New Trends in the Synthesis of Nanoparticles by Green Methods

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In this brief survey, we deal with green processes concerning the synthesis of zerovalent nanoparticles, enlighting some aspects motivating their choice with respect to traditional techniques generally relying upon toxic or noxious reactants and stabilizing agents. After a short discussion about health and environmental safety related to the use of standard reductants, we run through several green methods for metal nanoparticle synthesis and we split them into two basic classes, according to the electropositivity of the elements which the nanoparticles are made of. This classification has been proposed in order to account for strengths and weaknesses of processes based on active substances of biological origin that, though being effective in the production of noble metal nanoparticles, proved to be much less suitable when tested in the synthesis of nanoparticles made of more electropositive elements. The goal of this work is essentially oriented to stimulating new research trends for the eco-friendly production of nanosized non-noble elements deserving more attention by current nanobiotechnology.

1. Introduction

The production of nanomaterials is getting an increasing importance for its implications in many different aspects of technology (Pascariu et al., 2013). As a first rough classification, nanomaterials can be produced by large or small-scale manufacturing processes, according to their specific end-uses (Reverberi et al., 2016a). In the first case, as an example, the synthesis of inorganic nanosized powders by flame pyrolysis is one of the most important techniques in the production of dyes, pigments and refractories in raw chemical industry. In the latter, some classes of nanomaterials are still considered as niche products and they are still synthesized by batch chemical processes often requiring an accurate and problematic control in the relevant process variables (Reverberi et al., 2016b). We refer in particular to the synthesis of nanosized powders of organic and inorganic products for pharmaceutical uses, where the dimension of the particles and the presence of unwanted by-products as contaminants play a basic role in determining the quality of the final product. In this context, many efforts are recently focused on process optimization for the synthesis of functionalized nanoparticles (NPs) as carriers for the dispersion of antitumoral molecules by intravenous or transdermal route, possibly targeting single cells, so minimizing side-effects typical of systemic administration (Pastorino et al., 2016). When a chemical process is adopted for the synthesis of NPs, many species as reagents, complexants, stabilizers and surfactants are often adopted as basic chemicals, or correctors aiming at tuning the characteristics of the as-produced nanophase. In particular, a reduction process requires the use of inorganic or organic electron donors that may have toxic, irritating, mutagenic or somehow noxious effects to humans or to the environment. As a consequence, the research is now focused on finding new economic, innocuous and ecological reactants to be used in place of the aforementioned toxic molecules. Of course, such reagents substitution implies the search of a new trade-off among environmental sustainability, reaction yield and safety conditions related to changes of process variables settings (Thunugunta et al., 2015)

Examining purely physical methods leading to the synthesis of NPs is beyond the scopes of the present survey. Instead, we focus on processes where biochemistry has a basic role, in order to point out which aspects should be modified to reach a greater reliability in terms of environmental soundness and sustainability. This work addresses the problems of changes in reagents and process variables for NPs synthesis, according to inherent safety concepts applied already in conventional processes (e.g. Fabiano et al., 2012), in order to meet the restrictions enforced by recent constraints in terms of health and environmental safeguard.

2. Some problems related to "classical" reductants

The production of zerovalent elements generally require process schemes that differ from those adopted in the synthesis of metal compounds (like oxides), owing to unfavourable thermodynamic constraints often requiring drastic experimental conditions. The most widely used inorganic reductants in standard wet chemical processes for metal NPs are alkali metal borohydrides, hydrazine, hydrazonium salts, and alkali hypophosphites.

Sodium borohydride ($NaBH_4$) is perhaps one of the strongest reductants in small scale synthesis of metal nanoparticles and it is widely adopted in laboratories for its versatility in many organic syntheses. It has been used in a broad variety of operative conditions and it proved to be useful for its fast reaction kinetics of reduction even at relatively low temperatures, typically occurring in aqueous solutions at ordinary pressure. It has a good solubility in water and alcohols and generally it releases hydrogen among its decomposition products. However, in certain conditions, boranes are produced and this fact poses some concern in terms of human and environmental safeguard. These gaseous boron by-products are often spontaneously flammable in air, and explosions have occurred in reservoirs where this reagent was stored for long times. Besides warnings related to fire hazards, boranes have well-defined noxious effects on respiratory tract, as reported in many studies (Uemura et al., 1995), while their neurotoxicity is documented by previous statistics concerning workers occupationally subject to acute exposures by inhalation. The symptoms were not only limited to transitory effects, but even some cases of encephalopathy were reported.

