogenic bacteria (*Staphylococcus epidermidis* and *Eshrichia coli*), the effectiveness was well.

Key words: Nanoparticles, AgNPS, Morphology.

Introduction

The meaning of the word “Nano” is called to any parameter when it is represented as a measure of 10-9 times of SI units. NPs and their applications have been undetected and unknown in recent years. The term nanotechnology was associated with the name of the Japanese scientist (Taniguchi, 1974).

Nano science is slowly stepping up in various fields until a group of scientists began to report on how heavy metals are being processed by Microorganisms.

This remediated of metal crystal is imperceptible. After evidence of nanoparticles is available, the applications of these NPs have expanded. For example, AgNPs are used as antibacterial agents in many facilities and infrastructure in China. It is also used as an antibacterial in surgical procedures to reduce infection during the procedure and also as antifungal (Mullen *et al.*, 1989) (Kalishwaralal *et al.*, 2009) (Gurunathan *et al.*, 2009).

Silver - Nps: AgNps obtained from silver nitrate have replaced silver oxide for antibacterial use silver was used war (Sheikpranbabu *et al.*, 2009) (Chu *et al.*, 1988) (Deitch *et al.*, 2009) (Margraff *et al.*, 1977) (Silver, 2003)

(Atiyeh *et al.*, 2007) (Law *et al.*, 2008), where used as an ointment to heal wounds. AgNPs better than Ago NPs in therapeutic uses, Since AgONPs are effective for short periods. The applications of AgNPS have increased to enter Various fields such as Medicine, food production health care, due to their distinct chemical and physical properties (Mechanical, thermal, optical, electrical etc.) (Gurunathan *et al.*, 2015) (Li *et al.*, 2010) (Mukherjee *et al.*, 2001) and biological advantages like antibacterial anticancer cell (Chernousova *et al.*, 2013). These physical, chemical and biological properties result from surface to volume ratio. (Gurunathan *et al.*, 2009) (Li *et al.*, 2001) physical and chemical methods for the properties preparation AgNps are good but they contain negative sides such as a high cost and toxicity and require many tools. (Gurunathan *et al.*, 2015) (Sharma *et al.*, 2009). The characteristic of the biological method for the preparation of AgNPs is limited toxicity, more productivity, simplicity and excellent solubility. (Gurunathan *et al.*, 2015) Safety aspect of NPs.

The properties of many materials change when they are reduced to nano scale the surface area of the particles increased, resulting in increased interactions with environment Nanomaterial's are of limited toxicity (Gwinn *et al.*, 2006) and therefore have the advantage in an

*Author for correspondence : E-mail: karrarnadhim.hf@gmail.com

medicine, biochemistry and nanotechnology (Bruchez *et al.*, 1998) (Cao *et al.*, 2004) (Cao Y.C., 2002) (Akerman *et al.*, 2009).

Materials and Methods

The silver nanoparticles were prepared by biologic way from yeast (*Saccharomyces cerevisiae*) (Sowbarnika *et al.*, 2018) (Kaushik *et al.*, 2015), a new method for the preparation of nanoparticles, where the yeast was grown in Sabouraud Dextrose Broth (SDB).

By adding (0.1 gm) form dry (*Saccharomyces cerevisiae*) to SDB where growth yeast colonies after putting the flask in shaker incubation at (30°C, 110 rpm) for two days, then made centrifuge for the growth of (*Saccharomyces cerevisiae* + SDB). We take (50 ml) from supernatant, dissolved (0.5 gm) from AgNO_3 in (50ml) Deionized water, the concentration will be (0.6 M), when we add yeast (Drop wise) the concentration will be (0.3 M), after that stirring for solution by stirrer device during 30 minutes and incubated flask in shaker incubation with the (110 rpm at 30°C) for 24h, the color of solution is changing from light yellow to dark brown then red then black over time as shown in fig. 1.

Take the precipitate of solution after centrifuged (5000 rpm, 10 minutes) and washed it for several times by ethanol and then we take the extract dry up with under vacuum oven (200°C, 3 hrs, -0.06 MPa) and thus will get AgNPs (dry powder).

Results and Discussion

XRD analysis: AgNPS Crystals did not appear in XRD with one phase but appeared in different phases, there are four peaks for silver $2\theta=38.32^\circ$, 44.47° , 64.62° , 77.55° and strongest peaks at $2\theta=38.32^\circ$, 77.55° with max intensity $I/I_1 = 100, 51$ (in order) and Fall with half max (FWHM) = 0.56, 0.47 (in order) as shown in fig. 2.

