

Antimicrobial activity of biogenic silver nanoparticles, and silver chloride nanoparticles: an overview and comments

Nelson Durán^{1,2,3} · Gerson Nakazato⁴ · Amedea B. Seabra^{2,5,6}

Received: 10 April 2016 / Accepted: 27 May 2016 / Published online: 11 June 2016
© Springer-Verlag Berlin Heidelberg 2016

Abstract The antimicrobial impact of biogenic-synthesized silver-based nanoparticles has been the focus of increasing interest. As the antimicrobial activity of nanoparticles is highly dependent on their size and surface, the complete and adequate characterization of the nanoparticle is important. This review discusses the characterization and antimicrobial activity of biogenic synthesized silver nanoparticles and silver chloride nanoparticles. By revising the literature, there is confusion in the characterization of these two silver-based nanoparticles, which consequently affects the conclusion regarding to their antimicrobial activities. This review critically analyzes recent publications on the synthesis of biogenic silver nanoparticles and silver chloride nanoparticles by attempting to correlate the characterization of the nanoparticles with their antimicrobial

activity. It was difficult to correlate the size of biogenic nanoparticles with their antimicrobial activity, since different techniques are employed for the characterization. Biogenic synthesized silver-based nanoparticles are not completely characterized, particularly the nature of capped proteins covering the nanomaterials. Moreover, the antimicrobial activity of these nanoparticles is assayed by using different protocols and strains, which difficult the comparison among the published papers. It is important to select some bacteria as standards, by following international foundations (Pharmaceutical Microbiology Manual) and use the minimal inhibitory concentration by broth microdilution assays from Clinical and Laboratory Standards Institute, which is the most common assay used in antibiotic ones. Therefore, we conclude that to have relevant results on antimicrobial effects of biogenic silver-based nanoparticles, it is necessary to have a complete and adequate characterization of these nanostructures, followed by standard methodology in microbiology protocols.

✉ Nelson Durán
duran@iqm.unicamp.br

✉ Amedea B. Seabra
amedea.seabra@gmail.com; amedeaseabra@ufabc.edu.br

¹ Institute of Chemistry, Biological Chemistry Laboratory, Universidade Estadual de Campinas, C.P. 6154, Campinas, SP CEP 13083-970, Brazil

² Brazilian Nanotechnology National Laboratory (LNNano-CNPEN), Campinas, SP, Brazil

³ Laboratory of Nanomaterials Synthesis and Biological Interactions (NanoBioss), Institute of Chemistry, Universidade Estadual de Campinas, C.P., 6154, Campinas, SP CEP 13083-970, Brazil

⁴ Department of Microbiology, Universidade Estadual de Londrina, Londrina, PR, Brazil

⁵ Exact and Earth Sciences Department, Universidade Federal de São Paulo, Rua São Nicolau, 210, Centro, CEP, Diadema, SP 09913-030, Brazil

⁶ Center of Natural and Human Sciences, Universidade Federal do ABC, Santo André, SP, Brazil

Keywords Silver nanoparticles · Silver chloride nanoparticles · Biogenic synthesis · X-ray diffraction · Antimicrobial · Antibacterial

Introduction

Silver nanoparticles (Ag° nanoparticles) are the most used nanomaterials due to their potent antimicrobial activity. Biogenic synthesis of Ag° nanoparticles is from a long time known for many researchers, and several excellent reviews were published in the last 6 years (Abbasi et al. 2014; Faramarzi and Sadighi, 2013; de Lima et al. 2012; Durán et al. 2009, 2010, 2011, 2014a; 2015a; Ingál and Chaudhari, 2013; Keat et al. 2015; Mashwani et al. 2015; Mittal et al. 2014; Moghaddam et al. 2015; Natsuki et al. 2015; Rai et al.

2009). Biogenic synthetic routes have several advantages over traditional chemical or physical ones, since they are environmentally friendly technology (Seabra and Durán, 2015). However, the great limitation of biogenic synthesis is the relatively poor reproducibility process. Therefore, the adequate characterization of biogenic Ag⁰ nanoparticles is a crucial step prior to the evaluation of their antimicrobial activity. It is clear from the literature that the basic analysis of the structure of any metal-based nanomaterials is the x-ray diffraction (XRD). Although other techniques including UV–Vis, Fourier transform infrared (FTIR) spectroscopy, x-ray photoelectron spectroscopy (XPS), nuclear magnetic resonance (NMR), scanning electron microscopy (SEM), transmission electron microscopy (TEM), and dynamic light scattering (DLS) are important and complementary, XRD is a crucial technique to the correct identification of the silver-based nanoparticles.

In view of this statement, it is important to compare the silver derivative nanoparticles through XRD, especially in the case of biogenic synthesized Ag⁰ nanoparticles. For this discussion, Fig. 1 represents the XRD pattern from inorganic crystal structure database (ICSD) standards of Ag⁰ nanoparticles (Fig. 1a) and silver chloride nanoparticles (AgCl nanoparticles) (Fig. 1b), which are the most typical compounds found in the final product of biogenic synthesized silver-based nanoparticles. Figure 1a shows the XRD pattern 2θ peak values of around 38.10°, 44.47°, 64.63°, 77.44°, and 81.33° which correspond to the XRD pattern of indexed [111], [200], [220], [311], and [222] facets of Ag⁰ nanoparticles. In the case of AgCl nanoparticles (Fig. 1b), the XRD pattern in 2θ peak values is 27.9°, 32.3°, 46.3°, 55.0°, 57.6°, 67.6°, 74.6°, 76.9°, and 85.7° corresponding to the indexed (111), (200), (220), (311), (222), (400), (331), (420), and (422) facets of AgCl nanoparticles.

Besides typical XRD patterns of silver-based nanoparticles, another important aspect in the characterization of the

nanoparticles is the absorption bands of different silver nanoparticle derivatives. Pure Ag⁰ nanoparticles exhibit a typical plasmon resonance absorption band at the region ~400–500 nm (Fig. 2a), depending on the morphology and size of the nanoparticles (Fig. 2b). In contrast, pure AgCl nanoparticle exhibited only UV band (~250 nm), and not the plasmonic band (~400–500 nm) (Dong et al. 2009). However, upon contamination of AgCl nanoparticles with Ag⁰ nanoparticle clusters, the plasmonic band appeared (Fig. 3). As indicated in Fig. 3, AgCl nanoparticles are photosensitive, yielding Ag⁰ nanoparticles upon ambient light exposition (Trinh et al. 2015). The color of the AgCl nanoparticles suspension was initially opalescent, and upon irradiation, it changed to violet, and finally to yellow-brown (~420 nm). The presence of Ag⁰ layer, produced by the chemical reduction of AgCl, might lead to the appearance of the visible light band absorption. The authors suggested that the direct and indirect band gaps of AgCl nanoparticles were 5.15 eV (240 nm) and 3.25 eV (380 nm), respectively, which avoids AgCl from absorbing light with the wavelength above 380 nm. Furthermore, it is reported that Ag⁰ deposited on AgCl particles exhibited the plasmonic absorption of visible light (Trinh et al. 2015). Therefore, precaution must be taken during the synthesis process of AgCl nanoparticles, since Ag⁰ nanoparticles might be simultaneously produced, as in the case of biogenic routes.

Upon these clarifications, regarding to the characterization of Ag⁰ nanoparticles and AgCl nanoparticles and the differences among them, the next sections of this review present and discuss the most common misunderstanding related to the characterization of the nanostructure of biogenically synthesized Ag⁰ nanoparticle, and consequently, the erroneous conclusion about their antimicrobial activities. Although several reviews reported the importance of biogenically synthesized Ag⁰ nanoparticles for several applications (Abbasi et al. 2014; Faramarzi and Sadighi, 2013; de Lima et al. 2012; Durán et al.

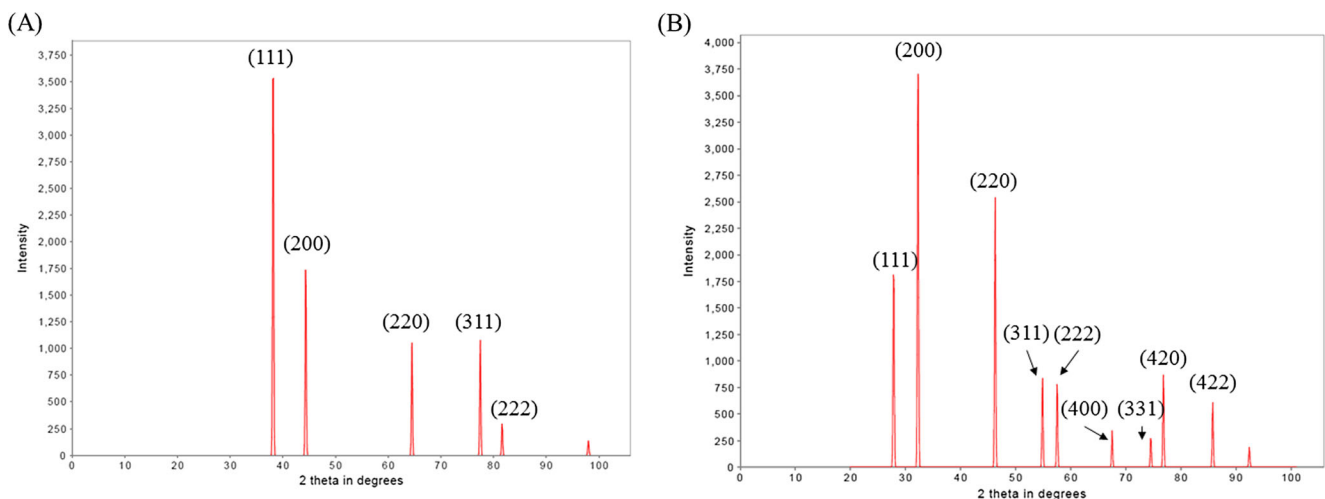


Fig. 1 XRD from International Crystal Structure Database (ICSD) standards: **a** Cubic Ag⁰ nanoparticles Code 64,994. **b** Cubic AgCl nanoparticles Code 64734 (modified from <http://bdec.dotlib.com.br/cliente/login>)

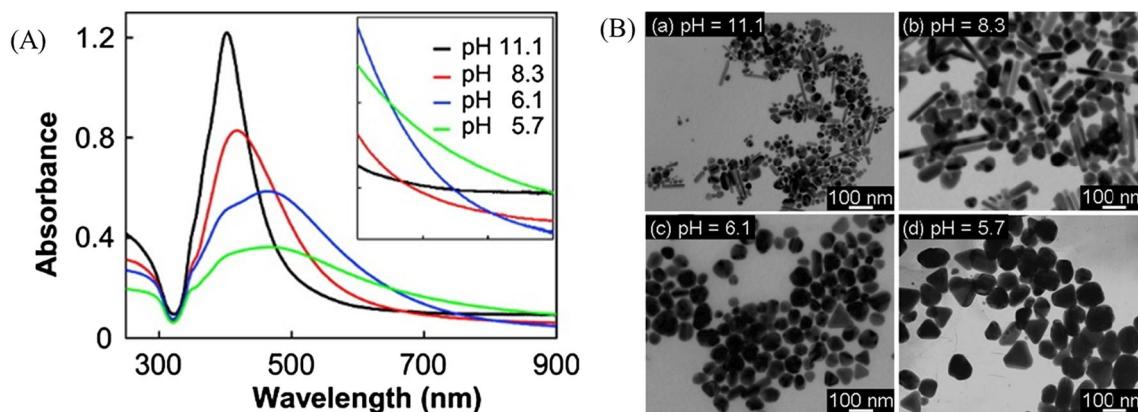


Fig. 2 **a** UV–Vis absorption spectra for the Ag[°] nanoparticles synthesized by citrate at different pH conditions, as indicated in the figure. **b** TEM images of the Ag[°] nanoparticles synthesized by different

pHs, as indicated in the figure. (Modified from reference Dong et al. 2009 with permission from the American Chemical Society)

2009, 2010, 2011, 2014a; 2015a; Ingal and Chaudhari, 2013; Keat et al. 2015; Mashwani et al. 2015; Mittal et al. 2014; Moghaddam et al. 2015; Natsuki et al. 2015; Rai et al. 2009) to our best knowledge, this is the first review to critically discuss the common problems on the erroneous characterization of Ag[°] nanoparticles obtained by biogenic routes. This review does not intend to cover all the literature; however, the aim of this paper is to alert that there are many published papers with confusing characterization of biogenic synthesized Ag[°] nanoparticles and their antibacterial effects, and it aims to be an alert for new developments in this field. In order to obtain relevant results on the antimicrobial effects of biogenic silver-based nanoparticles, it is necessary a complete and adequate characterization of these nanostructures.

