GREEN SYNTHESIZED SILVER NANOPARTICLES DERIVED FROM AN EXTRACT OF THE BETULA PENDULA TREE

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Abstract

This is a first report for the synthesis of silver nanoparticles using *branch* extract *of Betula pendula* - one of the most common trees in Europe, China and southwest Asia. The present study is dealing with ecofriendly green synthesis of silver nanoparticles (AgNPs) using the aqueous extract of branches from *B. pendula* tree. The reaction mixture contained only the extract (10 or 25 % of the total volume of the reaction mixture) from the branches and the source of silver ion without the addition of any catalyst, surfactant or template. The bio-synthesis was carried out at room temperature for 24 hours. The described method is perfectly green, simple, coast-effective and eco-friendly. Reducing substance from birch branches are purely natural and can be obtained simply without damaging the tree. The AgNPs were characterized by UV-vis., DLS and TEM. Surface plasmon spectra for AgNPs are centered at 430-436 nm. The synthesized AgNPs were found to be spherical in shape with size 22.7 ± 6.9 nm and 25.2 ± 7.5 nm, respectively for a reaction mixture containing 10 or 25 % of the extract. Biomolecules from branches acted as a reducing agent as well as stabilizer. Common green synthesis uses "green" parts of plants, especially leaves or fruits. It should be noted that in countries with alternating seasons, leaves are not available throughout the year. Branches, on the other hand, are available all year round and their composition is constant regardless of the season.

Keywords: Green synthesis, *Betula pendula*, silver nanoparticles, eco-friendly

1. INTRODUCTION

Nanoparticles (NPs) of noble metals are used in a wide range of applications from electronics to biology and medicine. Nanoparticles thanks to its "nano" size exhibit properties entirely different from the bulk material. This is essentially a high surface to volume ratio which increases with decreasing size of the nanoparticles. The large free surface is particularly advantageous for catalytic purposes [1,2], as well as for enhanced Raman spectroscopy (SERS) [3,4]. Among the nanoparticles of noble metals silver occupies an important place due to its unique antibacterial properties [5-7]. With increasing demand for silver nanoparticles is also growing effort to prepare them in a way that is effective, but also inexpensive. Chemical preparation methods of nanoparticles with different size and shape are very well known. These methods often utilize strong reducing agents e.g. sodium borohydride [8], run at elevated temperatures, and often are used toxic or hazardous substances [9,10] or high pH [11]. On the other hand, so-called green syntheses as defined by Sharma [12] are based on selection of suitable solvent medium, they are environmentally friendly and reducing substances comes from nature. The main sources of reducing agents are plants [13-23], fungi [24] and microorganisms [25,26]. Reducing agent figures often as a stabilizer and capping agent [20,23,27] too. Because green synthesis take place under mild conditions, sources of reducing agents comes from nature and the additional chemicals are not necessary, this procedure is not only environmentally friendly, but also inexpensive. As already mentioned, frequently used parts of plants are, e.g. leaves [14,19,25,26,28], roots [27,29], seeds [18] or fruits [23]. In areas where there is a change of seasons it is not possible to obtain the leaves or fruits all year round. In contrast, bark, or parts of trees are available year-round. Yet there have only minimal information on the use of the bark

[16] for the preparation of nanoparticles. Data on the use of branches or whole wood extract are altogether missing. Our aim was to test using branches extract to bio-reduction of silver nanoparticles. As a suitable source of reduction substances appears to be silver birch tree, which is wide-spread throughout Europe, China and southwest Asia. Birch tree (*B. pendula*, Linné, 1753) is a genus of deciduous trees or shrubs of the family *betulaceae*. It is a genus of mild to sub polar zone of the northern hemisphere, widespread mainly in the Eurasian region. *B. pendula* also known under the name of silver birch or warty birch is a small to medium-size tree with white peeling bark. Birch is typical for rapid growth; the thin ends of the branches can be easily removed without damaging the tree. The water content and composition of wood slightly varies depending on the season [30], but if dried branches are used, the water content does not play any role.

2. MATERIALS AND METHODS

2.1. Chemicals

All chemicals/reagents used in the study were of analytical grade and were purchased from Sigma-Aldrich, and they were used without further purification. All solutions were prepared with Milli Q water.

2.2. Synthesis of AgNPs (silver nanoparticles)

Accurate concentration of 10mM silver nitrate was prepared by dissolving $AgNO_3$ in 25 ml of Milli Q water. A fresh solution was prepared before each experiment. Birch tree (*Betula pendula*) branches were collected from the tree in Olomouc town, Czech Republic during the beginning of March, 2017, at a time when the tree does not have leaves yet. The branches were cut into pieces of about 2 cm, rinsed with distilled water thrice followed by Milli Q water to remove the dust and other contaminants and then dried at 60°C for 24 hours to remove the moisture. Weight of dried branches compared to fresh halved. Then 10 g of dried birch branches were weighed, mixed with 100 ml of Milli Q water and boiled in a 250 ml Erlenmeyer flask under continuous stirring for 10 minutes. After cooling the extract was filtered using Whatman No.1 filter paper and stored at 4°C for further use. We have observed how the concentration of the reducing agent will affect the formation of AgNPs. Two different volume of branch extract (10 ml or 25 ml) was added to $AgNO_3$ solution (final concentration of $AgNO_3$ was 1 mM, total volume 100 ml) for bio reduction process at room temperature and observed for change in color. The final solution containing bio-synthesized AgNPs was centrifuged at 10 000 rpm for 15 min. The precipitate was washed 3 times to remove unreacted silver ions, kept in petri dish and left in the oven at 60°C for 24 hrs. Dry AgNPs were scrapped and used for characterization.

2.3. Characterization of AgNPs

The bio reduction of Ag^+ ions in solutions was monitored by measuring a spectrum of the reaction medium. The UV-VIS spectral analysis of the sample was done by using UV−VIS absorption spectroscopy with the Specord S 600 spectrophotometer (Analytik Jena AG). The average sizes and size distributions of the prepared noble metal NPs were determined by dynamic light scattering (DLS) using the Zetasizer Nano ZS (Malvern). The nanodimensions of the synthesized silver NPs were confirmed by transmission electron microscopy using the JEM-2010 (Jeol). Zeta potential was measured using the Zetasizer Nano ZS (Malvern). Fourier transform infrared (FTIR) spectral measurements were carried out to identify the potential biomolecules in *Betula p.* branch extract which is responsible for reducing and capping the biogenic silver nanoparticles. FTIR absorption spectra were measured on an FTIR spectrometer iS5 Thermo Nicolet employing attenuated total reflection (ATR) on a ZnSe crystal, using 64 scans with the resolution of 2 cm^{-1}. Air-dried samples were placed directly on a ZnSe crystal and slightly pressed in order to remove any residual air containing humidity and CO_2.

3. RESULTS AND DISCUSSION

The addition of birch (*Betula pendula*) branch extract to the silver nitrate solution resulted in the change in color from light yellowish to dark yellow on incubation at room temperature (24 ºC). Color change of the reaction mixture containing 10 % and 25 % extract immediately after the addition of the extract, and after 24 hrs. are shown in **Figure 1**. Silver particles exhibit dark yellow color in the solution because of the excitation of surface plasmon vibrations caused by the silver nanoparticles as described elsewhere [31]. The color gradually darkened with increasing time. The increasing concentration of silver nanoparticles was confirmed by UV spectrophotometry (**Figure 2**). The presence of the surface plasmon provides information about the size and the morphology of nanoparticles [31-33]. The larger volume of the extract of the reaction mixture showed higher reduction efficiency as seen in the UV-VIS spectra, as well as larger size of the nanoparticles and high yield. Nanoparticles reduced 10 % extract exhibited plasmon with a maximum at 430 nm, 25 % at 436 nm respectively. Bio synthesis of silver nanoparticles was complete after 24 hours as can be seen from the spectra (**Figure 2**), location of maxima and peak height did not change since the twenty fourth hour.

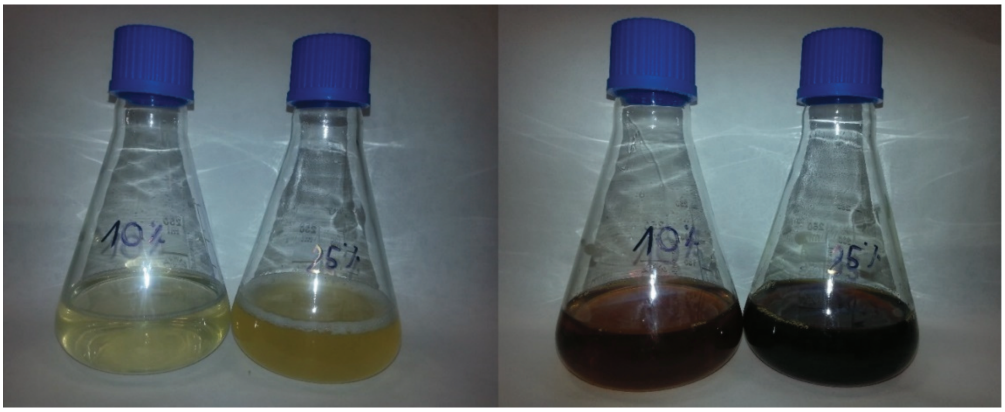

Figure 1 Reaction mixtures immediately after addition of birch extract (left), reaction mixture after 24 hrs. (right)

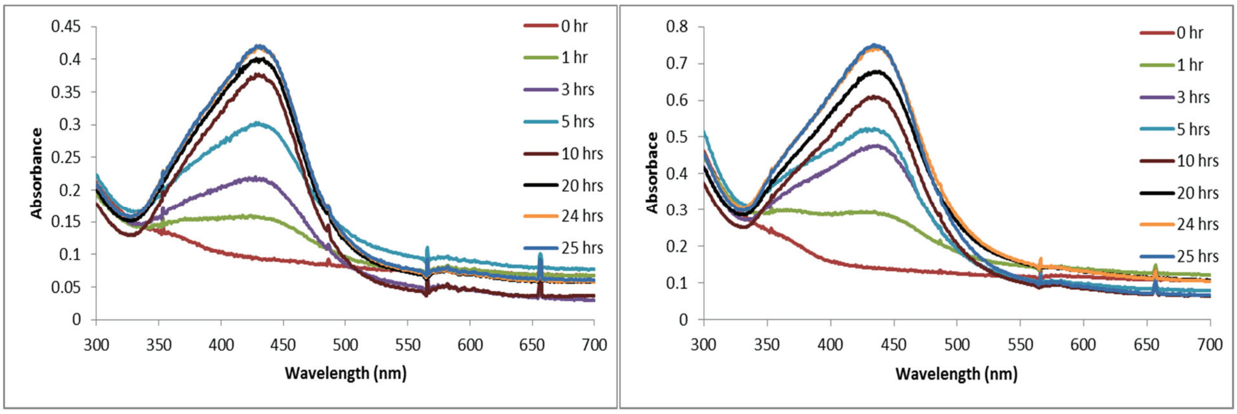

Figure 2 UV-VIS. Absorption spectra recorded in time horizon from 0 to 25 hours. Mixture containing 10 % (v/v) of extract (left), and 25 % (v/v) of extract (right).

The size and morphology of bio synthesized silver nanoparticles were examined using TEM (**Figure 3**) and DLS. The obtained data are summarized in **Table 1**. Size obtained from DLS was somewhat larger than TEM data. It is evident that the biosynthesized nanoparticles are more or less spherical in shape and their size variances in the interval between 7 and 37 nm. The nanoparticles prepared with 25 % extract showed more negative values of zeta potential, it can therefore be assumed that the stability of the dispersion is higher.

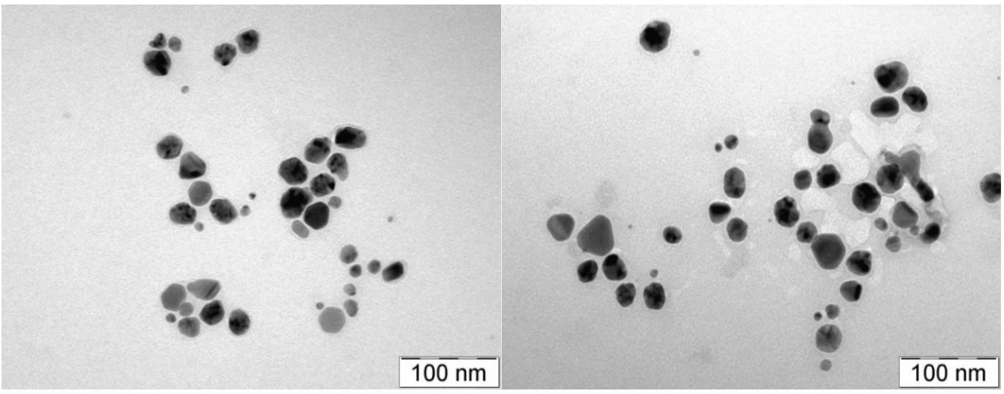

Figure 3 TEM images of silver NPs. 10 % extract (left), 25 % extract (right)

Table 1

	Size TEM	Size DLS	Zeta potential	Absorption peak
10 % extract	22.7 ± 6.9 nm	32.14 ± 0.08 nm	-22 ± 0.86 mV	430 nm
25 % extract	25.2 ± 7.5 nm	51.45 ± 1.49 nm	-24 ± 0.63 mV	436 nm

Fourier transform infrared (FTIR) spectral measurements were carried out to identify the potential biomolecules in birch tree branches extract which is responsible for reducing and capping the biogenic silver nanoparticles. Typical FTIR spectrum of *Betula pendula* branch extract is shown on **Figure 4**.

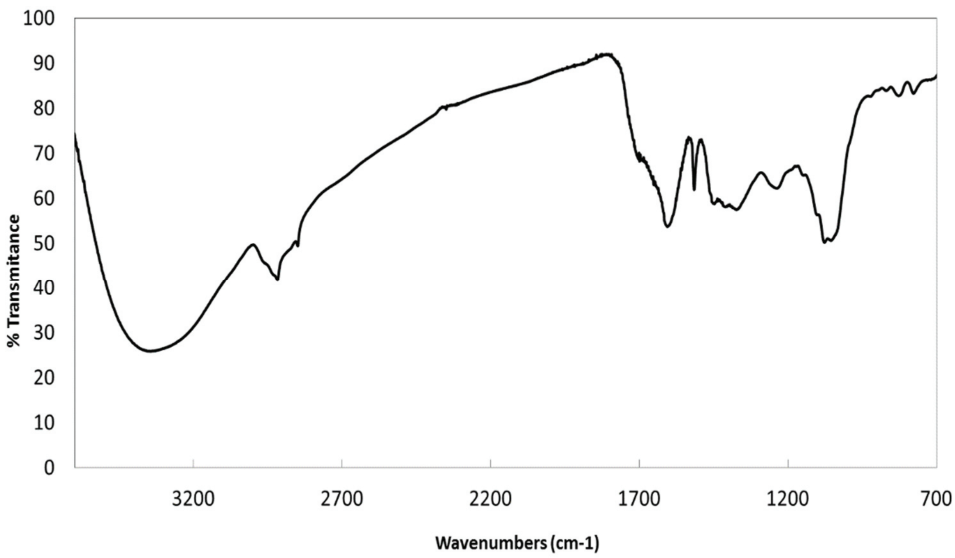

Figure 4 FTIR spectrum of *Betula pendula* branch extract

There are several characteristic bands. The finger prints region lied between wavenumbers ranging from 700 and 1600 cm^{-1} and typical stretching of chemical groups was observed at 2800 to 3500 cm^{-1}. The absorption bands at 1605 cm^{-1} and 1515 cm^{-1} are characteristic of phenyl ring skeletal vibrations of lignin macromolecules [34,35]. The broad band at 3350 cm^{-1} was due to presence of hydroxyls. The band 2917 cm^{-1} was contributed to methyl and methylene groups. In the finger print region band located at 1327 cm^{-1} was characteristic for the presence of methyl groups and 1076 cm^{-1} for alcohol groups. These bands originated from the functional groups present in the various tree metabolites of the branches extract. The functional groups such as -OH present in extract might be responsible for bio reduction of Ag^{+} to AgNPs. *Betula p.* is rich in tannins (an astringent, bitter plant polyphenolic compounds that binds to and precipitates proteins) and other (poly)-phenols. So the capping agents might be tannins or phenols and it's -OH groups might be involved in bio reduction of silver nitrate. These results also correlate with the literature that had already been reported [22,27].

3. CONCLUSION

This is a first report for the synthesis of silver nanoparticles using *branch* extract *of Betula pendula. B. pendula* branch extract was applied for bio reduction of silver ions and environmentally friendly green synthesis of spherical silver nanoparticles. Preparation of AgNPs was carried at room temperature for 24 hours and the reaction mixture was composed of only the extract from the branches and the source of silver ion. The described method is absolutely green, simple, coast-effective and eco-friendly. Biogenic silver nanoparticles are stable due to the layer of bioactive substances adsorbed on their surface and can be used for the same purposes as nanoparticles prepared by conventional techniques. Branches of a tree as a source of reducing agents are available for all seasons unlike the leaves or fruits, the composition of the branches is more less constant regardless of the season and the end parts of the branches used for the reduction can be obtained simply without greater damage to the tree.

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