Miller/indices in order

$$2\theta = 38.32^\circ \rightarrow (111) \quad 2\theta = 44.47^\circ \rightarrow (200)$$

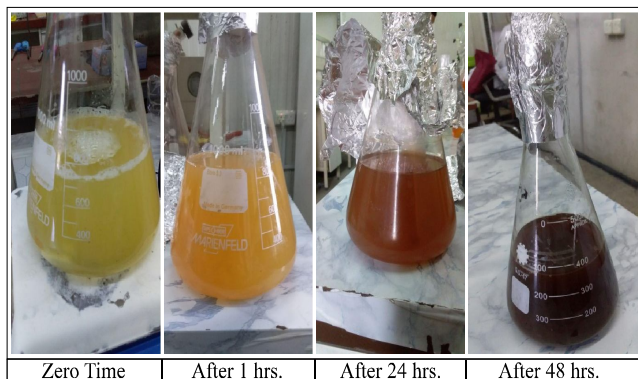


Fig.1: Changing colors during time.

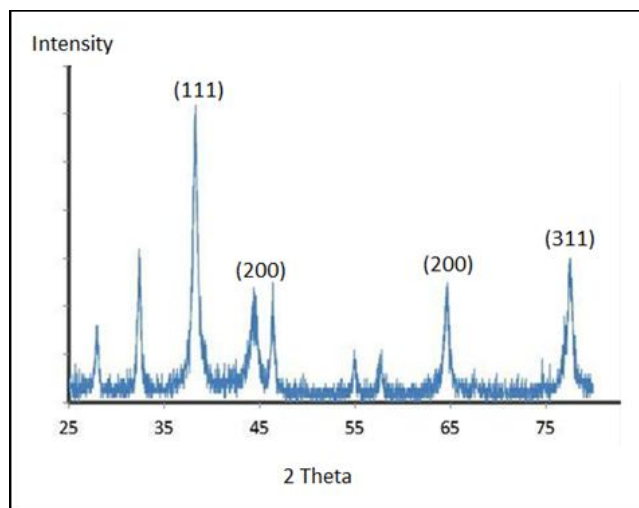


Fig. 2: X-ray diffraction of biosynthesized AgNps.

$$2\theta = 64.62^\circ \rightarrow (200) \quad 2\theta = 77.55^\circ \rightarrow (311)$$

UV-Vis spectroscopy

AgNPS solution was examined after 48h in UV-Vis device for the purpose of measuring absorption and the peak absorption was at wavelength (470)nm which corresponds to the wavelength for AgNPS, peak of the absorption at (470)nm indicate to AgNPS in solution as illustrated in fig. 3.

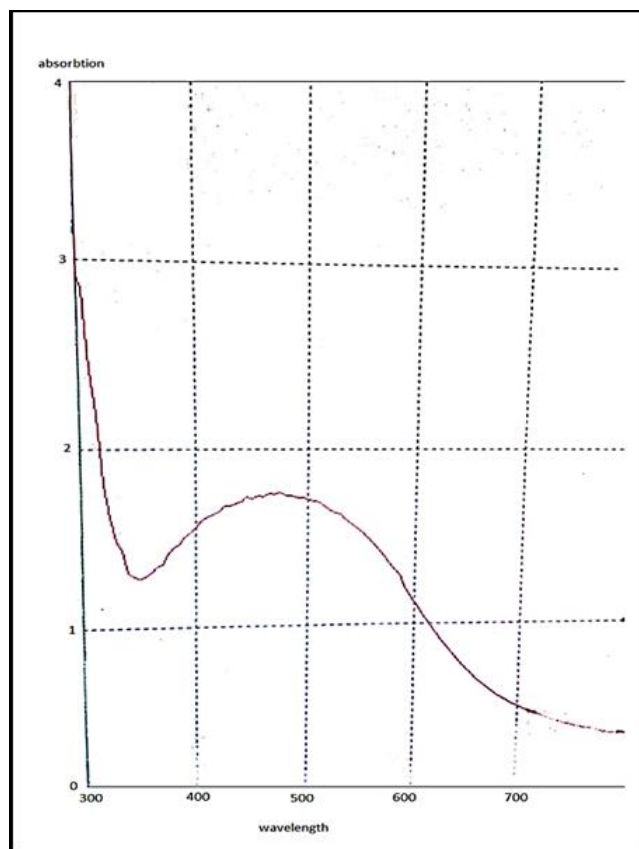


Fig. 3: Uv spectra of biosynthesis AgNps after 48 hrs.