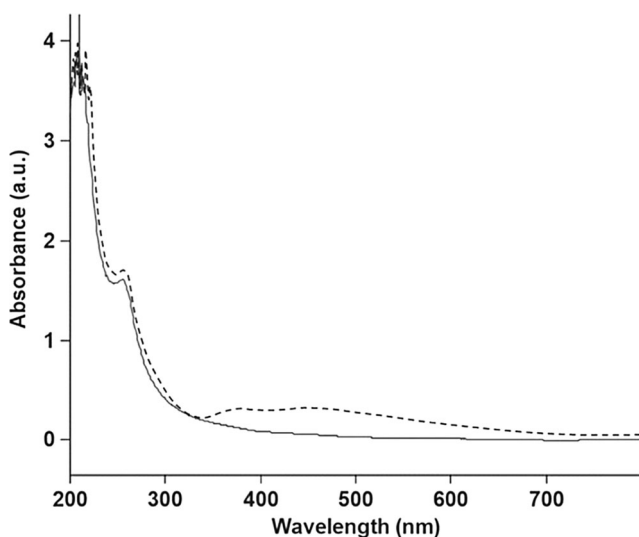


Fig. 3 UV–Vis absorption spectra of AgCl nanoparticle suspension before (*solid line*) and after 5 h of UV irradiation with $\lambda = 254$ nm irradiation (*dashed line*). The irradiation of AgCl nanoparticles led to the formation of the plasmonic band (~ 420 nm) due to the presence of Ag[°] nanoparticles. (Reproduced from reference Trinh et al. 2015 with permission from Vietnam Academy of Science and Technology, under Creative Commons Attribution license of an open access article)

Although several previous studies reported the antibacterial effects of biogenic Ag[°] nanoparticles towards different bacteria, this review highlights the importance to report the antimicrobial effects of silver-based nanoparticles towards at least one standard strain in the microbiology protocols, to allow appropriated comparison among the antibacterial efficacy of different silver nanostructures.

Biogenic synthesis of Ag[°] nanoparticles

The bacterium *Aeromonas* sp. SH10 rapidly reduced silver ions, $[\text{Ag}(\text{NH}_3)_2]^+$, to Ag[°] in the solution at basic pH, and the XRD pattern obtained clearly showed the Bragg reflections corresponding to (111), (200), (220), (311), and (222) sets of lattice planes of Ag[°] nanoparticles (Fu et al. 2006). The same results was reported for the reduction of silver ions to Ag[°] (Kalishwaralal et al. 2008) in the presence of *Bacillus licheniformis*, from *Morganella* sp. (Parikh et al. 2008, 2011), from *Streptomyces hygrosopicus* (Sadhasivam et al. 2010), from *Streptomyces albobrisesolus* (Samundeeswari et al. 2012), from *Shewanella oneidensis* MR-1 (Suresh et al. 2010), from *Bacillus thuringiensis* IS1 strain (Jain et al. 2010), and from *Pseudomonas aeruginosa* (Jeevan et al. 2012).

In the case of biogenic synthesis of Ag[°] nanoparticles by fungi, *Phoma glomerata* (MTCC-2210) (Gade et al. 2014) showed XRD pattern of 2θ peaks related to Ag[°] nanoparticles (Fig. 4a), identical to that from *Aspergillus oryzae* (MTCC 1846) (Phanjom and Ahmed, 2015), similar to those Ag[°] nanoparticles produced by *Vitex negundo* L. leaf extract (Zargar et al. 2011) (Fig. 4b). The same pattern were also published for Ag[°] nanoparticles synthesized from *Phanerochaete chrysosporium* (MTCC 787/ATCC 24725) culture (Vigneshwaran et al. 2006), *Schizophyllum radiatum* HE 863742.1 (Metuku et al. 2014), or from *Aspergillus fumigatus* (Bhainsa and D'Souza, 2006) following the Joint

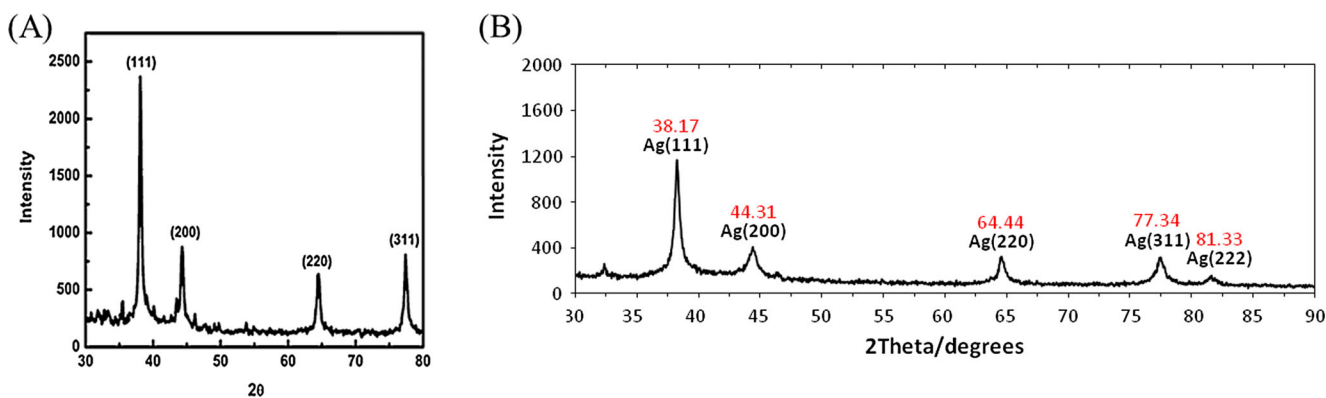


Fig. 4 XRD graphs of biogenic Ag^o nanoparticles synthesized from **a** *Phoma glomerata* (MTCC-2210) and from **b** from *Vitex negundo* L. extract. (Reproduced with modification from reference Gade et al. 2014

with permission from Elsevier and reproduced from reference Zargar et al. 2011 with permission from MDPI, Basel, Switzerland, under Creative Commons Attribution license of an open access article)

Committee on Powder Diffraction Standards (JCPDS; silver file No. 04-0783 or to the ICSD Code 64994).

Plant sources such as *Annona squamosa* leaf broth (Arjunan et al. 2012) produced only Ag^o nanoparticles, the same was observed in the case of biogenic synthesis of Ag^o nanoparticles from aqueous extract of *Gloriosa superba* Linn. (Glory Lily) (Ashokkumar et al. 2013), *Dioscorea bulbifera* (Ghosh et al. 2012), *Sinapis arvensis* seeds (Khatami et al. 2015), gum tragacanth exudate of the bark from *Astragalus gummifer* (Fabaceae family) (Kora and Arunachalam, 2012), Gum ghatti from *Anogeissus latifolia* (Kora et al. 2012), *Medicago sativa* seed exudates (Lukman et al. 2011), from *Cinnamomum zeylanicum* bark extract (Sathishkumar et al. 2009), from extract of *Lawsonia inermis* (Henna) (Gupta et al. 2014), and from latex of *Jatropha curcas* (Bar et al. 2009).

Baker's yeast clearly produced Ag^o nanoparticles, as revealed by the XRD pattern of the obtained nanoparticle (Jha et al. 2008). The same results were reported from yeast strain from an acid mine drainage in Portugal (Mourato et al. 2011), and from yeast species (strain MKY3), which were isolated from garden soil (Kowshik et al. 2003).

Biogenic synthesis of AgCl nanoparticles

Apparently, the formation of AgCl nanoparticles is associated with the biosynthesis of Ag^o nanoparticles, thus it is important to correctly distinguish the XRD patterns between those two nanoparticles. In this direction, this section presents relevant reports on the biogenic synthesis of AgCl nanoparticles and their XRD characterization.

Using the *Bacillus subtilis* MTCC 3053, Paulkumar et al. (2013) demonstrated the biogenic synthesis of AgCl nanoparticles, in the total absence of Ag^o nanoparticles (Fig. 5). The XRD spectrum of silver chloride nanoparticles synthesized by *B. subtilis* exhibits strong peaks at 2 θ values corresponding to (111), (200), (220), (311), (222), (400), (331), (420), and (422)

planes, respectively (Fig. 5A). The XRD spectrum was in accordance with the standard AgCl nanoparticle from Joint Committee on Powder Diffraction Standards (JCPDS file no. 85–1355). Figure 5B shows the energy dispersive x-ray analysis (EDAX) of the AgCl nanoparticles demonstrating the presence of silver and chloride. Indeed, the quantitative percentage of silver and chloride atoms is represented in Fig. 5c. AgCl nanoparticles showed spherical-shaped morphology with average size in the range of 20–60 nm, as demonstrated by SEM analysis (Fig. 5d). Figure 5e shows the UV–Vis spectra recorded during the formation of AgCl nanoparticles from 6 to 48 h of monitoring (Paulkumar et al. 2013). Curiously, Fig. 5e clearly shows the presence of absorption band at around 400 nm, the authors attributed this band due to the presence of AgCl nanoparticles. However, the presence of absorption band at ~400 nm is not expected for AgCl nanoparticles, in contrast, this band indicates the formation of Ag^o nanoparticles (plasmonic band) (Fig. 3). It can be assumed that for UV–Vis analysis, the author measured the real-time biogenic formation of AgCl nanoparticles (from 6 to 48 h), and Ag^o nanoparticles were also yielded, leading to the appearance of the absorption band at ~400 nm. It can be assumed that for UV–Vis analysis, the samples were not washed. In contrast, for XRD analysis, Ag^o nanoparticles were removed by washing/purification processes; therefore, the XRD pattern corresponded solely to the formation of AgCl nanoparticles, with no indication of the presence of Ag^o nanoparticles. Hence, biogenic synthesis of AgCl nanoparticles by *B. subtilis* led to the formation of Ag^o nanoparticles (as indicated by UV–Vis analysis, Fig. 5e), and upon AgCl purification, the Ag^o nanoparticles were efficiently removed, as indicated by the XRD analysis (Fig. 5a). Therefore, caution must be taken with the methodology of biogenic synthesis of AgCl nanoparticles, in particular with the purification process and adequate characterization and discussion of the results in order to avoid confusing between the characterization of AgCl and Ag^o nanoparticles.

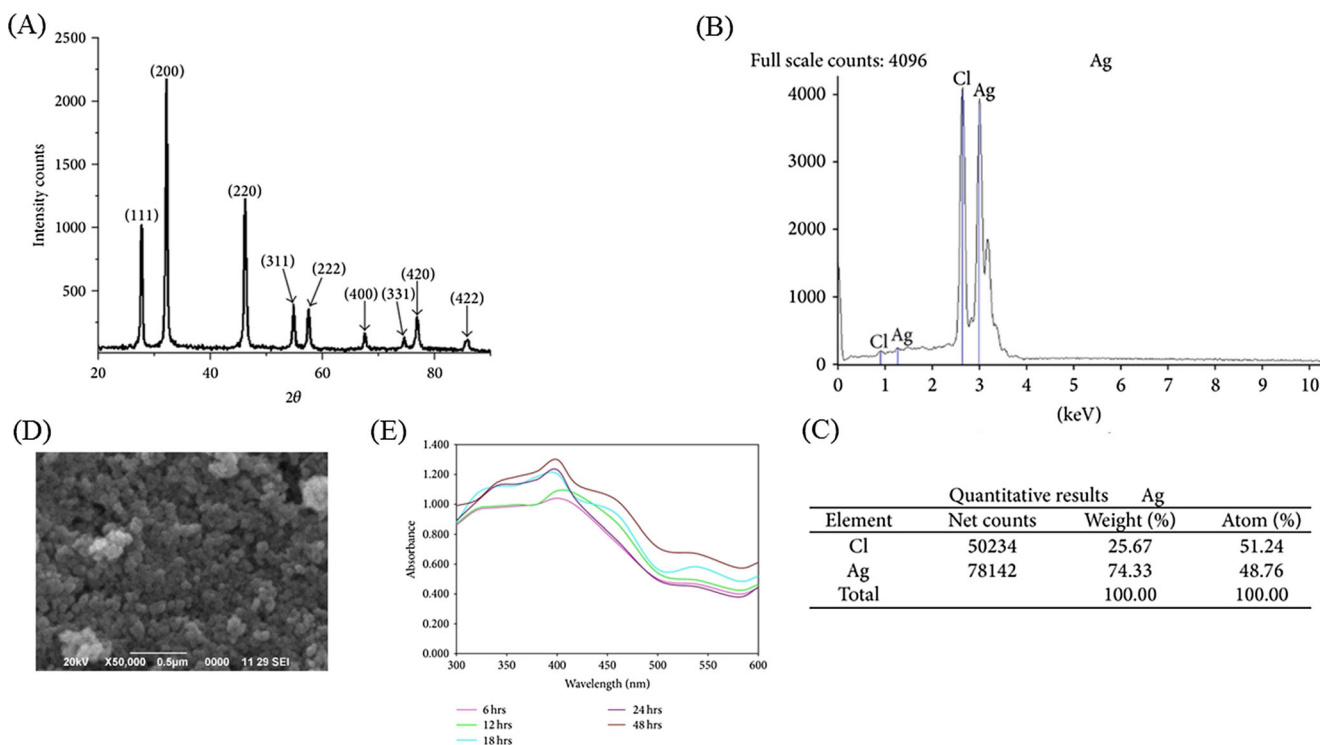


Fig. 5 Characterization of biogenic AgCl nanoparticles synthesized by *B. subtilis*. **a** XRD pattern. **b, c** EDAX analysis. **d** SEM image showing spherical-shaped AgCl nanoparticles. The particle size was characterized in the range of 20–60 nm for *B. subtilis* (scale bar = 500 nm). **e** UV–Vis spectra recorded during the formation of AgCl nanoparticle. Silver nitrate