Hydrazine (N_2H_4) an its derivatives are more economical reductants than alkali borohydrides, and they could be considered as a milestone in many metal NPs synthesis. Depending on the cation to be reduced, hydrazine has a slightly slower reduction kinetics than borohydrides and it has been used both in aqueous and in non-aqueous solvents. Industrially, it is largely adopted as oxygen-scavenger that makes it a valid additive to prevent oxidation in boilers and reservoirs, even in marine applications. In nanotechnology, it is generally employed as hydrate, as in the pure state it poses some problems of storage and handling owing to its reactive, corrosive and toxic properties. This last aspect has been extensively and thoroughly treated in a report of the US Department of Health and Human Services (Choudhary et al., 1997) where systemic, neurological, immunological and degenerative effects of this compound have been pointed out and documented by exhaustive studies on animals. The human effects rely upon a scanty documentation, generally based on acute or repeated effects due to periodical weekly exposures.

The biological mechanisms by which hydrazine compound show their noxious effects are mainly related to its metabolic fate, as there are both enzymatic and non-enzymatic process of transformation. Moreover, cytochrome P450 catalyses hydrazocompounds degradation in vivo with the onset of free radicals, whose formation is enhanced by the presence of cations of transition elements, such as Cu and Fe, strengthening a cellular oxidative stress and ultimately compromising cellular functions (Sinha and Mason, 2014).

Alkali metal hypophosphites, such as sodium hypophosphite (NaH_2PO_2), though being powerful reducing agents, are somewhat less employed than the previously cited reagents for some unwanted side reactions typically occurring in the synthesis of metal NPs. In particular, the formation of phosphides in the nanostructured solid phase is one of the main drawbacks actually limiting their uses, except in those cases where they are explicitly required for the synthesis of metal-P composites. The technology using such reagents is often described as a green one, as the toxicity of metal hypophosphites depends generally on the cation in question. However, it should be pointed out that, in certain operative conditions occurring at temperatures above the range 100 – 150 °C, metal hypophosphites are subject to thermolysis with production of phosphine (PH_3) according to the following reaction:

$$y\, Me(H_2PO_2)_x \rightarrow Me_y(P_2O_7)_{\frac{xy}{4}} + \frac{xy}{2}\, PH_3 + \frac{xy}{4} H_2O \tag{1}$$

where Me is whatever cation and the product xy is an integer number with xy$\geq$4. For this reason, the use of hypophosphites as reductants in NPs synthesis is usually limited to wet chemical reactions carried out in aqueous solvents, where temperature values are low enough to hinder the onset of reaction (1). Phosphine poses some hazards for human and environmental safety as it is a highly toxic, reactive and spontaneous flammable gas with severe acute and chronic effects on man.

The list of reductants in NPs manufacture includes also many organic compounds, which are typically used for noble metal zerovalent NPs synthesis. The paper by Han and Kim (2009) contains an exhaustive list of organic compounds to be used in this context and we refer the reader to the references quoted there. It should be noted that most organic reductants have milder effects than inorganic ones towards the reduction of cations. A possible explanation of this aspect can be found provided we compare the list of redox potential values of organic reductants with respect to the ones typical of inorganic reductants, as it has been pointed out by the mentioned authors. As a consequence, most experimental studies concerning the synthesis of zerovalent NPs by "green" methods are generally focused on noble cations owing to their faint electropositive character that makes them more easily reducible by organic compounds. The paper of Mittal et al. (2014) presents a detailed list of recent works on NPs green fabrication, where the mentioned reasoning seems to find a good motivation. In the following section, we briefly summarize the mostly adopted eco-friendly methods suitable for the synthesis of noble metals NPs, whose synthesis by green processes got an impressive acceleration in last years. Admittedly, the same protocols are not so efficient when applied to the manufacture of chemically reactive zerovalent NPs, despite this drawback seems to be customarily overlooked by most current literature. Up to last years, these constraints represented a serious limitation to a further extension of phytosynthesis techniques to the production of NPs made of moderately, or highly electropositive elements, but there are further promising developments that is worth considering.