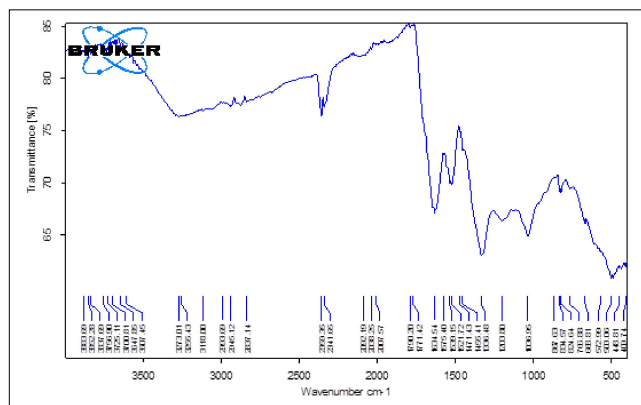


Fig. 4: FTIR spectra of the biosynthesized AgNps.

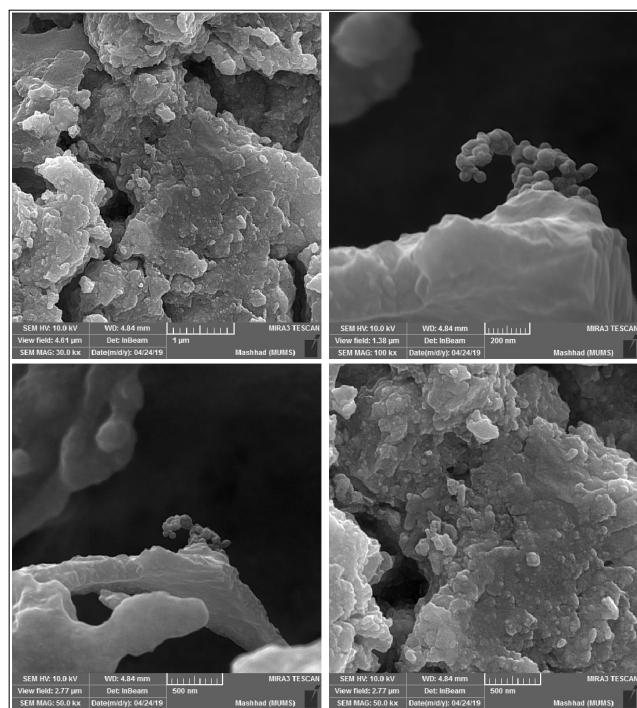


Fig. 5: SEM images of biogenic AgNps.

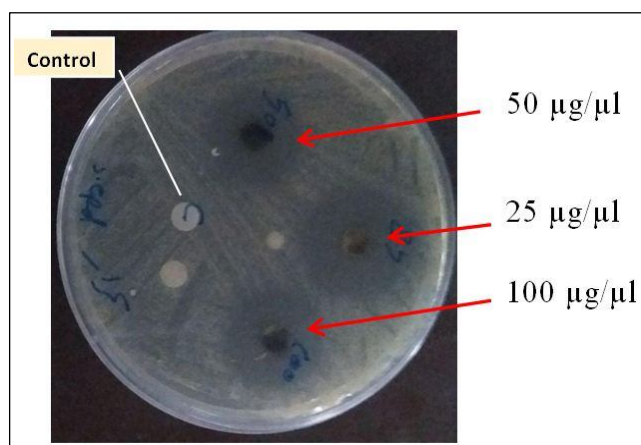


Fig. 6: Inhibition Zone of *Staphylococcus epidermidis*.

FTIR

From FTIR spectra of the biosynthesized AgNps we

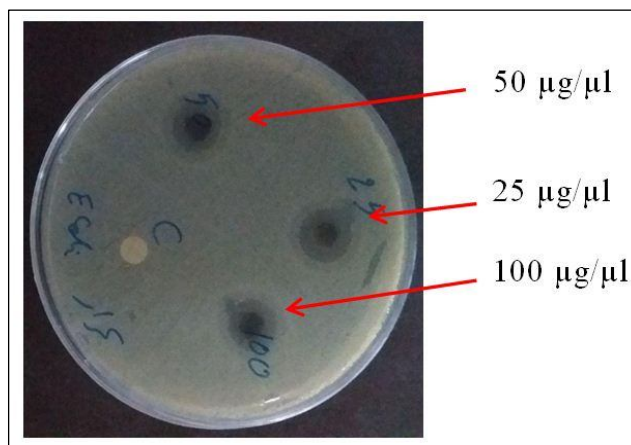


Fig. 7: Inhibition Zone of *E. coli*.

notice the following bands as shown in fig. 4.

- Band at 668.81 cm^{-1} (low band) Corresponding to Cl stretching.
- Band at 1634 cm^{-1} (Mid band) Corresponding to N-H banding frequency.
- Band at 2007 cm^{-1} corresponding to C-N stretching of any R-N=C=S

SEM

Interactions during the biosynthesis process. Leads to the production of NPS that take different morphology and structures and identify the morphology are done during FESM.

The morphology of NPs in the images were multiple shapes, (rock, Clusters, Semi spherical, semi cubic).

It was pH between (5, 6) acid, perhaps it is the reason for these morphologies grain size of SEM Images greater than crystal size at XRD, the reason: (XRD) depends on size or measure particles without taking the number of defects in the crystal.

Anti bacterial activity of yeast extract synthesized AgNPS

We measured the inhibitory effect of different concentration of AgNps against *Staphylococcus epidermidis* and *Escherichia coli*. As shown in table 1.

References

Table 1: Relation between concentration AgNps and Inhibition zone.

Concentration	Bacterial type	Inhibition zone
100 $\mu\text{g}/\mu\text{l}$	<i>Staphylococcus epidermidis</i>	21 mm
50 $\mu\text{g}/\mu\text{l}$		20 mm
25 $\mu\text{g}/\mu\text{l}$		24 mm
100 $\mu\text{g}/\mu\text{l}$	<i>Escherichia coli</i>	11 mm
50 $\mu\text{g}/\mu\text{l}$		12 mm
25 $\mu\text{g}/\mu\text{l}$		13 mm

- Akerman, M.E., W.C. Chan, P. Laakkonen, S.N. Bhatia and E. Ruoslahti (2002). Nanocrystal targeting *in vivo*. *Proc. Nat. Acad. Sci., USA* **99**: 12617-12621.
- Atiyeh, B.S., M. Costagliola, S.N. Hayek and S.A. Dibo (2007). Effect of silver on burn wound infection control and healing: review of the literature. *Burns.*, **33**:139-148.
- Bruchez, J., M.M. Moronne, P. Gin, S. Weiss and A.P. Alivisatos (1998). *Semiconductor nanocrystals as fluorescent biological labels Science.*, **281**: 2013-2016.
- Cao, G. (2004). Nanostructures and nanomaterials: synthesis, properties and applications. Imperial College Press, London.
- Cao, Y.C., R. Jin and C.A. Mirkin (2002). Nanoparticles with Raman spectroscopic fingerprints for DNA and RNA detection. *Science.*, **297**: 1536-1540.
- Chernousova, S. and M. Epple (2013). Silver as antibacterial agent: Ion, nanoparticle and metal. *Angew. Chem. Int. Ed.*, **52**: 1636-1653. (CrossRef) (PubMed).
- Chu, C.S., A.T. McManus, B.A. Pruitt and A.D. Mason (1988). Therapeutic effects of silver nylon dressing with weak direct current on *Pseudomonas aeruginosa* infected burn wounds. *J. Trauma.*, **28**:1488-1492.
- Deitch, E.A., A. Marin, V. Malakanov and J.A. Albright (1987). Silver nylon cloth: *in vivo* and *in vitro* evaluation of antimicrobial activity. *J. Trauma.*, **27**: 301-304.
- Gurunathan, S., K. Kalishwaralal, R. Vaidyanathan, D. Venkataraman, S.R.K. Pandian, J. Muniyandi, N. Hariharan and S.H. Eom (2009). Biosynthesis, purification and characterization of silver nano particles using *Escherichia coli*. *Colloids Surf.*, **B74(1)**: 328-335.
- Gurunathan, S., K.I. Lee, K. Kalishwaralal, S. Sheikpranbabu, R. Vaidyanathan and S.H. Eom (2009). Antiangiogenic properties of silver nanoparticles. *Biomaterials.*, **30**: 6341-6350.
- Gurunathan, S., K. Kalishwaralal, R. Vaidyanathan, D. Venkataraman, S.R. Pandian, J. Muniyandi, N. Hariharan and S.H. Eom (2009). Biosynthesis, purification and characterization of silver nanoparticles using *Escherichia coli*. *Colloids Surf. B. Biointerfaces.*, **74**: 328-335. (CrossRef) (PubMed).
- Gurunathan, S., J.H. Park, J.W. Han and J.H. Kim (2015). Comparative assessment of the apoptotic potential of silver nanoparticles synthesized by *Bacillus tequilensis* and *Calocybe indica* in MDA-MB-231 human breast cancer cells: Targeting p53 for anticancer therapy. *Int. J. Nanomed.*, **10**: 4203-4222. (CrossRef) (PubMed).
- Gwinn, M.R. and V. Vallyathan (2006). Nanoparticles: health effects: pros and cons. *Environ. Health Persp.*, **114**: 1818-1825.
- Kalishwaralal, K., E. Banumathi, S.B.R.K. Pandian, V. Deepak, J. Muniyandi and S.H. Eom (2009). Silver nanoparticles inhibit VEGF induced cell proliferation and migration in bovine retinal endothelial cells. *Colloids Surf B.*, **73**: 51-7.
- Kaushik Roy, C.K. Sarkar and C.K. Ghosh (2015). Photocatalytic activity of biogenic silver nanoparticles synthesized using yeast (*Saccharomyces cerevisiae*) extract Authors Authors and affiliations., November 2015, **5(8)**: 953-959.
- Law, N., S. Ansari, F.R. Livens, J.C. Renshaw and J.R. Lloyd (2008). The formation of nano-scale elemental silver particles *via* enzymatic reduction by *Geobacter sulfurreducens*. *Appl. Environ. Microbiol.*, **74**: 7090-7093.
- Li, L., J. Hu, W. Yang and A.P. Alivisatos (2001). Band gap variation of size-and shape-controlled colloidal CdSe quantum rods. *Nano Lett.*, **1**: 349-351. (Cross Ref).
- Li, W.R., X.B. Xie, Q.S. Shi, H.Y. Zeng, Y.S. Ou-Yang and Y.B. Chen (2010). Antibacterial activity and mechanism of silver nanoparticles on *Escherichia coli*. *Appl. Microbiol. Biotechnol.*, **8**: 1115-1122. (CrossRef) (PubMed).
- Margraff, H.W. and T.H. Covey (1977). A trial of silver-zinc-allantoin in the treatment of leg ulcers. *Arch. Surg.*, **112**: 699-704.
- Mukherjee, P., A. Ahmad, D. Mandal, S. Senapati, S.R. Sainkar, M.I. Khan, P. Renu, P.V. Ajaykumar, M. Alam and R. Kumar *et al.*, (2001). Fungus-mediated synthesis of silver nanoparticles and their immobilization in the mycelial matrix: A novel biological approach to nanoparticle synthesis. *Nano Lett.*, **1**: 515-519. (CrossRef).
- Mullen, M.D., D.C. Wolf, F.G. Ferris, T.J. Beveridge, C.A. Flemming and G.W. Bailey (1989). Bacterial sorption of heavy metals. *Appl. Environ. Microbiol.*, **55**: 3143-3149.
- Sharma, V.K., R.A. Yngard and Y. Lin (2009). Silver nanoparticles: Green synthesis and their antimicrobial activities. *Adv. Colloid Interface.*, **145**: 83-96. (Cross Ref) (PubMed).
- Sheikpranbabu, S., K. Kalishwaralal, D. Venkataraman, S.H. Eom, J. Park and S. Gurunathan (2009). Silver nanoparticles inhibit VEGF-and IL-1B-induced vascular permeability *via* Src dependent path way in porcine retinal endothelial cells. *J. Nanobiotechnol.*, **7**: 8.
- Silver, S. (2003). Bacterial silver resistance: molecular biology and uses and misuses of silver compounds. *FEMS Microbiol. Rev.*, **27**: 341-353.
- Sowbarnika, R., S. Anhuradha and B. Preetha (2018). Enhanced Antimicrobial Effect of Yeast Mediated Silver Nanoparticles Synthesized From Baker's Yeastm. *Int. J. Nanosci. Nanotechnol.*, **14(1)**: March. 2018, 33-42.
- Taniguchi, N. (1974). Proceedings of International Conference on Precision Engineering (ICPE), Tokyo, Japan.