Mokhtari et al. (2009) reported that *Klebsiella pneumoniae* synthesized AgCl nanoparticles as the final sole biogenic product. To justify their findings, the authors cited a reference that correctly demonstrated the characterization of chemically synthesized AgCl nanoparticles (Potiyaraj et al. 2007). Unfortunately, it was erroneously designated that the 2θ scale for the XRD pattern published by Mokhtari et al. (2009). In this case, the UV–Vis of as-prepared AgCl nanoparticles showed clearly no plasmon band before light irradiation, as shown in Fig. 3.

Synthesis of AgCl nanoparticles by using *Rosa macdub* petal extract was also reported by Shete et al. (2014), in which the XRD pattern clearly showed the presence of AgCl nanoparticles and the absence of Ag⁰ nanoparticles. However, the UV–Vis spectra showed the presence of the plasmon absorption band, which is assigned to the presence of Ag⁰ nanoparticles (probably the concentration of Ag⁰ nanoparticles was very low, and not detectable by XRD analysis, or AgCl nanoparticles were purified for XRD analysis and not for UV–Vis analysis), as reported by Wang et al. (2008). Therefore, attention should be observed in the adequate purification process of the biogenically obtained nanoparticles, and in the correct form to analyze the different techniques for nanoparticle characterization. Adequate discrimination between AgCl and Ag⁰ is required.

Micrococcus luteus (Vimalanathan et al. 2013) was able to produce Ag⁰ nanoparticles; however, the XRD pattern

at 1 mM was added to *B. subtilis* and incubated at different growth periods. (Reproduced with modifications from reference Paulkumar et al. 2013 with permission from Hindawi Publishing Corporation, under Creative Commons Attribution license of an open-access article)

exhibited only the formation of AgCl nanoparticles, in the absence of Ag⁰ nanoparticles. Similar erroneous interpretation was reported by Roy et al. (2015), which used *Saccharomyces cerevisiae* to biogenically produced AgCl nanoparticles, as revealed by XRD analysis; however, the authors assumed the formation of Ag⁰ nanoparticles (Roy et al. 2015). In a similar manner, *Streptomyces* sp. LK3 (Karthik et al. 2014) apparently produced AgCl nanoparticles, and not Ag⁰ nanoparticles, as suggested by the authors. Therefore, from the revised literature, it is clear that several papers published erroneous interpretation regarding to the characterization of biogenic synthesized Ag⁰ and AgCl nanoparticles.

Biogenic synthesis of Ag⁰ nanoparticles/AgCl nanoparticles

The next section discusses some relevant publications of biogenic synthesis of both Ag⁰ nanoparticles and AgCl nanoparticles, probably as Ag/AgCl nanocomposite.

Fungi

In the case of *Penicillium brevicompactum* WA 2315, it was suggested the exclusively formation of Ag⁰ nanoparticles by

XRD pattern (Shaligram et al. 2009). However, the pattern does not correspond solely to Ag⁰ nanoparticles; probably, there is a mixture of both Ag⁰ and AgCl nanoparticles, as following the JCPDS or ICSD standards. Dhanasekaran and Thangaraj (2013) reported the biogenic synthesis of Ag⁰ nanoparticles from *Agaricus bisporus* acting on *Culex* mosquito larvae, however, the XRD pattern revealed the presence of both Ag⁰ nanoparticles and AgCl nanoparticles. Therefore, it is not possible to assign if the effective antimicrobial action of the nanomaterial against larvae was due to Ag⁰ nanoparticles or to AgCl nanoparticles, or the combination of both nanoparticles.

Li et al. (2012) reported the biogenic synthesis of Ag⁰ nanoparticles from *Aspergillus terreus* (BC0603), and for the crystalline nature of the Ag⁰ nanoparticles intense XRD peaks were observed. However, the authors pointed out that some of the intense diffraction peaks observed were related to the presence of AgCl nanoparticles. The authors attributed the presence of AgCl nanoparticles to the addition of chloride ions involved during the preparation of the cell filtrate. Similarly, Vanaja et al. (2015) demonstrated XRD patterns of both Ag⁰ and AgCl nanoparticles upon biogenic synthesis by using *Aspergillus niger*, *Aspergillus pseudodeflectus*, and the synthesized nanoparticles were found to be effective against *Malassezia furfur* (dandruff diseases). The authors attributed the formation of exclusively Ag⁰ nanoparticles, although XRD analysis showed mainly the formation of AgCl nanoparticles. Therefore, AgCl nanoparticles were effective against *Malassezia*, not Ag⁰ nanoparticles, as concluded by the authors (Mathew et al. 2015). Vigneshwaran et al. (2007) produced both nanoparticles, Ag⁰ and AgCl nanoparticles from *Aspergillus flavus* (NCIM 650). Gade et al. (2011) reported that *Phoma sorghina* was capable of synthesizing the Ag⁰ nanorods extracellularly. However, negligible amounts of AgCl were observed in the XRD pattern.

Patil et al. (2011) reported that *A. flavus* challenged with silver nitrate solution accumulated Ag⁰ nanoparticles on the surface of its cell wall. However, the XRD pattern revealed that Ag⁰ nanoparticles are in minor proportion in comparison with AgCl nanoparticles, and other impurities. *Cochliobolus lunatus* produced Ag⁰ nanoparticles; however, AgCl nanoparticles and probably silver oxides were also produced, as demonstrated by the XRD analysis (Salunkhe et al. 2011). The formation of AgCl nanoparticles and silver oxides was not discussed in the publication (Salunkhe et al. 2011). *Fusarium oxysporum*, isolated from mangrove soil samples, showed the presence of only Ag⁰ nanoparticles although no complete pattern was analyzed (Selvi and Sivakumar, 2012). However, Gaikwad et al. (2013) showed that *F. oxysporum*, from infected plant materials such as fruits, vegetable, and food collected from different places of India, exhibited by its XRD analysis mainly Ag⁰ nanoparticles, and a very small amount of AgCl nanoparticles. Similarly, Husseiny et al.

(2015) produced Ag⁰ nanoparticles from *F. oxysporum* f. sp. *lycopersici*; however, XRD pattern showed also the presence of AgCl nanoparticles. Soni and Prakash (2011) demonstrated the presence of Ag⁰ nanoparticles together with silver oxides mixtures and other impurities from *F. oxysporum* Schldtl 2480 and from *Chrysosporium tropicum* Carmichael-2828 strains. *Penicillium atramentosum* KM was suggested to be able to produce Ag⁰ nanoparticles; however, the XRD pattern represented a mixture of silver oxide nanoparticles and not Ag⁰ nanoparticles (Sarsar et al. 2015). *Aspergillus foetidus* (Roy and Das 2015) demonstrated mainly the presence of Ag⁰ nanoparticles; however, traces of AgCl nanoparticles were also observed in the XRD pattern and not discussed in the publication.

Taken together, from the revise literature, there is a misunderstanding in the XRD analysis of biogenically synthesized Ag⁰ and AgCl nanoparticles. It is important to correctly attribute the presence of Ag⁰ and/or AgCl nanoparticles in order to investigate the antimicrobial action of the obtained nanoparticles in a proper manner.

Bacteria

Chauhan et al. (2013) reported that the culture supernatant *Streptomyces* sp. JAR1 was able to synthesize Ag⁰ nanoparticles; however, the XRD pattern clearly revealed the presence of AgCl nanoparticles that was not discussed by the authors. In similar manner, *Streptomyces* sp. ERI-3 produced both Ag⁰ nanoparticles and AgCl nanoparticles, as assayed by XRD analysis, and this fact was not discussed by the authors in their publication (Zonooz and Salouti 2011). The same mistake was observed for the biogenic formation of both AgCl and Ag⁰ nanoparticles by *Brevibacterium frigoritolerans* DC2, and not appropriated discussed in the publication (Singh et al. 2015). Therefore, bacteria may lead to the synthesis of both Ag⁰ and AgCl nanoparticles, and appropriate XRD interpretation is necessary.

Plants

As in the case of fungi and bacteria, plants might result in the formation of both Ag⁰ and AgCl nanoparticles; therefore, caution must be taken to the correct interpretation of the nanomaterial characterization. Gopinath et al. (2013) reported the synthesis of AgCl nanoparticles by leaf extract of *Cissus quadrangularis* Linn, and the crystalline nature of the synthesized AgCl nanoparticles was confirmed using XRD analysis showing the typical peaks at 2 θ planes of the cubic AgCl nanoparticles (ICDD file no. 00-001-1013; ICSD Code 64734). However, the authors demonstrated a minor production of Ag⁰ nanoparticles, as shown by the peaks that

correspond to (111), (200), and (220) planes for cubic Ag⁰ nanoparticles (ICDD file no. 00-087-0718 or ICSD Code 64, 994). Similarly, *Satureja hortensis* L produced Ag⁰ nanoparticles together with AgCl nanoparticles; however, this issue was not discussed by the authors (Afshar P, Sedaghat 2016). Awwad et al. (2015a,b) reported the formation of Ag/AgCl nanoparticles by *Albizia julibrissin* flowers extract. *Murraya koenigii* leaves extract produced mainly Ag⁰ nanoparticles; however, the presence of AgCl was also evident from XRD pattern of the final silver nanostructured material, and not discussed in the paper (Bonde et al. 2012).

Ibrahim (2015) reported the production of Ag⁰ nanoparticles from banana (*Musa paradisiaca*) peels, as suggested by XRD analysis. However, the XRD pattern demonstrated the presence of extra peaks, which the authors attributed to organic compounds, found in the extract, and responsible for the reduction of silver ions as well as to the stabilization of resultant nanoparticles. Unfortunately, the XRD pattern was clearly due to the presence of AgCl nanoparticles. Similarly, Johnson and Prabu (2015) reported that *Cycas circinalis*, *Ficus amplissima*, *Commelina benghalensis*, and *Lippia nodiflora* produced Ag⁰ nanoparticles; however, in the case of *F. amplissima*, AgCl nanoparticles were clearly produced.

Parthenium hysterophorus extract yielded Ag⁰ nanoparticles; however, AgCl nanoparticles were also produced as assayed by XRD and not discussed in the paper (Kumar 2012), and similar mistake was also reported by *Prosopis farcta* extract (Miri et al. 2015) or *Pongamia pinnata* (L) Pierre (Fabaceae family) plant leaves extract (Naik et al. 2014) or from *Curcuma longa* tuber powder and extract (Sathishkumar et al. 2010). Leela and Vivekanandan (2008) suggested the presence of Ag⁰ nanoparticles in their publication; however, the XRD pattern does not correspond to the Ag⁰ nanoparticle. In the presence of Asiatic Pennywort and *Bryophyllum* leaf extracts, the formation of Ag⁰ nanoparticles was wrongly assigned by XRD technique, and the presence of AgCl nanoparticles was ignored (Saikia et al. 2015). Subarani et al. (2013) reported the formation of Ag⁰ nanoparticles from *Vinca rosea* (L.) (*Catharanthus roseus*); however, some silver oxides probably contaminated nanostructure, as revealed by XRD analysis. The *Opuntia ficus-indica* produced both Ag⁰ nanoparticles and AgCl nanoparticles (Gade et al. 2010).

Ag⁰ nanoparticles were suggested to be obtained by using *Rhinacanthus nasutus* leaf extract; however, the XRD pattern did not correspond to Ag⁰ nanoparticles. Probably, traces of Ag⁰ nanoparticles and some AgCl nanoparticles were presented in the low-quality XRD diffractions pattern reported (Pasupuleti et al. 2013). *Clitoria ternatea* and *Solanum nigrum* extracts were suggested to produce Ag⁰ nanoparticles; however, from the XRD pattern, it is clear the presence of AgCl nanoparticles (Krithiga et al. 2015). Therefore, inadequate XRD analysis occurs during characterization of biogenic synthesized silver-based nanoparticles.

Algae

Azziz et al. (2015) reported the synthesis of Ag⁰ nanoparticles from *Chlorella pyrenoidosa* (NCIM 2738), and the authors cited that the characterization of Ag⁰ nanoparticles was in accordance to the results reported by Yang et al. (2011), in which the peaks at 2θ values were correctly ascribed to the reflection of (111), (200), (220), (311) planes of the face-centered cubic silver, respectively (matched well with the patterns of silver) (JCPDS file No. 04-0783 or ICSD Code 64994). However, Azziz et al. (2015) showed XRD peaks at 2θ values that clearly correspond to AgCl nanoparticles, not to Ag⁰ nanoparticles. Vivek et al. (2011) clearly obtained, from *Gelidiella acerosa*, AgCl nanoparticles and probably traces of Ag⁰ nanoparticles, as stated by the authors, and as shown by XRD patterns and UV–Vis spectra. Aqueous extract of *Sargassum plagiophyllum* produced mainly AgCl nanoparticles, as stated by the authors (Dhas et al. 2014); however, traces of Ag⁰ nanoparticles were also shown by UV–Vis spectra, since pure AgCl nanoparticles did not exhibit plasmon absorption spectra. *Gracilaria crassa* was able to produce Ag⁰ nanoparticles as stated by the authors (Lavakumar et al. 2015); however, presence of AgCl nanoparticles can be observed from the XRD pattern. The authors ignored the presence of AgCl nanoparticles.

The formation of synthesized Ag⁰ nanoparticles by *Spirulina platensis* was suggested by XRD measurements (Namasivayam et al. 2015). The authors stated that the distinct high diffraction peaks at 21.6°, 35.5°, and 43.5° erroneously corresponded to the face-centered cubic structure of crystalline silver nanoparticles. Probably (since the published XRD graphic has low quality), there is the appearance of Ag⁰ nanoparticles at 38°, 44°, and 64°; however, there is mainly the presence of AgCl nanoparticles (monoclinic) (ICSD Code 56543). Therefore, erroneous interpretation of XRD results can be observed for biogenic synthesis of Ag⁰ and AgCl nanoparticles from algae.

Yeasts

Candida diversa strain JA1 was reported to synthesize Ag⁰ nanoparticles together with some AgCl nanoparticles (Chauhan et al. 2014), in a similar manner as described by *Candida lusitaniae* (strain TERM 73 and TERM77) (Eugenio et al. 2016). Indeed, *Candida albicans* produced AgCl nanoparticles as shown by XRD pattern, together with Ag⁰ nanoparticles, and the authors unfortunately did not discuss this pattern (Saminathan 2015).

Enzymes

Kumar et al. (2007) reported the synthesis of Ag⁰ nanoparticles from lysozyme, although the XRD analysis confirmed the

formation of the Ag⁰ nanoparticles, the authors ignored the presence of AgCl nanoparticles, as revealed by XRD. Das et al. (2009) and Kumar et al. (2012) reported the lysozyme-mediated synthesis of Ag⁰ nanoparticles in aqueous solution under controlled pH and light conditions. XRD pattern showed that the reaction of laccase, obtained from *Trametes versicolor*, with silver ions was compatible with the cubic phase of Ag⁰ nanoparticles with diffractions points at planes of the facet centered cubic (FCC) structure (JCPDS file: 65-2871 or ICSD Code 64994) that coexisted with the cubic phase of AgCl planes (JCPDS file: 31-1238 or ICSD Code 64734) (Durán et al. 2014b).

Similarly, Zhao et al. (2015) generated both Ag⁰ and AgCl nanoparticles by *Trichoderma hamatum* extract as shown by its XRD pattern. Plasmonic silver-embedded silver chloride (Ag/AgCl) nano-photocatalysts were prepared in a microbe-free aqueous lysogeny broth (Miller) solution, with sunlight exposure and at room temperature, as shown by XRD analysis. Probably, in situ photo-reduction and photo-accelerated ripening of AgCl colloids were promoted by the attached biomolecular ligands (Shen et al. 2015).

Antibacterial activities of biogenic Ag⁰ nanoparticles, AgCl nanoparticles, and Ag/AgCl nanocomposite

From the revised literature, there is a confusion regarding to the characterization of biogenic synthesized Ag⁰ nanoparticles, AgCl nanoparticles, and Ag/AgCl nanocomposites. This misunderstanding affects the appropriate interpretation of the antimicrobial effects of biogenic silver-based nanomaterials and impairs the establishment of regulatory nanotechnological platform in countries that are using silver nanoparticles for the manufacturing of medical or technical products. All silver-based biogenic nanomaterials (Ag⁰ nanoparticles, AgCl nanoparticles, and Ag⁰/AgCl) are reported to have antimicrobial activity. Tables 1, 2, and 3 summarize the antibacterial activity of biogenic synthesized Ag⁰ nanoparticles, AgCl nanoparticles and Ag⁰/AgCl nanoparticles, respectively. The tables present the conclusion from XRD pattern analysis, the biogenic organism used for the synthesis, the size and zeta potential of the nanoparticles, the bacteria strain employed for the microbiological assay, the minimum inhibitory concentration (MIC) values, and the corresponding reference number.

As can be seen in Tables 1, 2, and 3, the most important parameters to compare the antimicrobial efficacy of a compound are the MIC and/or minimal bactericidal concentration (MBC) (CLSI, 1999) values. However, most of studies reported the antimicrobial effect of silver-based nanomaterials in terms of zone of inhibition, which is a qualitative measurement, and in many cases, the concentration of silver-based nanomaterial is not reported.

While the antimicrobial efficacy of biogenic silver-based nanomaterials is increasing, irregular reporting of both nanomaterial characterization and microbiology parameters continue to limit the development of biogenic silver-based nanomaterials because comparison between reports are difficult. Microbiological results must be reported using standard conventions to allow for appropriate comparison among the therapeutic activity between the biogenic nanomaterials.

From Tables 1, 2, and 3, it can be conclude that MIC values of *Escherichia coli* from different strains were completely different, as shown in the strain BL-21 (Sathishkumar et al. 2009) vs ATCC 25922 (Suresh et al. 2010), in which the MIC values were found to be 59 and 2 µg/mL, respectively. Obviously, this discrepancy depends on the intermediate resistant of the strains. Therefore, this is an indication that it is necessary to use standard strains in order to compare the results in an adequate manner. Indeed, from the results presented in Table 2, it is not possible to compare the antibacterial activity, since the studies used different strains, and unfortunately, MIC values were not reported. Analyzing Tables 1, 2, and 3, it can be observed that very few studies reported MIC values and employed same bacterial strains for the biological tests.

Attempt to correlate the antibacterial activities among the silver-based nanoparticles

This section attempts to compare the microbiological results published in the literature regarding to the antimicrobial effects of biogenic synthesized silver-based nanomaterials (Ag⁰ nanoparticles, AgCl nanoparticles, and Ag/AgCl). Table 4 presents the biogenic silver-based nanostructure material, the size of the nanoparticle, the MIC values towards *E. coli*, *Staphylococcus aureus*, *C. albicans*, *B. subtilis*, *P. aeruginosa*, and the corresponding references. As can be observed, comparison between the nanostructures are complicated since the size of the nanoparticles are reported from different techniques ranging from solid-state measurements (TEM and SEM) to aqueous suspension measurements (DLS). It should be noted that generally, DLS values are found to be higher in comparison with nanoparticle size obtained from TEM/SEM analyses, since in aqueous environment there is the presence of extra hydrate layers on the nanoparticle surface. Therefore, standardization of size measurements of nanoparticles is required. Moreover, in microbiological assays, different strains are selected for the antibacterial activities, and a few papers reported the results in terms of MIC or MBC, in contrast, zone of inhibition is still observed in several publications as the single microbiological assay.

Despite these differences, Table 4 aims to highlight and summarize the published results regarding to antibacterial effects of biogenic silver-based nanomaterials against *E. coli*, *S.*

Table 1 Biogenic Ag⁰ nanoparticles characterized by XRD technique and their antibacterial activities

Nanomaterial characterized by XRD	Biogenic synthetic route	Size/zeta potential	Bacteria or fungi	MIC ($\mu\text{g/mL}$)/ Zone (μg)	Reference
Ag	<i>Lawsonia inermis</i> (plant)	30 nm/–32.8 mV	<i>Candida albicans</i> NCIM-3100	12/20 mm	Gupta et al. (2014)
Ag	<i>Schizophyllum radiatum</i> (fungus)	15 nm/non determined (ND)	<i>Escherichia coli</i> (ND)	ND/14 mm (9.6)	Metuku et al. (2014)
Ag	<i>Astragalus gummifer</i> (gum)	13–28 nm/ND	<i>Staphylococcus aureus</i> (ND)	ND/15 mm (9.6)	Kora and Arunachalam (2012)
			<i>Escherichia coli</i> ATCC 25922	ND/9.5 mm (5.0)	
Ag	<i>Anogeissus latifolia</i> (gum)	27 nm/ND	<i>Staphylococcus aureus</i> ATCC 25923	ND/11.5 mm (5.0)	Kora et al. (2012)
			<i>Escherichia coli</i> ATCC 25922	ND/9 mm (5)	
			<i>Escherichia coli</i> ATCC 35218	ND/8.0 (5)	
Ag	<i>Pseudomonas aeruginosa</i> (bacterium)	50 nm/ND	<i>Staphylococcus aureus</i> ATCC 25923	ND/12.3 (5)	Jeevan et al. (2012)
			<i>Escherichia coli</i> (ND)	ND/18 mm (2)	
Ag	<i>Cinnamon zeylanicum</i> (plant)	30–40 nm/–28 mV	<i>Escherichia coli</i> BL-21	10/ND	Sathishkumar et al. (2009)
Ag	<i>Streptomyces hydroscoppicus</i> (bacterium)	20–30 nm/ND	<i>Escherichia coli</i> KCTC-1682	ND/14 mm (2.1)	Sathishkumar et al. (2010)
			<i>Candida albicans</i> KACC 30068	ND/18 mm (2.1)	
Ag	<i>Streptomyces albogriseolus</i> (bacterium)	16 nm/ND	<i>Escherichia coli</i> MCIM 2809	ND/16.5 (10.7)	Samundeeswari et al. (2012)
			<i>Staphylococcus aureus</i> MCIM 2672	ND/18.5(10.7)	
Ag	<i>Bacillus thuringiensis</i> (bacterium)	10–20 nm/ND	<i>Escherichia coli</i> (Multi-drug resistant MDR human pathogens)	ND/12 mm (50)	Jain et al. (2010)
			<i>Staphylococcus aureus</i> (MDR human pathogens)	ND/10 mm (50)	
Ag	<i>Dioscorea bulbifera</i> (plant)	14 nm/ND	<i>Escherichia coli</i> (ND)	ND/9 mm (20)	Ghosh et al. (2012)
			<i>Staphylococcus aureus</i> (ND)	ND/no effect	
Ag	<i>Vitex negundo</i> (plant)	10–20 nm/ND	<i>Escherichia coli</i> ATCC 25922	ND/12 mm (2.1)	Zargar et al. (2011)
			<i>Staphylococcus aureus</i> ATCC 25923	ND/11 mm (2.1)	
Ag	<i>Shewanella oneidensis</i> (algae)	2–11 nm/–12 mV 82 nm	<i>Escherichia coli</i> ATCC 25922	2/ND	Suresh et al. (2010)
			<i>Bacillus subtilis</i> ATCC 9372	0.5/ND	

aureus, *C. albicans*, *B. subtilis*, and *P. aeruginosa*. In the case of Ag⁰ nanoparticles acting on *E. coli*, no correlation between the published papers was possible to make, since different strains and different size measurements were performed. For Ag/AgCl nanoparticles; generally, it can be conclude that MIC values of 2–7 $\mu\text{g/mL}$ were obtained by treating *E. coli* with 20–70 nm nanoparticle sizes in different strains including ATCC 8739 and ATCC 25922. In the case of *S. aureus*, MIC values in the range of 1.9–3.4 $\mu\text{g/mL}$ were found for ~20 nm of Ag/AgCl nanoparticles (ATCC 06538 and 05, 923) (Marcato and Durán 2011; Longhi et al. 2016; Ballottin et al. 2016). Table 4 reveals that higher values of MIC are observed for nanoparticles with higher size values, as measured by DLS technique.

In the case of antimicrobial activity of silver-based biogenic nanoparticles towards different strains (ATCC 26790, ATCC 10231) of *C. albicans*, the nanocomposite Ag/AgCl showed MIC values in the range of 4–12 $\mu\text{g/mL}$ and the size values range from 24–70 nm, as measured by DLS technique. Although, Ag/AgCl synthesized from banana peel no effect was observed against *C. albicans*. Probably, in this case, the protein corona effect (Durán et al. 2015b) (capped protein on the surface of Ag/AgCl nanoparticle) impaired the nanoparticle antimicrobial activity.

In the case of antimicrobial activity against *B. subtilis* and *P. aeruginosa*, different MIC values and different particle sizes are reported. Therefore, these results highlight the importance of standardization of the

Table 2 Biogenic AgCl nanoparticles characterized by XRD technique and their antibacterial activities

Nanomaterial characterized by XRD	Biogenic synthetic route	Size/zeta potential	Bacteria and fungi	MIC ($\mu\text{g/mL}$)/ Zone (μg)	Reference
AgCl	<i>Rosa macdub</i> petal (plant)	35–40 nm /ND	<i>Escherichia coli</i> (human pathogenic)	ND/positive (no values)	Shete et al. (2014)
AgCl	<i>Bacillus subtilis</i> (bacterium)	47 nm/ND	<i>Candida albicans</i> (ND)	ND/17 mm (2.2)	Paulkumar et al. (2013)

Table 3 Biogenic Ag/AgCl nanoparticles characterized by XRD technique and antibacterial activities

Nanomaterial characterized by XRD	Biogenic sythetic route	Size/zeta potential	Bacteria	MIC ($\mu\text{g/mL}$)/ Zone mm (μg)	Reference
Ag/AgCl	<i>Aspergillus niger</i> (fungus)	43–63 nm/ND	<i>Escherichia coli</i> (ND)	ND/ 12 mm (5.4)	Vanaja et al. (2015)
Ag/AgCl	Banana peel (plant)	24 nm/ND	<i>Escherichia coli</i> ATCC 8739 <i>Staphylococcus aureus</i> ATCC 6538 <i>Bacillus subtilis</i> (local) <i>Candida albicans</i> ATCC 120231 <i>Pseudomonas aeruginosa</i> ATCC 9027	5.1/17 mm (2) 3.4/16 mm (2) 6.8/12 mm (2) No effect 1.7/30 mm (2)	Ibrahim (2015)
Ag/AgCl	<i>Cissus quadrangularis</i> Linn (plant)	15–20 nm/ND	<i>Escherichia coli</i> (ND) <i>Staphylococcus aureus</i> (ND)	5/9 mm(ND) 3/8 mm(ND)	Gopinath et al. (2013)
Ag/AgCl	<i>Gracilaria crossa</i> (algae)	123 nm/–34.9 mV	<i>Escherichia coli</i> MTCC 443	ND/38 (20)	Lavakumar et al. (2015)
Ag/AgCl	<i>Prosopis farcta</i> (plant)	11 nm/ND	<i>Escherichia coli</i> PTCC 1431 <i>Staphylococcus aureus</i> PTCC 1399	ND/9.5 mm (1) ND/9.5 mm (1)	Miri et al. (2015)
Ag/AgCl	<i>Spirulina platensis</i> (algae)	27 nm/ND	<i>Staphylococcus aureus</i> (ND) <i>Pseudomonas aeruginosa</i> ATCC 10145	29.2/22 mm (25) 31.2/22 mm (25)	Namasivayam et al. (2015)
Ag/AgCl	<i>Aspergillus terreo</i> (fungus)	1–20 nm/ND	<i>Escherichia coli</i> ATCC 25923 <i>Staphylococcus aureus</i> ATCC 25922	ND/13 mm (20) ND/16 mm (20)	Li et al. (2012)
Ag/AgCl	<i>Streptomyces sp.</i> JAR1 (bacterium)	68 nm/ND	<i>Escherichia coli</i> (ND)	ND/12 mm (5.3)	Chauhan et al. (2013)
Ag/AgCl	<i>Candida diversa</i> A1	93 nm/ND	<i>Escherichia coli</i> (ND) <i>Staphylococcus aureus</i> (ND)	ND/14 mm (1.1) ND/12 mm (1.1)	Chauhan et al. (2014)
Ag/AgCl	<i>Satureja hortensis L</i> (plant)	15 nm/ND	<i>Escherichia coli</i> ATCC 25922 <i>Staphylococcus aureus</i> TCC 25923	MBC 0.37/ND MBC 0.18/ND	Afshar and Sedaghat (2016)
Ag/AgCl	<i>Fusarium oxysporum</i> (fungus)	20–50 nm/–20 mV	<i>Escherichia coli</i> ATCC 25923 <i>Staphylococcus aureus</i> ATCC 05923 <i>Bacillus subtilis</i> ATCC 6633	1.8/ND 1.9/ND 1.9/ND	Marcato and Durán (2011)
Ag/AgCl	<i>Fusarium oxysporum</i> (fungus)	20 nm/–13.2 mV 55 nm	<i>Escherichia coli</i> ATCC 25923 <i>Staphylococcus aureus</i> ATCC 6538	6.6/ND 12/ND	Marcato et al. (2013)
Ag/AgCl	<i>Fusarium oxysporum</i> (fun.)	55 nm/–43 mV	<i>Escherichia coli</i> ATCC 25923	6.7/ND	Unpublished results
Ag/AgCl	<i>Fusarium oxysporum</i> (fun.)	55 nm/–43 mV	<i>Candida albicans</i> ATCC 26790	4.35/ND	Longhi et al. (2016)
Ag/AgCl	<i>Fusarium oxysporum</i> (fun.)	55 nm/–37.1 mV	<i>Candida albicans</i> ATCC 10231	6.5/ND	Unpublished Results
Ag/AgCl	<i>Rhinacanthus nasutus</i> (plants)	22 nm/–18.1 mV 329 nm*	<i>Escherichia coli</i> (ND) <i>Staphylococcus aureus</i> (ND)	ND/17 mm (50) ND/18 mm (50)	Pasupuleti et al. (2013)
Ag/AgCl	<i>Curcuma longa</i> (plant)	20–30 nm/ND	<i>Escherichia coli</i> BL-21	MBC 50/ND	Sathishkumar et al. (2010)
Ag/AgCl	<i>Aspergillus foetidus</i> (fungus)	10–30 nm/ND	<i>Escherichia coli</i> (ND) <i>Staphylococcus aureus</i> (ND)	ND/9 mm (10.7) ND/ 11 mm(10.7)	Roy and Das (2015)
Ag/AgCl	<i>Sargassum plagiophyllum</i> (alga)	18–42 nm/ND	<i>Escherichia coli</i> ATCC 25922	ND/5 mm (20)	Dhas et al. (2014)
Ag/AgCl	<i>Candida lusitanae</i> (yeast)	3–83 nm (strain TERM77)/ND	<i>Staphylococcus aureus</i> ATCC 25923	~30/ND	Eugenio et al. (2016)

characterization and the microbiological assays of silver-based biogenic nanoparticles.

According to Table 4, in the case of *E. coli*, few publications reported the MIC values for silver-based nanoparticles. Among them, Ag⁰ nanoparticles (size of 30–40 nm) obtained from the plant extract of *C. zeylanicum* demonstrated an MIC

value of 10 $\mu\text{g/mL}$, whereas Ag⁰ nanoparticles (size of 2–11 nm) obtained from *S. oneidensis* showed an MIC value of 2 $\mu\text{g/mL}$ (Table 4). This is an indication that small-sized Ag⁰ nanoparticles are more active against the studied bacterium. Similar results were observed for Ag/AgCl nanoparticles, the decrease of MIC values for small-sized nanoparticles

Table 4 Comparison between MIC and size values of biogenically synthesized Ag⁰ nanoparticles and Ag⁰/AgCl

Nanomaterial /Biogenic route	<i>E. coli</i> strain	MIC (μg/mL)	Size (nm)	Reference
Ag/AgCl Banana peel	ATCC 8739	5.1	24 (DLS)	Ibrahim (2015)
Ag/AgCl <i>Cissus quadrangularis</i>	ND	5.0	20 (SEM)	Gopinath et al. (2013)
Ag/AgCl <i>Fusarium oxysporum</i>	ATCC 25922	1.9	~20 (TEM) 82 (DLS)	Marcato and Durán (2011)
Ag/AgCl <i>Fusarium oxysporum</i>	ATCC 25922	6.6	~70 (DLS)	Marcato et al. (2013)
Ag/AgCl <i>Fusarium oxysporum</i>	ATCC 25922	6.7	~70 (DLS)	Unpublished results
Ag/AgCl <i>Candida lusitanae</i>	ND	ND	ND	Eugenio et al. (2016)
Ag <i>Shewanella oneidensis</i>	ATCC 25922	2.0	2–11 (TEM) 82 nm (DLS)	Suresh et al. (2010)
Ag <i>Cinnamom zeylanicum</i>	BL-21	10	30–40 (SEM)	Sathishkumar et al. (2009)
Nanostructure/ Biogenic route	<i>S. aureus</i> strain	MIC (μg/mL)	Size (nm)	Reference
Ag/AgCl Banana peel	ATCC 06538	3.4	24 (DLS)	Ibrahim et al. (2015)
Ag/AgCl <i>Cissus quadrangularis</i>	ND	3.0	20 (SEM)	Gopinath et al. (2013)
Ag/AgCl <i>Fusarium oxysporum</i>	ATCC 05923	1.9	~20 (TEM) 82 (DLS)	Marcato and Durán (2011)
Ag/AgCl <i>Fusarium oxysporum</i>	ATCC 06538	12.0	~70 (DLS)	Marcato et al. (2013)
Ag/AgCl <i>Fusarium oxysporum</i>	ND	ND	ND	Unpublished results
Ag/AgCl <i>Candida lusitanae</i>	ATCC 25923	~30	2–22 (TEM)	Eugenio et al. (2016)
Ag <i>Shewanella oneidensis</i>	ND	ND	ND	Suresh et al. (2010)
Ag <i>Cinnamom zeylanicum</i>	ND	ND	ND	Sathishkumar et al. (2009)
Nanostructure/ Biogenic route	<i>C. albicans</i> strain	MIC (μg/mL)	Size (nm)	Reference
Ag/AgCl / <i>Fusarium oxysporum</i>	ATCC 26790	4.35	~70 (DLS)	Longhi et al. (2016)
Ag/AgCl / <i>Fusarium oxysporum</i>	ATCC 10231	6.5	28 (DLS)	Unpublished results
Ag/AgCl BANANA peel	ATCC 10321	No effect	24 (DLS)	Ibrahim et al. (2015)
Ag <i>Lawsonia inermis</i>	NCIM-3100	12	30 (DLS)	Gupta et al. (2014)
Nanostructure/ biogenic route	<i>B. subtilis</i> strain	MIC (μg/mL)	Size (nm)	Reference
Ag/AgCl /Banana peel	Local	6.8	24 (DLS)	Ibrahim (2015)
Ag/AgCl <i>Fusarium oxysporum</i>	ATCC 6633	1.9	55 (TEM)	Marcato and Durán (2011)
Ag <i>Shewanella oneidensis</i>	ATCC 9372	0.5	82 (DLS)	Suresh et al. (2010)
Nanostructure/ biogenic route	<i>P. aeruginosa</i> strain	MIC (μg/mL)	Size (nm)	Reference
Ag/AgCl /banana peel	ATCC 9027	1.7	24 (DLS)	Ibrahim (2015)
Ag/AgCl / <i>Spirulina platensis</i>	ATCC 10145	31.2	27 (TEM)	Namasivayam et al. (2015)

(Table 4). Unfortunately, in the case of AgCl nanoparticles, the literature does not report enough data to allow comparison, as indicated in Table 2. Therefore, more studies are necessary.

Final remarks

From the selected publications presented and discussed in this review, it can be observed that the biogenic synthesis of silver-based nanomaterials (Ag⁰ nanoparticles, AgCl nanoparticles, and Ag/AgCl nanocomposites) has become an attractive field of research due to the combination of the biological/technological impact of silver nanoparticles and the use of environment-friendly routes in the place of traditional

chemical/physical synthetic protocols. Although much progress in this area (Durán et al. 2016), the characterization of the obtained silver-based biogenic nanostructures contains errors, which are perpetuating in many recent publications. In special, the XRD and UV–Vis analyses of the nanoparticles, reported in several papers, demonstrate incorrect interpretations and conclusions.

Besides the lack of appropriate characterization of biogenic silver-based nanomaterials, the antimicrobial experiments, reported in the literature, make difficult comparisons among the reports. As demonstrated in this review, it is difficult to compare the results reported by different papers, since the measurements of nanoparticle size varies among the reports, as well as the authors used different microorganism strains, and

in most of the cases, the results are not reported in terms of MIC values. In addition, an important point to be considered is the thermodynamic stabilization of Ag⁰ and AgCl nanoparticles, which may interfere in their antibacterial activities (de Lima et al. 2012). In general, bare nanoparticles obtained by chemical/physical routes are unstable and tend to aggregate. To avoid this, the nanoparticle stabilization is achieved by the addition of capping agents via covalent bonds or chemical interactions. In contrast, biogenically synthesized nanoparticles have a superior stability due to the presence of capping agents. Indeed, biogenically synthesized nanoparticles are obtained from the biological resources, which act not only as a reducing agent but also as efficient capping agents (Seabra and Durán, 2015). The presence of capping agents is important to avoid the oxidation of biogenic Ag⁰ nanoparticles to silver ions. During biogenic synthesis of Ag⁰ nanoparticles, oxidation of the metallic nanoparticle is avoided by the presence of reducing/capping agents (Durán et al. 2015a,b).

This review highlights that irregular reporting of both nanomaterial characterization and microbiological parameters impairs the development of biogenic silver-based nanomaterials, since the comparison between the reports are difficult. In contrast, comparable results from different research groups are necessary for the development of regulatory issues in safety norms and procedures. Microbiological results need to be reported using standard conventions allowing appropriate comparison among the therapeutic activity between the biogenic nanomaterials synthesized from different research groups.

For microbiological evaluations, this review highlights the importance to select the target microorganism and the protocol by following the international foundations, such as the Pharmaceutical Microbiology Manual (2014). ORA.007, Version 1.2 date: March 30, 2015 (<http://www.fda.gov/downloads/ScienceResearch/FieldScience/UCM397228.pdf>). According to this manual, the protocol for the evaluation of antimicrobial effectiveness of a certain compound should use cultures with no more than five passages removed from the original stock culture. Moreover, the manual suggests the following standard microorganisms: *C. albicans* (ATCC 10231), *Aspergillus brasiliensis* (ATCC 16404) (formerly *A. niger*), *E. coli* (ATCC 8739), *P. aeruginosa* (ATCC 9027), and *S. aureus* (ATCC 6538). There are other suggestions of regulatory manuals to be followed, including the US Pharmacopeia. US Pharmacopeia Convention, Rockville, MD, USA, 2009, pp. 87–93), and British Pharmacopeia. The stationary office, London, 2011, pp. A348–A355). Determination of MIC values by broth microdilution assays in 96-well microplates (Corning®) as suggested by the Clinical and Laboratory Standards Institute (CLSI, 2015) is the most common assay (Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria That Grow Aerobically. Approved Standards. Tenth Edition. M07-A10 Vol. 35 (2).

Replaces M07-A9. Jan. 2015) (http://shop.clsi.org/c.1253739/site/Sample_pdf/M07A10_sample.pdf), and for MBC (CLSI, 1999) (http://shop.clsi.org/site/Sample_pdf/M26A_sample.pdf).

Finally, as stated in this review, it is necessary an exhaustive adequate characterization and interpretation of the final products obtained from biogenic synthesis of silver-based nanomaterials, followed by comparable reports of their antimicrobial activities.

Acknowledgments Support from INOMAT (CNPq), Brazilian Network on Nanotechnology (MCTI/CNPq), NanoBioSS (MCTI), BIOTEC 28/2013 (CNPq), and FAPESP are acknowledged.

Compliance with ethical standards

Conflict of interest The authors declare that they have no competing interests.

References

- Abbasi E, Milani M, Aval SF, Kouhi M, Akbarzadeh A, Nasrabadi HT, Nikasa P, Joo SW, Hanifehpour Y, Nejati-Koshki K, Samiei M (2014) Silver nanoparticles: synthesis methods, bio-applications and properties. *Crit Rev Microbiol*:1549–7828. doi:10.3109/1040841X.2014.912200
- Afshar P, Sedaghat S (2016) Bio-synthesis of silver nanoparticles using water extract of *Satureja hortensis* L and evaluation of the antibacterial properties. *Curr Nanosci* 12:90–93. doi:10.2174/1573413711666150529202238
- Arjunan NK, Murugan K, Rejeeth C, Madhiyazhagan P, Barnard DR (2012) Green synthesis of silver nanoparticles for the control of mosquito vectors of malaria, filariasis, and dengue. *Vector-borne Zoonotic Dis* 12:262–268. doi:10.1089/vbz.2011.0661
- Ashokkumar S, Ravi S, Velmurugan S (2013) Green synthesis of silver nanoparticles from *Gloriosa superba* L. leaf extract and their catalytic activity. *Spectr Acta Part A: Mol Biomol Spectr* 115:388–392. doi:10.1016/j.saa.2013.06.050
- Awwad AM, Salem NM, W. Khurfan W, Ibrahim Q (2015a) FT-IR spectroscopy and x-ray diffraction characterization of biosynthesized silver/silver chloride nanoparticles. *Arab J Phys Chem* 2:15–19 <http://aphyschem.org/index.php/article/2969880>
- Awwad AM, Salem NM, Ibrahim QM, Abdeen AO (2015b) Phytochemical fabrication and characterization of silver/silver chloride nanoparticles using *Albizia julibrissin* flowers extract. *Adv Mater Lett* 6:726–730. doi:10.5185/amlett.2015.5816
- Aziz N, Faraz M, Pandey R, Shakir M, Fatma T, Varma A, Barman I, Prasad R (2015) Facile algae-derived route to biogenic silver nanoparticles: synthesis, antibacterial, and photocatalytic properties. *Langmuir* 31:11605–11612. doi:10.1021/acs.langmuir.5b03081
- Bar H, Bhui DK, Sahoo GP, Sarkar P, De SP, Misra A (2009) Green synthesis of silver nanoparticles using latex of *Jatropha curcas*. *Colloids Surf A Physicochem Eng Asp* 339:134–139. doi:10.1016/j.colsurfa.2009.02.008
- Bhainsa KC, D'Souza SF (2006) Extracellular biosynthesis of silver nanoparticles using the fungus *Aspergillus fumigatus*. *Colloids Surf B: Biointerfaces* 47:160–164. doi:10.1016/j.colsurfb.2005.11.026
- Bonde SR, Rathod DP, Ingle AP, Ade RB, Gade AK, Rai MK (2012) *Murraya koenigii*-mediated synthesis of silver nanoparticles and its

- activity against three human pathogenic bacteria. *Nanosci Methods* 1:25–36. doi:10.1080/17458080.2010.529172
- Chauhan R, Kumar A, Abraham J (2013) A biological approach to the synthesis of silver nanoparticles with *Streptomyces sp* JAR1 and its antimicrobial activity. *Sci Pharm* 81:607–621. doi:10.3797/scipharm.1302-02
- Chauhan R, Reddy A, Abraham J (2014) Biosynthesis and antimicrobial potential of silver and zinc oxide nanoparticles using *Candida diversa* strain JA1. *Der Pharma Chem* 6:39–47 <http://derpharmachemica.com/archive.html>
- Das R, Jagannathan R, Sharan C, Kumar U, Poddar P (2009) Mechanistic study of surface functionalization of enzyme lysozyme synthesized Ag and Au nanoparticles using surface enhanced Raman spectroscopy. *J Phys Chem* 113:21493–21500. doi:10.1021/jp905806t
- de Lima R, Seabra AB, Durán N (2012) Silver nanoparticles: a brief review of cytotoxicity and genotoxicity of chemically and biogenically synthesized nanoparticles. *J Appl Toxicol* 32:867–879. doi:10.1002/jat.2780
- Dhanasekaran D, Thangaraj R (2013) Evaluation of larvicidal activity of biogenic nanoparticles against filariasis causing *Culex* mosquito vector. *Asian Pac J Trop Dis* 3:174–179. doi:10.1016/S2222-1808(13)60035-3
- Dhas TS, Kumar VG, Karthick V, Angel KJ, Govindaraju K (2014) Facile synthesis of silver chloride nanoparticles using marine alga and its antibacterial efficacy. *Spectrochim Acta Part A: Mol Biomol Spect* 120:416–420. doi:10.1016/j.saa.2013.10.044
- Dong X, Ji X, Wu H, Zhao L, Li J, Yang W (2009) Shape control of silver nanoparticles by stepwise citrate reduction. *J Phys Chem C* 113:6573–6576. doi:10.1021/jp900775b
- Durán N, Marcato PD, Teixeira Z, Durán M, Costa FTM, Brocchi M (2009) State of art of nanobiotechnology applications in neglected diseases. *Curr Nanosci* 5:396–408. doi:10.2174/157341309789378069
- Durán N, Marcato PD, De Conti R, Alves OL, Costa FTM, Brocchi M (2010) Potential use of silver nanoparticles on pathogenic bacteria, their toxicity and possible mechanisms of action. *J Braz Chem Soc* 21:949–959. doi:10.1590/S0103-50532010000600002
- Durán N, Marcato PD, Durán M, Yadav A, Gade A, Rai M (2011) Mechanistic aspects in the biogenic synthesis of extracellular metal nanoparticles by peptides, bacteria, fungi and plants. *Appl Microbiol Biotechnol* 90:1609–1624. doi:10.1007/s00253-011-3249-8
- Durán N, Seabra AB, De Lima R (2014a) Cytotoxicity and genotoxicity of biogenically synthesized silver nanoparticles. In *Nanotoxicology: materials, methodologies, and assessments*. (N. Durán, S.S. Guterres, O.L. Alves, Eds). Springer, Chap 11. 245–263. ISBN 978-3-642-24427-8
- Durán N, Cuevas R, Cordi L, Rubilar O, Diez MC (2014b) Biogenic silver nanoparticles associated with silver chloride nanoparticles (Ag@AgCl) produced by laccase from *Trametes versicolor*. Springer Plus, 3:645. <http://www.springerplus.com/content/3/1/645>
- Durán M, Silveira CP, Durán N (2015a) Non enzymatic biogenic metallic nanoparticles associated to protein catalytic agents: a mini-review. *IET Nanobiotechnol* 9:314–323. doi:10.1049/iet-nbt.2014.0054
- Durán N, Silveira CP, Durán M, Martínez DST (2015b) Silver nanoparticle protein corona and toxicity: a mini-review. *J Nanomater* 13:55. doi:10.1186/s12951-015-0114-4
- Durán N, Durán M, de Jesus MB, Seabra AB, Fávoro WJ, Nakazato G (2016) Silver nanoparticles: a new view on mechanistic aspects on antimicrobial activity. *Nanomedicine: NBM* 12:789–799. doi:10.1016/j.nano.2015.11.016
- Eugenio M, Müller N, Frasés S, Almeida-Paes R, Lima LMTR, Lemgruber L, Farina M, de Souza W, Sant'Anna C (2016) Yeast-derived biosynthesis of silver/silver chloride nanoparticles and their antiproliferative activity against bacteria. *RSC Adv* 6:9893–9904. doi:10.1039/C5RA22727E
- Faramarzi MA, Sadighi A (2013) Insights into biogenic and chemical production of inorganic nanomaterials and nanostructures. *Adv Colloid Interf Sci* 189–190:1–20. doi:10.1016/j.cis.2012.12.001
- Fu M, Li Q, Sun D, Lu Y, He N, Deng X, Wang H, Hunag J (2006) Rapid preparation process of silver nanoparticles by bioreduction and their characterizations. *Chin J Chem Eng* 14:114–117. doi:10.1016/S1004-9541(06)60046-3
- Gade A, Gaikwad S, Tiwari V, Yadav A, Ingle A, Rai M (2010) Biofabrication of silver nanoparticles by *Opuntia ficus-indica*: *in vitro* antibacterial activity and study of the mechanism involved in the synthesis. *Curr Nanosci* 6:370–375. doi:10.2174/157341310791659026
- Gade A, Rai M, Kulkarni S (2011) *Phoma sorghina*, a phytopathogen mediated synthesis of unique silver rods. *Inter J Green Nanotechnol* 3:153–159. doi:10.1080/19430892.2011.628573
- Gade A, Gaikwad S, Durán N, Rai M (2014) Green synthesis of silver nanoparticles by *Phoma glomerata*. *Micron* 59:52–59. doi:10.1016/j.micron.2013.12.005
- Gaikwad SC, Birla SS, Ingle AP, Gade AK, Marcato PD, Rai M, Durán N (2013) Screening of different *Fusarium* species to select potential species for the synthesis of silver nanoparticles. *J Braz Chem Soc* 24:1974–1982. doi:10.5935/0103-5053.20130247
- Ghosh S, Patil S, Ahire M, Kitture R, Kale S, Pardesi K, Cameotra SS, Bellare J, Dhavale DD, Jabgunde A, Chopade BA (2012) Synthesis of silver nanoparticles using *Dioscorea bulbifera* tuber extract and evaluation of its synergistic potential in combination with antimicrobial agents. *Int J Nanomedicine* 7:483–496. doi:10.2147/IJN.S24793
- Gopinath V, Priyadarshini S, Priyadarshini NM, Pandian K, Velusamy P (2013) Biogenic synthesis of antibacterial silver chloride nanoparticles using leaf extracts of *Cissus quadrangularis* Linn. *Mater Lett* 91:224–227. doi:10.1016/j.matlet.2012.09.102
- Gupta A, Bonde SR, Gaikwad S, Ingle A, Gade AK, Rai M (2014) *Lawsonia inermis*-mediated synthesis of silver nanoparticles: activity against human pathogenic fungi and bacteria with special reference to formulation of an antimicrobial nanogel. *IET Nanobiotechnol* 8:172–178. doi:10.1049/iet-nbt.2013.0015
- Husseiny SM, Salah TA, Anter HA (2015) Biosynthesis of size controlled silver nanoparticles by *Fusarium oxysporum*, their antibacterial and antitumor activities. *Beni-Suef University J Basic Appl Sci* 4:225–231. doi:10.1016/j.bjbas.2015.07.004
- Ibrahim HMM (2015) Green synthesis and characterization of silver nanoparticles using banana peel extract and their antimicrobial activity against representative microorganisms. *J Rad Res Appl Sci* 8:265–275. doi:10.1016/j.jrras.2015.01.007
- Ingal AG, Chaudhari AN (2013) Biogenic synthesis of nanoparticles and potential applications: an ecofriendly approach. *J Nanomed Nanotechnol* 4:165. doi:10.4172/2157-7439.1000165
- Jain D, Kachhwaha S, Jain R, Srivastava G, Kothari SL (2010) Novel microbial route to synthesize nanoparticles using spore crystal mixture of *Bacillus thuringiensis*. *Indian J Exp Biol* 48:1152–1156 <http://nopr.niscair.res.in/handle/123456789/10471>
- Jeevan P, Ramya K, Rena AE (2012) Extracellular biosynthesis of silver nanoparticles by culture supernatant of *Pseudomonas aeruginosa*. *Indian J Biotechnol* 11:72–76 <http://www.niscair.res.in/sciencecommunication/researchjournals/rejour/ijbt/ijbt0.asp>
- Jha AK, Prasad K, Kulkarni AR (2008) Yeast mediated synthesis of silver nanoparticles. *Inter J Nanosci Nanotechnol* 4:17–21 http://www.ijnonline.net/article_3994_812.html.
- Johnson I, Prabu HJ (2015) Green synthesis and characterization of silver nanoparticles by leaf extracts of *Cycas circinalis*, *Ficus amplissima*, *Commelina benghalensis* and *Lippia nodiflora*. *Int Nano Lett* 5:43–51. doi:10.1007/s40089-014-0136-1
- Kalishwaralal K, Deepak V, Ramkumarpanidian S, Nellaiah H, Sangiliyandi G (2008) Extracellular biosynthesis of silver

- nanoparticles by the culture supernatant of *Bacillus licheniformis*. Mater Lett 62:4411–4413. doi:10.1016/j.matlet.2008.06.051
- Karthik L, Kumar G, Kirthi AV, Rahuman AA, Rao KVB (2014) *Streptomyces* sp. LK3 mediated synthesis of silver nanoparticles and its biomedical application. Bioprocess Biosyst Eng 37:261–267. doi:10.1007/s00449-013-0994-3
- Keat CL, Aziz A, Eid AM, Elmarzugi NA (2015) Biosynthesis of nanoparticles and silver nanoparticles. Bioresour Bioprocess 2:47. doi:10.1186/s40643-015-0076-2
- Khatami M, Pourseyedi S, Khatami M, Hamidi H, Zaeifi M, Soltani L (2015) Synthesis of silver nanoparticles using seed exudates of *Sinapis arvensis* as a novel bioresource, and evaluation of their antifungal activity. Bioresour Bioproc 2:19. doi:10.1186/s40643-015-0043-y
- Kora AJ, Arunachalam J (2012) Green fabrication of silver nanoparticles by gum tragacanth (*Astragalus gummifer*): a dual functional reductant and stabilizer. J Nanomat 2012: Article ID 869765. http://dx.doi.org/10.1155/2012/869765.
- Kora AJ, Beedu SR, Jayaraman A (2012) Size-controlled green synthesis of silver nanoparticles mediated by gum ghatti (*Anogeissus latifolia*) and its biological activity. Org Med Chem Lett 2:17 http://www.orgmedchemlett.com/content/2/1/17
- Kowshik M, Ashtaputre S, Kharrazi S, Vogel W, Urban KSK, Paknikar KM (2003) Extracellular synthesis of silver nanoparticles by a silver-tolerant yeast strain MKY3. Nanotechnology 14:95–100
- Krithiga N, Rajalakshmi A, Jayachitra A (2015) Green synthesis of silver nanoparticles using leaf extracts of *Clitoria ternatea* and *Solanum nigrum* and study of its antibacterial effect against common nosocomial pathogens. J Nanosci Article ID 928204. http://dx.doi.org/10.1155/2015/928204.
- Kumar DA (2012) Rapid and green synthesis of silver nanoparticles using the leaf extracts of *Parthenium hysterophorus*. A novel biological approach. Inter Res J Pharm 3:169–173 http://www.irjponline.com
- Kumar SA, Abyaneh MK, Gosavi SW, Kulkarni SK, Pasricha R, Ahmad A, Khan MI (2007) Nitrate reductase-mediated synthesis of silver nanoparticles from AgNO₃. Biotechnol Lett 29:439–445. doi:10.1007/s10529-006-9256-7
- Kumar U, Ranjan AK, Sharan C, Hardikarc AA, Pundle A, Poddar P (2012) Green approach towards size-controlled synthesis of biocompatible antibacterial metal nanoparticles in aqueous phase using lysozyme. Curr Nanosci 8:130–140. doi:10.2174/1573413711208010130
- Lavakumar V, Masilamani K, Ravichandiran V, Venkateshan N, Saigopal DVR, Kumar CKA, Sowmya C (2015) Promising upshot of silver nanoparticles primed from *Gracilaria crassa* against bacterial pathogens. Chem Centr J 9:42. doi:10.1186/s13065-015-0120-5
- Leela A, Vivekanandan M (2008) Tapping the unexploited plant resources for the synthesis of silver nanoparticles. Afr J Biotechnol 7:3162–3165 http://www.ajol.info/index.php/ajb/article/view/59252/47550
- Li G, He D, Qian Y, Guan B, Gao S, Cui Y, Yokoyama K, Wang L (2012) Fungus-mediated green synthesis of silver nanoparticles using *Aspergillus terreus*. Int J Mol Sci 13:466–476. doi:10.3390/ijms13010466
- Longhi C, Santos JP, Morey AT, Marcato PD, Durán N, Pinge-Filho P, Nakazato G, Yamada-Ogatta SF, Yamauchi LM (2016) Combination of fluconazole with silver nanoparticles produced by *Fusarium oxysporum* improves antifungal effect against planktonic cells and biofilm of drug-resistant *Candida albicans*. Med Mycol 54:428–432. doi:10.1093/mmy/myv036
- Lukman AI, Gong B, Marjo CE, Roessner U, Harris AT (2011) Facile synthesis, stabilization, and anti-bacterial performance of discrete Ag nanoparticles using *Medicago sativa* seed exudates. J Colloid Interface Sci 353:433–444. doi:10.1016/j.jcis.2010.09.088
- Marcato PD, Durán N (2011) Metal nanoparticles in microbiology. In: M.Rai, Durán N (eds) Biogenic silver nanoparticles: applications in medicines and textiles and their health implications. Springer verlag, Berlin Chap. 11, pp. 249–267
- Marcato PD, Parizotto NV, Martinez DST, Paula AJ, Ferreira IR, Melo PS, Durán N, Alves OL (2013) New hybrid material based on layered double hydroxides and biogenic silver nanoparticles: antimicrobial activity and cytotoxic effect. J Braz Chem Soc 24:266–272 http://dx.doi.org/10.5935/0103-5053.20130034
- Mashwani Z, Khan T, Khan MA, Nadhman A (2015) Synthesis in plants and plant extracts of silver nanoparticles with potent antimicrobial properties: current status and future prospects. Appl Microbiol Biotechnol 99:9923–9934. doi:10.1007/s00253-015-6987-1
- Mathew J, Rathod V, Singh D, Ninganagouda S, Singh AK, Kulkarni P (2015) Enhanced efficacy of ketoconazole coated silver nanoparticles against the fungus *Malassezia furfur* a dandruff causing agent. World J Pharm Pharm Sci 4:1246–1258 http://www.wjpps.com/wjpps_controller/abstract_id/3272
- Metuku RP, Pabba S, Burra S, Hima BNSVSSSL, Gudikandula K, Charya MAS (2014) Biosynthesis of silver nanoparticles from *Schizophyllum radiatum* HE 863742.1: their characterization and antimicrobial activity. Biotech 4:227–234. doi:10.1007/s13205-013-0138-0
- Miri A, Sarani M, Bazaz MR, Darroudi M (2015) Plant-mediated biosynthesis of silver nanoparticles using *Prosopis farcta* extract and its antibacterial properties. Spectrochim Acta Part A: Mol Biomol Spectr 141:287–291. doi:10.1016/j.saa.2015.01.024
- Mittal J, Batra A, Singh A, Sharma MM (2014) Phytofabrication of nanoparticles through plant as nanofactories. Adv Nat Sci: Nanosci Nanotechnol 5:043002. doi:10.1088/2043-6262/5/4/043002
- Moghaddam AB, Namvar F, Moniri M, PMd T, Azizi S, Mohamad R (2015) Nanoparticles biosynthesized by fungi and yeast: a review of their preparation, properties, and medical applications. Molecules 20:16540–16565. doi:10.3390/molecules200916540
- Mokhtari N, Daneshpajouh S, Seyedbagheri S, Atashdehghan R, Abdi K, Sarkar S, Minaian S, Shahverdi HR, Shahverdi AR (2009) Biological synthesis of very small silver nanoparticles by culture supernatant of *Klebsiella pneumoniae*: the effects of visible-light irradiation and the liquid mixing process. Mater Res Bull 44:1415–1421. doi:10.1016/j.materresbull.2008.11.021
- Mourato A, Gadanho M, Lino AR, Tenreiro R (2011) Biosynthesis of crystalline silver and gold nanoparticles by extremophilic yeasts. Bioinorg Chem Appl 2011:Article ID 546074. doi:10.1155/2011/546074.
- Naik BR, Gowreeswari GS, Singh Y, Satyavathi R, Daravath SS, Reddy PR (2014) Bio-synthesis of silver nanoparticles from leaf extract of *Pongamia pinnata* as an effective larvicide on dengue vector *Aedes albopictus* (Skuse) (Diptera: Culicidae). Advan Entomol 2:93–101 http://dx.doi.org/10.4236/ae.2014.22016
- Namasivayam SKR, Jayakumar D, Kumar R, Bharani RSA (2015) Antibacterial and anticancerous biocompatible silver nanoparticles synthesised from the cold-tolerant strain of *Spirulina platensis*. J Coastal Life Med 3:265–272. doi:10.12980/JCLM.3.201514B324
- Natsuki J, Natsuki T, Hashimoto Y (2015) A review of silver nanoparticles: synthesis methods, properties and applications. Inter J Mat Sci Appl 4:325–332. doi:10.11648/j.ijmsa.20150405.17
- Parikh RY, Singh S, Prasad BLV, Patole MS, Sastry M, Shouche YS (2008) Extracellular synthesis of crystalline silver nanoparticles and molecular evidence of silver resistance from *Morganella* sp.: towards understanding biochemical synthesis mechanism. Chem Bio Chem 9:1415–1422. doi:10.1002/cbic.200700592
- Parikh RY, Ramanathan R, Coloe PJ, Bhargava SK, Patole MS, Shouche YS, Bansal V (2011) Genus-wide physicochemical evidence of extracellular crystalline silver nanoparticles biosynthesis by

- Morganella* spp. PLoS One 6:e21401. doi:10.1371/journal.pone.0021401
- Pasupuleti VR, TNVKV P, RA S, SK B, Narasimhulu G, CS R, IA R, SH G (2013) Biogenic silver nanoparticles using *Rhinacanthus nasutus* leaf extract: synthesis, spectral analysis, and antimicrobial studies. Intern J Nanomed 8:3355–3364
- Patil HB, Borse SV, Patil DR, Patil UK, Patil HM (2011) Synthesis of silver nanoparticles by microbial method and their characterization. Arch Phys Res 2:153–158 <http://scholarsresearchlibrary.com/archive.html>
- Paulkumar K, Rajeshkumar S, Gnanajobitha G, Vanaja M, Malarkodi C, Annadurai G (2013) Biosynthesis of silver chloride nanoparticles using *Bacillus subtilis* MTCC 3053 and assessment of its antifungal activity. ISRN Nanomat Article ID 317963. <http://dx.doi.org/10.1155/2013/317963>
- Phanjom P, Ahmed G (2015) Biosynthesis of silver nanoparticles by *Aspergillus oryzae* (MTCC no. 1846) and its characterizations. Nanosci Nanotechnol 5:14–21. doi:10.5923/j.nn.20150501.03
- Potiyaraj P, Kumlangdudsana P, Dubas ST (2007) Synthesis of silver chloride nanocrystal on silk fibers. Mater Lett 61:2464–2466. doi:10.1016/j.matlet.2006.09.039
- Rai M, Yadav A, Gade A (2009) Silver nanoparticles as a new generation of antimicrobials. Biotechnol Adv 27:76–83. doi:10.1016/j.biotechadv.2008.09.002
- Roy S, Das TK (2015) Protein capped silver nanoparticles from fungus: x-ray diffraction studies with antimicrobial properties against bacteria. Int J ChemTech Res 7:1452–1459 [http://sphinxσαι.com/2015/ch_vol7_no3_ICONN/16/PC30%20\(1452-1459\).pdf](http://sphinxσαι.com/2015/ch_vol7_no3_ICONN/16/PC30%20(1452-1459).pdf)
- Roy K, Sarkar CK, Ghosh CK (2015) Photocatalytic activity of biogenic silver nanoparticles synthesized using yeast (*Saccharomyces cerevisiae*) extract. Appl Nanosci 5:953–959. doi:10.1007/s13204-014-0392-4
- Sadhasivam S, Shanmugam P, Yun K (2010) Biosynthesis of silver nanoparticles by *Streptomyces hygroscopicus* and antimicrobial activity against medically important pathogenic microorganisms. Colloids Surf B: Biointerfaces 81:358–362. doi:10.1016/j.colsurfb.2010.07.036
- Saikia D, Gogoi PK, Phukan P, Bhuyan N, Borchetia S, Saikia J (2015) Green synthesis of silver nanoparticles using Asiatic pennywort and *Bryophyllum* leaves extract and their antimicrobial activity. Adv Mater Lett 6:260–264. doi:10.5185/amlett.2015.5655
- Salunkhe RB, Patil SV, Salunkhe BK, Patil CD, Sonawane AM (2011) Studies on silver accumulation and nanoparticle synthesis by *Cochliobolus lunatus*. Appl Biochem Biotechnol 165:221–234. doi:10.1007/s12010-011-9245-8
- Saminathan K (2015) Biosynthesis of silver nanoparticles from dental caries causing fungi *Candida albicans*. Int J Curr Microbiol App Sci 4:1084–1091 www.ijcmas.com
- Samundeeswari A, Dhas SP, Nirmala J, John SP, Mukherjee A, Chandrasekaran N (2012) Biosynthesis of silver nanoparticles using actinobacterium *Streptomyces albogriseolus* and its antibacterial activity. Biotechnol Appl Biochem 59:503–507. doi:10.1002/bab.1054
- Sarsar KMV, Selwal MK, Selwal KK (2015) Biofabrication, characterization and antibacterial efficacy of extracellular silver nanoparticles using novel fungal strain of *Penicillium atramentosum*. J Saudi Chem Soc 19:682–688. doi:10.1016/j.jscs.2014.07.001
- Sathishkumar M, Sneha K, Won SW, Cho C-W, Kim S, Yun Y-S (2009) *Cinnamon zeylanicum* bark extract and powder mediated green synthesis of nano-crystalline silver particles and its bactericidal activity. Colloids Surf B: Biointerfaces 73:332–338. doi:10.1016/j.colsurfb.2009.06.005
- Sathishkumar M, Sneha K, Yun Y-S (2010) Immobilization of silver nanoparticles synthesized using *Curcuma longa* tuber powder and extract on cotton cloth for bactericidal activity. Bioresour Technol 101:7958–7965. doi:10.1016/j.biortech.2010.05.051
- Seabra AB, Durán N (2015) Nanotoxicology of metal oxide nanoparticles. Metals 5:934–975. doi:10.3390/met5020934
- Selvi KV, Sivakumar T (2012) Isolation and characterization of silver nanoparticles from *Fusarium oxysporum*. Int J Curr Microbiol App Sci 1:56–62 <http://ijcmas.com/Archives/vol-1/PDF/K.%20Vanmathi%20Selvi%20and%20T.%20Sivakumar.pdf>
- Shaligram NS, Bule M, Bhambure R, Singhal RS, Singh SK, Szakacs G, Pandey A (2009) Biosynthesis of silver nanoparticles using aqueous extract from the compacting producing fungal strain. Process Biochem 44:939–943. doi:10.1016/j.procbio.2009.04.009
- Shen Z, Liu B, Pareek V, Wang S, Li X, Liu L, Liu S (2015) Sustainable synthesis of highly efficient sunlight driven Ag embedded AgCl photocatalysts. RSC Adv 5:80488–80495. doi:10.1039/C5RA17696D
- Shete S, Shende S, Bhagwat K, Dagade S, Deshpande N, Waghmode S (2014) Green synthesis of silver chloride nanoparticles by using *Rosa macdub* petal extract. Inter J Biosci Nanosci 1:96–99 http://www.ijbsans.com/journal14/oct14/MS-10-14-08_1.pdf
- Singh P, Kim YJ, Singh H, Wang C, Hwang KH, Farh ME-A, Yang DC (2015) Biosynthesis, characterization, and antimicrobial applications of silver nanoparticles. Int J Nanomedicine 10:2567–2577. doi:10.2147/IJN.S72313
- Soni N, Prakash S (2011) Factors affecting the geometry of silver nanoparticles synthesis in *Chrysosporium tropicum* and *Fusarium oxysporum*. Am J Nanotechnol 2:112–121. doi:10.3844/ajnsnp.2011.112.121
- Subarani S, Sabhanayakam S, Kamaraj C (2013) Studies on the impact of biosynthesized silver nanoparticles (AgNPs) in relation to malaria and filariasis vector control against *Anopheles stephensi* Liston and *Culex quinquefasciatus* Say (Diptera: Culicidae). Parasitol Res 112:487–499. doi:10.1007/s00436-012-3158-5
- Suresh AK, Pelletier DA, Wang W, Moon J-W, Gu B, Mortensen NP, Allison DP, Joy DC, Phelps TJ, Doktycz MJ (2010) Silver nanocrystallites: biofabrication using *Shewanella oneidensis*, and an evaluation of their comparative toxicity on gram-negative and gram-positive bacteria. Environ Sci Technol 44:5210–5215. doi:10.1021/es903684r
- Trinh ND, Nguyen TTB, Nguyen TH (2015) Preparation and characterization of silver chloride nanoparticles as an antibacterial agent. Adv Nat Sci: Nanosci Nanotechnol 6 045011. doi:10.1088/2043-6262/6/4/045011
- Vanaja M, Rajeshkumar S, Paulkumar K, Gnanajobitha G, Chitra K, Malarkodi C, Annadurai G (2015) Fungal assisted intracellular and enzyme based synthesis of silver nanoparticles and its bactericidal efficiency. Inter Res J Pharm Biosci 2:8–19 <https://www.researchgate.net/publication/281774783>
- Vigneshwaran N, Kathe AA, Varadarajan PV, Nachane RP, Balasubramanya RH (2006) Biomimetics of silver nanoparticles by white rot fungus, *Phaenerochaete chrysosporium*. Colloids Surf B: Biointerfaces 53:55–59. doi:10.1016/j.colsurfb.2006.07.014
- Vigneshwaran N, Ashtaputre NM, Varadarajan PV, Nachane RP, Paralikar KM, Balasubramanya RH (2007) Biological synthesis of silver nanoparticles using the fungus *Aspergillus flavus*. Mater Lett 61:1413–1418. doi:10.1016/j.matlet.2006.07.042
- Vimalanathan AB, Ernest V, Arumugasamy K, Tyagi MG (2013) Biosynthesis of silver nano-particles by the bacterium *Micrococcus luteus*. Inter J. Appl Biol Pharm Technol 4:1–5 <http://www.ijabpt.org/applied-biology/biosynthesis-of-silver-nanoparticles-by-the-bacterium-micrococcus-luteus.php?aid=3901>
- Vivek M, Kumar PS, Steffi S, Sudha S (2011) Biogenic silver nanoparticles by *Gelidiella acerosa* extract and their antifungal effects. Avicenna J Med Biotech 3:143–148. <https://archive.org/details/pubmed-PMC3558184>
- Wang P, Huang B, Qin X, Zhang X, Dai Y, Wei J, Whangbo MH (2008) Ag@AgCl: a highly efficient and stable photocatalyst active under visible light. Angew Chem 120:8049–8051. doi:10.1002/ange.200802483

- Yang X, Du Y, Li D, Lv Z, Wang E (2011) One-step synthesized silver micro-dendrites used as novel separation mediums and their applications in multi-DNA analysis. *Chem Commun* 47:10581–10583. doi:10.1039/c1cc11374g
- Zargar M, Hamid AA, Bakar FA, Shamsudin MN, Shameli K, Jahanshahi F, Farahani F (2011) Green synthesis and antibacterial effect of silver nanoparticles using *Vitex negundo* L. *Molecules* 16:6667–6676. doi:10.3390/molecules16086667
- Zhao X, Zhang J, Wang B, Zada A, Humayun M (2015) Biochemical synthesis of Ag/AgCl nanoparticles for visible-light-driven photocatalytic removal of colored dyes. *Materials* 8:2043–2053. doi:10.3390/ma8052043
- Zonooz NF, Salouti M (2011) Extracellular biosynthesis of silver nanoparticles using cell filtrate of *Streptomyces* sp. ERI-3. *Scientia Iranica F* 18:1631–1635. doi:10.1016/j.scient.2011.11.029