3. Green methods for the synthesis of zerovalent nanoparticles of noble elements.

3.1 Nanoparticles by plant extracts and agro-waste
Many techniques relying upon the choice of plant extracts have been proposed and tested in several experimental works, which have been collected in the excellent review paper by Adelere and Lateef (2016).
According to FTIR investigations, the most important reactants carrying out the reduction process can be ascribed to polysaccharides, polyphenols, terpenoids, flavonoids, alkaloids, carotenoids, catechins, anthocyanidins, sterols, ascorbic acid, reducing sugars, lignin, proteins and related amino-acids. The functional groups involved in such processes are hydroxyl, carboxyl, aldehyde, ketone, amine and even thiol and disulfide bonds, the latter having a basic role when by products deriving from enzyme transformation take part to the electron transfer (Duran et al., 2015). The details of mechanisms and the reaction steps involved in such reduction processes remain often obscure for many reasons, probably owing to the presence of many active substances simultaneously taking part as electron-donors in the same process. Santos et al. (2014) proposed an interesting and comprehensive investigation about this aspect, and they pointed out that, when a bark extract of Eucalyptus globulus is employed, the role of phenolic compounds is much more effective than the one of proteins in the reduction process. They identified 16 different phenolic compounds in the extract and concluded that gallic acid and galloyl compounds have a main role in the synthesis of Au and Ag NPs, while sugars do not essentially take part to the reduction process. Rather, they act as particle stabilizers, according to Z-potential data collected in tests artificially reproducing the composition of a natural extract.
An exhaustive list of different species of plants suitable for NPs synthesis can be found in the work of Iravani (2011). By the way, enzymes present in plants may act as catalysts modifying the reduction speed or acting simply as chemicals having a direct reducing activity toward the cation. The pH of the medium has a basic role in determining the reaction yield as the protonation may strongly condition the role of active sites as electro-donors, thus modulating both the reduction process and the following capping step.
Pigments have proven to be likewise useful as biomolecules showing promising properties as reductants for metals like Ag, Au and for compounds as oxides and sulfides of transition elements (Adelere and Lateef, 2016). A common point characterizing most NPs produced by biosynthesis is a decrease in the systemic toxicity and an increase of biocompatibility and selectivity against carcinogenic cells in mammalians with respect to the same NPs produced by traditional chemical route.

3.2 Nanoparticles by microorganisms, fungi, algae, yeasts and living plants
Microorganisms have been utilized as "bio-nanoreactors" for the synthesis of NPs according to two different processes, essentially divided into intracellular and extracellular assembling methods, both generally adopted when algae (Shankar et al., 2016) and fungi (Siddiqi and Husen, 2016) are employed. In the former, cations cross the membrane cell and undergo a reduction process inside the cell itself, subsequently forming NPs by reciprocal aggregation. The isolation of nanoparticles requires a further process of cell membrane breaking, which may be performed by sonication and further extraction of NPs by a sequence of chemical-physical methods, like centrifugation and solvent extraction. To overcome these drawback, which may be a source of operational costs at industrial production scale, Singh et al. (2016) stressed the advantages offered by extracellular methods, where cations are attracted by specific functional groups lying at the membrane surface, acting as electro-donors and further reducing the cations with formation of neutral atoms as precursors for NPs, which grow without crossing the cell surface. Likewise, unconventional fuel bioproduction, effective design and

scale-up of the equipment require the definition of an appropriate kinetic model of the process and operating conditions optimization (Palazzi et al., 2002). Ponnuchamy and Jacob (2016) reviewed several processes where green, red and brown seaweeds were employed in the preparation of metal NPs from the corresponding salts. They presented an exhaustive list of different seaweed species adopted in literature, where it can be seen that the success of such technology is essentially limited to the least electropositive metals like Au, Pt and Ag. Yeasts, which have long been used for intracellular syntheses of sulphides as semiconductors for electronic applications, were also adopted for the production of noble metal NPs by extracellular methods in more recent years (Mandal et al., 2006). Living plants have been proposed as organisms acting like substrates adsorbing a cation which is further reduced by metabolic processes (Marchiol, 2012), according to reaction steps remaining still partially unknown. In this context, Haverkamp and Marshall (2009) studied the formation of Ag NPs by phytoextraction and they were ones of the rare authors expressing scepticism toward the use of plants in the synthesis of NPs, as they reported many unsuccessful cases involving Pb, Zn, Cd, Ni and Tl ions where the formation of zerovalent metal NPs was not observed. Moreover, they pointed out that, even for noble cations like $Ag+$, the reaction yield is strongly dependent on different ways of precursor complexation and they finally concluded that such methods are recommendable for precious or semiprecious elements only. Their reasoning is still globally valid, but some exceptions have been found and they are listed in the following chapter.

4. Green methods for the synthesis of zerovalent nanoparticles of non-noble elements.

Madhavi et al. (2013) obtained zerovalent iron NPs (NZVI) in the range 50 - 80 nm using leaf extract of Eucalyptus globulus and they proved the efficiency of such a solid phase as a substrate adsorbing chromium compounds as pollutants. The reduction process was fast, totally green and it was carried out at room temperature. They reasonably supposed that polyphenols like oenothein were the main reagents complexating the Fe ion, reducing it and finally stabilizing the as-formed NZVI. These hypotheses were confirmed by FTIR analyses, proving that the oxydryl grops present in polyphenols acted as carriers for the bioreduction process.

Machado et al. (2013) compared the reducing power of 26 different leaf extracts as likely candidates to be used in the preparation of NZVI. The extracts were divided in three different classes according to the corresponding antioxidant capacity toward Fe ions. The process led to NZVI of good quality in terms of dimension (10 - 30 nm), despite a certain tendency to agglomeration, depending on the specific characteristics of the plant extract. Best results were obtained using tea, oak and pomegranate leaves when they leave the corresponding trees, namely at a dry state. Water and, in some cases, a mixture of water-ethanol gave the highest extraction yields and the concentration of polyphenols proved to be the key aspect in the process.

Copper metal NPs were successfully obtained by means of several plant extracts in some studies collected in the review paper by Din and Rehan (2017). In particular, a bark extract from Terminalia Arjuna proved to be satisfactory in a microwave-assisted reduction of copper nitrate ($Cu(NO_3)_2$), and the particles were characterized by X-Ray Diffraction (XRD), showing a high grade of purity in the absence of contamination by oxides (Yallappa et al. 2013).

Nazari et al. (2012) prepared bismuth NPs supplying bismuth nitrate to a nutrient broth where a microorganism (Serratia marcescens) isolated from the Caspian Sea was bred. The colonies were further isolated by centrifugation and subsequently dried and mechanically crushed to free the nanoparticulate phase, which underwent a sequence of purifications leading to a pure elemental bismuth NPs of diameter <150 nm. The authors were the first who successfully produced such element in nanosized scale by a process totally relying upon phytosynthesis.

Lead NPs were produced by specific bacteria colonies of the species Shewanella capable of reducing a lead soluble salt ($Pb(NO_3)_2$) in the presence of sodium s as electron donor (Liu and Jen, 2016). It was ascertained that NPs nucleate and grow inside bacteria strains by means of high-resolution TEM images. Besides, the particles were analysed in composition and their size was dynamically monitored, showing characteristic patterns following the geometry of the host cell.

Koyyati et al. (2016) synthesized cobalt NPs by chemical reduction of cobalt acetate with an aqueous extract of Raphanus sativus at 40°C. The particles were characterized by scanning electron microscopy and they were made of metallic cobalt, showing antibacterial activity by disc diffusion method and cytotoxic activity against human cancer.

An eco-friendly method for the synthesis of nickel NPs was proposed by Elango et al. (2016), who used an extract of Cocos nucifera in methanol to reduce a solution of nickel acetate at 60 °C. Several instrumental analyses proved the formation of elemental nickel in the solid phase. The NPs dispersion was stable and it was tested for its larvicidal and pesticidal properties, with promising results.

Manganese and indium zerovalent NPs merit a little mention apart. In fact, a patent exists explicitly claiming the possibility of obtaining such elements by reduction operated by plant extracts (Hoag et al., 2009). However, to the best of our knowledge, no experimental study has been found in literature where elemental Mn or In have

been obtained by such a synthesis process. It is intriguing to note that, while indium occupies a position lower than Fe in the list of redox potentials ordered by growing electropositive elements, Mn is far more electropositive than Fe, thus making the reduction process carried out by biological organic reductants much more difficult and unlikely.

5. Conclusions

We have briefly summarized some of the mostly adopted methods for the synthesis of metal nanoparticles by eco-friendly processes, taking into account some drawbacks affecting the use of traditional toxic and environmentally noxious compounds as reductants in standard wet chemical processes. In many cases, the reductants proved to act as capping or stabilizing agents, thus making green synthesis a valid and cost effective protocol to be transferred to a larger production scale. Despite an initial reasonable and theoretically motivated scepticism in the use of green methods for the production of zerovalent NPs of non-noble metals, many works seem to encourage this trend and the results are challenging both in terms of quality of the final product, as well as in process safety. Regretfully, in most nanobiotechnology techniques for NPs synthesis, a general lack of modelling studies is observable, probably owing to intrinsic difficulties in describing complex reaction/diffusion phenomena that often overlap in the presence of a biological substrate (Reverberi et al., 2008). The broader production and application of nano-materials will cause a demand for new approaches in process safety, also considering that new processes often are carried out at very high temperatures and/or in unusual pressure ranges, so that material properties under these conditions are largely unknown (De Rademaeker et al., 2014).

References

Adelere I.A., Lateef A., 2016, A novel approach to the green synthesis of metallic nanoparticles: the use of agro-wastes, enzymes, and pigments, Nanotechnology Review 5, 567–587.

Choudhary G., Hansen H., Donkin S., Kirman C., 1997, Toxicological profile for hydrazines. US Department of Health and Human Services, Agency for Toxic Substances and Disease Registry, 1-185.

Din M.I., Rehan R., 2017, Synthesis, characterization, and applications of copper nanoparticles, Analytical Letters 50, 50-62.

Durán M, Silveira C.P., Durán N., 2015, Catalytic role of traditional enzymes for biosynthesis of biogenic metallic nanoparticles: a mini-review, IET Nanobiotechnol. 9, 314-323.

De Rademaeker E., Suter G., Pasman H.J., Fabiano B., 2014, A review of the past, present and future of the European Loss Prevention and Safety Promotion in the Process Industries, Process Safety and Environmental Protection 92, 280-291.

Elango G., Roopan S.M., Dhamodaran K.I., Elumalai K., Al-Dhabi N.A., Arasu M.V., 2016, Spectroscopic investigation of biosynthesized nickel nanoparticles and its larvicidal, pesticidal activities, Journal of Photochemistry and Photobiology B: Biology 162, 162-167.

Fabiano B., Reverberi A.P., Del Borghi A., Dovi V.G., 2012, Biodiesel production via transesterification: Process safety insights from kinetic modeling, Theoretical Foundations of Chemical Engineering 46, 673-680.

Han K.N., Kim N.S., 2009, Challenges and opportunities in direct write technology using nano-metal particles, KONA Powder and Particle Journal 27, 73-83.

Haverkamp R.G., Marshall A.T., 2009, The mechanism of metal nanoparticle formation in plants: Limits on accumulation, Journal of Nanoparticle Research 11,(6), 1453-1463.

Hoag G.E., Collins J.B., Varma R.S., Nadagouda M., 2009, Green synthesis of nanometals using plant extracts and use thereof, Patent WO 2009140694 A2.

Iravani, S., 2011, Green synthesis of metal nanoparticles using plants, Green Chemistry 13, 2638.

Koyyati R., Kudle K.R., Padigya P.R.M., 2016, Evaluation of antibacterial and cytotoxic activity of green synthesized cobalt nanoparticles using raphanus sativus var. Longipinnatus leaf extract, International Journal of PharmTech Research 9(3), 466-472.

Liu C-L, Jen J-H., 2016, Characterization of lead nanoparticles formed by Shewanella sp. KR-12, Journal of Nanoparticle Research 18 (1), article number 30, 1-10.

Machado S., Pinto S.L., Grosso J.P., Nouws H.P.A., Albergaria J.T., Delerue-Matos C., 2013, Green production of zero-valent iron nanoparticles using tree leaf extracts, Science of the Total Environment 445–446, 1–8.

Madhavi V., Prasad T.N.V.K.V., Reddy A.V.B., Reddy B.R., Madhavi G., 2013, Application of phytogenic zerovalent iron nanoparticles in the adsorption of hexavalent chromium, Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy 116, 17–25.

Mandal D., Bolander M.E., Mukhopadhyay D., Sarkar G., Mukherjee P., 2006, The use of microorganisms for the formation of metal nanoparticles and their application, Appl. Microbiol. Biotechnol. 69, 485-492.

Marchiol, L., 2012, Synthesis of metal nanoparticles in living plants, Italian J. of Agronomy 7, (3), 274-282.

Mittal J., Batra A., Singh A., Sharma M.M., 2014, Phytofabrication of nanoparticles through plant as nanofactories, Adv. Nat. Sci.: Nanosci. Nanotechnol. 5, 043002, 1-10.

Nazari P., Faramarzi M.A., Sepehrizadeh Z., Mofid M.R., Bazaz R.D., Shahverdi A.R., 2012, Biosynthesis of bismuth nanoparticles using Serratia marcescens isolated from the Caspian Sea and their characterisation, IET Nanobiotechnol. 6, (2), 58–62.

Palazzi E., Perego P., Fabiano B., 2002, Mathematical modelling and optimization of hydrogen continuous production in a fixed bed bioreactor, Chemical Engineering Science 57, 3819–3830.

Pascariu V., Avadanei O., Gasner P., Stoica I., Reverberi A.P., Mitoseriu L., 2013, Preparation and characterization of PbTiO 3-epoxy resin compositionally graded thick films, Phase Transitions 86, 715-725.

Pastorino L., Dellacasa E., Dabiri M.H., Fabiano B., Erokhina S., 2016, Towards the fabrication of polyelectrolyte-based nanocapsules for bio-medical applications, BioNanoScience 6 (4), 496-501.

Ponnuchamy K., Jacob J.A., 2016, Metal nanoparticles from marine seaweeds – a review, Nanotechnology Review 5, (6), 589–600.

Reverberi A.P., Salerno M., Fabiano B., 2016a, Inorganic nanoparticles synthesis by an aerosol-assisted wet chemical method, Chemical Engineering Transactions 47, 115-120.

Reverberi A.P., Salerno M., Lauciello S., Fabiano B., 2016b, Synthesis of copper nanoparticles in ethylene glycol by chemical reduction with vanadium (+2) salts, Materials 9, (10), Article 809, 1-11.

Reverberi A.P., Bagnerini P., Maga L., Bruzzone A.G., 2008, On the non-linear Maxwell-Cattaneo equation with non-constant diffusivity: Shock and discontinuity waves, International Journal of Heat and Mass Transfer 51, (21-22), 5327-5332.

Santos S.A.O., Pinto R.J.B., Rocha S.M., Marques P.A.A.P., Neto C.P., Silvestre A.J.D., Freire C.S.R., 2014, Unveiling the chemistry behind the green synthesis of metal nanoparticles, ChemSusChem 7, 2704 – 2711.

Shankar P.D., Shobana S., Karuppusami I., Pugazhendhi A., Ramkumar V.S., Arvindnarayan S., Kumar G., 2016, A review on the biosynthesis of metallic nanoparticles (gold and silver) using bio-components of microalgae: Formation mechanism and applications, Enzyme and Microbial Technology 95, 28-44.

Siddiqi K.S., Husen A., 2016, Fabrication of metal nanoparticles from fungi and metal salts: scope and application, Nanoscale Research Letters 11, (1), article 98, 1-15.

Singh P., Kim Y-J, Zhang D., Yang D-C, 2016, Biological synthesis of nanoparticles from plants and microorganisms, Trends in Biotechnology 34, (7), 588-599.

Sinha B.K., Mason R.P., 2014, Biotransformation of hydrazine dervatives in the mechanism of toxicity, Journal of Drug Metabolism and Toxicology 5, 168, 1-6.

Thunugunta T., Reddy A.C., Lakshmana Reddy D.C., 2015, Green synthesis of nanoparticles: Current prospectus, Nanotechnology Reviews 4, (4), 303-323.

Uemura T., Omae K., Nakashima H., Sakurai H., Yamazaki K., