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Production of Metal Nanoparticles by Agro-Industrial Wastes: A Green Opportunity for Nanotechnology

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The feasibility of producing silver nanoparticles (Ag NPs) using phenolic extracts from agro-industrial wastes as reducing agents was investigated. Phenolic extracts were obtained from bilberry wastes (BW) and spent coffee grounds (SCG) with aqueous ethanol as extraction solvent. Experiments were carried out in batch at 25 °C by adding appropriate amounts of phenolic extracts to a silver nitrate aqueous solution. The formation of Ag NPs was monitored spectrophotometrically by measuring the intensity of the surface plasmon resonance (SPR) band of silver at 415–435 nm. Depending on the process conditions, the synthesis of Ag NPs was completed in 3 to 5 hours. Characterization of the resulting reaction products by XRD, SEM and DLS showed that nanoparticles were formed with a spherical shape and an average size of 10–20 nm. Overall, the results obtained suggest that BW and SCG could be used as a source of reducing agents for the production of metal NPs and that agro-industrial wastes may represent a valid alternative to the use of microorganisms, whole plants or plant parts for the biogenic synthesis of NPs.

1. Introduction

Owing to their unique properties resulting from the exceptionally high surface-to-volume ratio, metal nanoparticles (NPs) have attracted increasing attention in the scientific as well as technological fields. Current methods for the production of NPs are based on the use of hazardous compounds as reducing agents and often require costly devices and instrumentation (Gutiérrez et al., 2012). Although these methods produce NPs quite efficiently, they have a significant environmental impact and the downstream operations needed to achieve a high level of product purity can be expensive and time-consuming. For these reasons, alternative greener routes to the production of NPs, such as biogenic synthesis from bacteria, fungi and plant extracts, are actively being explored (Kulkarni and Muddapur, 2014). Regarding the latter, plant parts such as leaves (Khalil et al., 2013), roots (Suman et al., 2013) and stem bark (Shameli et al., 2012) have been successfully used.

Plant-based production of NPs is very simple and cost-effective (Marchiol, 2012). Furthermore, the size and shape of the resulting NPs can be modulated to some extent by varying the temperature, pH and reaction conditions. Finally, plant extracts may contain NP stabilizers in addition to the reducing agents, so that no chemicals other than the metal source are needed (Kharissova et al., 2013).

Recently, as a further step to the development of fully green and sustainable methods for the production of NPs, attempts have been made to replace plant parts with agro-industrial wastes, e.g., grape pomace (Krishnaswamy et al., 2014), mango (Yang et al., 2014) and citrus (Dauthal and Mukhopadhyay, 2015) peels. However, very few studies have so far been performed on this topic and little is known about the real potential of this approach. In particular, the effects of the waste type or the process conditions on the characteristics of the synthesized NPs are not fully understood.

In this contribution we have investigated the feasibility of using bilberry wastes (BW) and spent coffee grounds (SCG) as a raw material for the green synthesis of silver NPs. BW and SCG are commonly found in Italy and are characterized by the presence of relatively high amounts of phenolic compounds, which are known to be effective reducing agents for the production of metal NPs (Jacob et al., 2008). According to our previous studies, the phenolic content of these materials may reach values above 25 mg GAE/g dw in BW (Zuorro and Lavecchia, 2014) and close to 20 mg GAE/g dw in SCG (Zuorro, 2015). Furthermore, because of the

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differences in their respective plant sources, the phenolic compounds originating from the two wastes can be expected to vary both in type and relative amounts. Thus, by comparing the results obtained using BW and SCG, useful information can be gained on the influence that each waste may have on the properties of NPs.

2. Experimental

2.1 Chemicals and waste materials

Silver nitrate (CAS 7761−88−8, purity 99.8%), gallic acid (CAS 149-91-7) and the Folin-Ciocalteu reagent were purchased from Sigma-Aldrich (Milan, Italy). Ethanol (CAS 64-17-5), hydrochloric acid (CAS 7647-01-0) and sodium carbonate (CAS 497-19-8) were obtained from Carlo Erba (Milan, Italy). All chemicals were reagent grade and used without further purification. Aqueous solutions were prepared with demineralized water (18.2 µS cm^{-1}).

BW were obtained as the solid residue of the processing of fresh Italian bilberries in a household food centrifuge (Moulinex, Italy). SCG were collected from an automatic espresso machine.

2.2 Analytical methods

Moisture content was determined by oven drying at 105 °C to constant weight. Total phenolics were determined by the Folin-Ciocalteu method as described by Zuorro and Lavecchia (2011). The results were expressed as gallic acid equivalents (GAE) per unit volume of liquid using a calibration curve obtained with gallic acid standards.

The absorption spectra of the NP solution were recorded at room temperature in the wavelength range 200–800 nm using a double-beam UV-Vis spectrophotometer (UV-2700, Shimadzu, Japan) and quartz cells of 1-cm path length.

X-ray diffraction (XRD) measurements were carried out on an X’Pert PRO diffractometer (Philips, The Netherlands) with a monochromatic Cu Kα radiation (λ = 0.154 nm) at 1.6 kW (40 kV, 40 mA). Diffraction patterns were recorded in a step-scan mode between 20° and 80° 2θ values, with a step size of 0.04° and a counting time of 16 s per step.

Morphological analyses of Ag NPs were performed by a field-emission scanning electron microscope (Fe-SEM Auriga, Zeiss, Germany). Images were acquired on freshly prepared dehydrated samples with an applied voltage of 2–6 kV.

Dynamic light scattering (DLS) and zeta-potential measurements were made using a Malvern Zetasizer instrument (Malvern Instruments, UK).

2.3 Production of phenolic extracts

Phenolic extracts were obtained from BW and SCG following the procedure described by Zuorro et al. (2014). The extraction was carried out in batch mode using aqueous ethanol of different concentrations (0–100% v/v) as solvent. Briefly, appropriate amounts of solvent and plant material were placed into screw-capped glass flasks. The flasks were thermostated (± 0.1 °C) and magnetically stirred for the required time. Then, an aliquot of the liquid was taken, filtered and assayed for phenolic content. The extraction time was 160 min, the liquid-to-solid ratio was 10 mL g^{-1} and the temperature was 40 °C.

2.4 Synthesis of Ag NPs

For the synthesis of Ag NPs, a known volume of the phenolic extract obtained from BW or SCG was added to a given amount of aqueous $AgNO_3$ solutions in order to achieve the desired silver-to-polyphenol ratio (Ag^+/GAE = 10 mol/mol). The reaction was carried out at 25 °C in thermostated (± 0.1 °C) and magnetically stirred screw-capped vials. The formation of silver NPs was monitored by measuring the intensity of the surface plasmon resonance band in the UV-Vis spectra. Ag NPs were further characterised by XRD, SEM and DLS measurements.

3. Results and discussion

As a first step in the production of Ag NPs using phenolic compounds from BW and SCG as reducing agents, we investigated the influence of solvent composition on the phenolic extraction yields. The results shown in Figure 1 indicate that, for both materials, a maximum extraction yield was achieved with 50% (v/v) ethanol. The concentration of polyphenols in the resulting extracts was 17.4 mmol GAE L^{-1} for SCG and 4 mmol GAE L^{-1} for BW. Phenolic concentrations in extracts from SCG were about 5-fold higher than those from BW, with the exception of the extracts obtained with pure ethanol, where much lower differences were found (2.85 mmol GAE L^{-1} for SCG and 1.96 mmol GAE L^{-1} for BW).

The presence of a maximum in the dependence of the extraction yield on the concentration of aqueous ethanol, lying approximately around 50% ethanol, has also been reported for other plant materials, such as peanut skins (Nepote et al., 2005), kiwi by-products (Sun-Waterhouse et al., 2009), artichoke waste (Zuorro et al., 2014) and olive pomace (Lavecchia and Zuorro, 2015). When interpreting this result, it should be considered that polyphenols encompass a wide range of compounds with different affinities for ethanol and for water. Accordingly, an optimal aqueous ethanol concentration may exist which maximizes the total amount of polyphenols extracted. Factors other than chemical affinity, such as solvent-induced perturbations of solute–matrix interactions or plant swelling, could also be involved but their role in the extraction process remains uncertain (Zuorro and Lavecchia, 2013).

Typical UV-Vis spectra of the solution containing silver nitrate and hydroalcoholic phenolic extracts at different reaction times are displayed in Figure 2. The formation of Ag NPs is clearly evidenced by the appearance of a strong surface plasmon resonance (SPR) band at around 420 nm. The intensity of this band, arising from the collective oscillation of free metal electrons at the NP surface in resonance with the incident light, became apparent after 1-h reaction and increased gradually up to about 5 h, when the reaction was completed. The formation of Ag NPs was accompanied by a colour change of the solution to golden yellow (Figure 3), which can be attributed to the excitation of surface plasmon vibrations in these NPs (Dang et al., 2015).

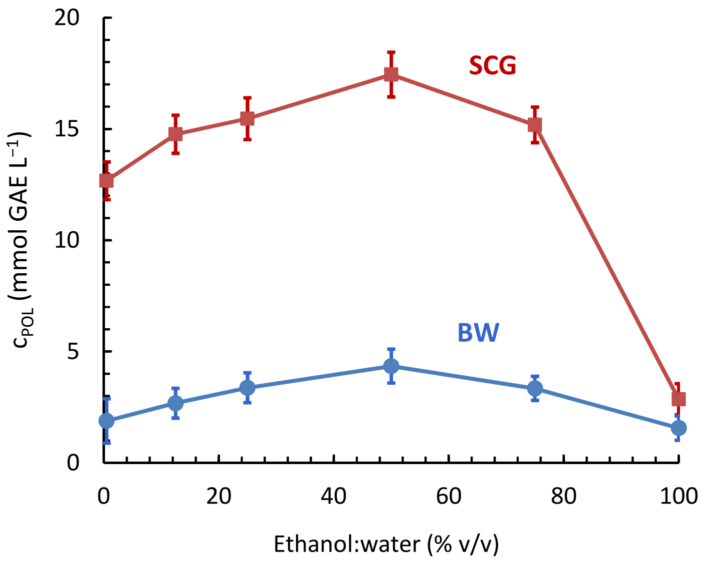

Figure 1: Effect of solvent composition on the extraction of polyphenols from BW and SCG

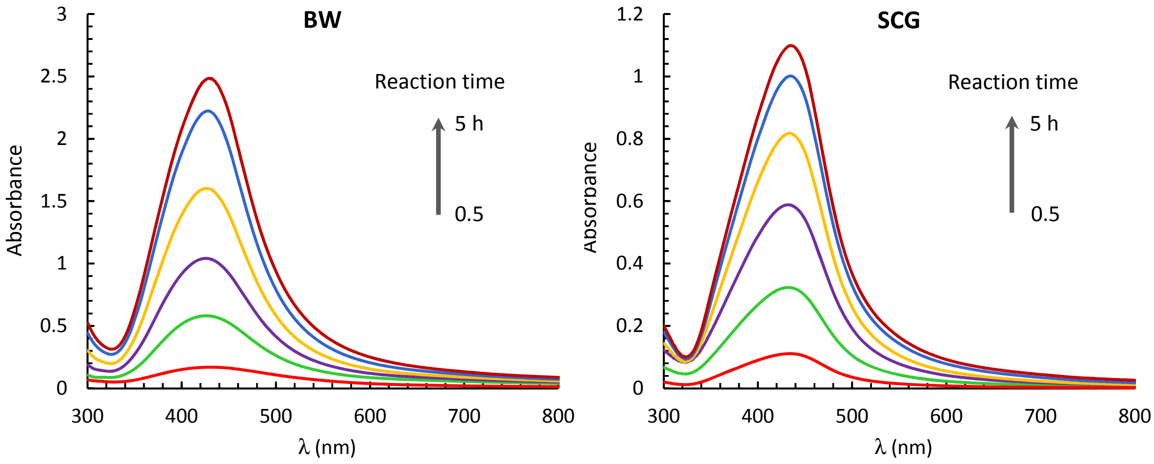

Figure 2: UV-Vis spectra of the reaction mixture containing silver nitrate and 50% aqueous ethanol extracts from BW and SCG (the absorption spectrum of the solution at t = 0 was subtracted from those at time t)

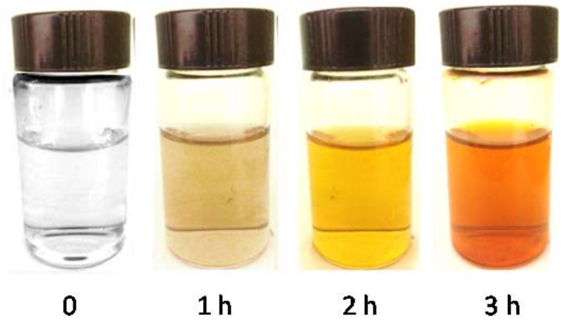

Figure 3: Appearance of the reaction mixture (silver nitrate solution and 50% aqueous ethanol extract from BW) at different times

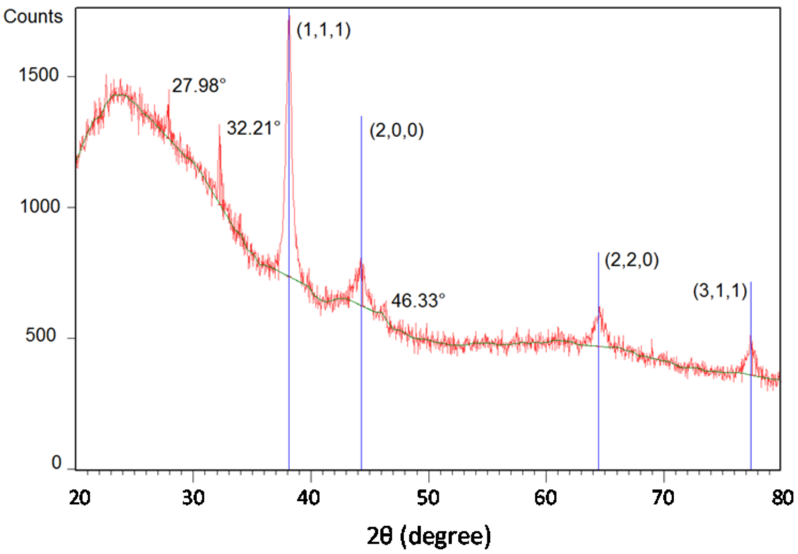

Figure 4: XRD pattern of Ag NPs obtained from 50% hydroalcoholic BW extract

XRD analysis of NP samples provided evidence for their crystalline nature (Figure 4). In the 2θ range 20–80°, four intense peaks at 38.13°, 44.41°, 64.60°, 77.42° were observed, corresponding to the planes (111), (200), (220) and (311) of the face-centered cubic (fcc) structure of metallic silver. The additional peaks at 27.98°, 32.21° and 46.33° were not clearly identified but their presence can be tentatively attributed to the formation on the surface of NPs of a crystalline organic film of polyphenols or other compounds present in the extracts.

Since the shape and size of NPs affect the position and width of the SPR band (Rauwel et al., 2015), a quantitative analysis of the UV-Vis spectra can provide further insight into the characteristics of the synthesized NPs. Table 1 reports the wavelength of maximum absorbance (λ_{max}), the maximum intensity of the SPR band (A_{max}) and the full width at half maximum (FWHM) for NPs produced from BW and SCG.

Examination of the data in Table 1 and inspection of the absorption spectra of Ag NPs reveal that:

a) SPR bands for all samples were highly symmetrical and no other peaks were present at higher wavelengths;
b) the wavelengths of maximum absorbance for NPs from BW extracts were always higher than those from SCG extracts;
c) for both types of extracts, an increase in ethanol concentration from 0 to 50% (v/v) resulted in a slight blue shift of the SPR bands, while a marked red shift was observed when ethanol concentration was increased from 50 to 100% (v/v).

Since a blue shift of the SPR band is indicative of a reduction in the size of NPs (Burda et al., 2005), it can be inferred that smaller Ag NPs are obtained when phenolic extracts in 50% ethanol are used. Furthermore, extracts from SCG seemed to be more effective in producing smaller-sized NPs. Finally, the symmetry of the SPR bands and the lack of additional peaks at longer wavelengths suggest the absence of NP aggregation

(Bindhu and Umadevi, 2014). Although further studies are needed to provide supporting evidence for the above considerations, the results obtained clearly suggest a role for the characteristics of the phenolic extracts in modulating the properties of the synthesized NPs.

The average size of Ag NPs (D) was estimated from the strongest diffraction peak ($2\theta = 38.13°$) using the Debye-Scherrer equation:

$$D = \frac{0.92\lambda}{\beta \cos\theta} \tag{1}$$

where λ is the incident X-ray wavelength, β is the full width at half-maximum height and θ is the Bragg angle. We obtained: D = 16.6 nm, in very good agreement with the size range of 11.7–21.5 nm resulting from SEM measurements (Figure 5). In contrast, a higher value (D = 48.9 ± 1.4 nm) was provided by DLS. This overestimation, which is frequently observed when using the DLS technique, can be attributed to the influence that solvation phenomena and/or the presence of capping agents may have on the hydrodynamic diameter of NPs (Cumberland and Lead, 2009).

The zeta potential of the NPs was –34.5 ± 4.41 mV, indicating that the particle surface was negatively charged and hence that Ag NPs can be expected to be quite stable against aggregation.

4. Conclusions

The results of the present study indicate that BW and SCG could be used as a valuable source of phenolic compounds for the green synthesis of Ag NPs. We have also shown that the characteristics of the resulting NPs are affected by both the type of waste used and the extraction conditions. This is probably a reflection of the nature of the extracted phenolic compounds and their relative amounts in the hydroalcoholic extracts.

Future studies should be directed at identifying the major phenolic compounds present in the extracts and elucidating the mechanisms involved in NP formation and stabilization.

Table 1: Characteristics of the SPR band of Ag NPs produced from extracts with solvents at different ethanol concentrations (EC). A_{max} is the absorbance at λ_{max} and FWHM is the full width at half maximum of the peak. Synthesis conditions: T = 25 °C, reaction time = 4 h, Ag^+/GAE = 10 mol/mol

BW				SCG			
EC (% v/v)	λ_{max} (nm)	A_{max}	FWHM (nm)	EC (% v/v)	λ_{max} (nm)	A_{max}	FWHM (nm)
0	430	1.991	94	0	435	1.813	224
12.5	424	1.773	97	10	448	2.036	157
25	423	1.907	104	20	436	1.555	150
50	420	2.164	99	40	431	1.295	110
75	431	1.644	122	50	422	0.909	118
100	435	1.416	121	60	431	0.626	138

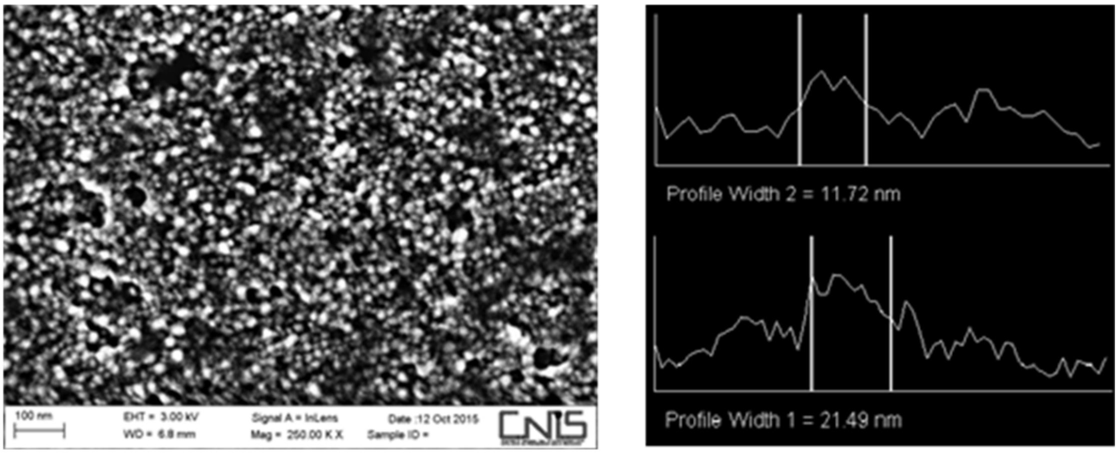

Figure 5: Imaging of Ag NPs and evaluation of NP size from SEM measurements

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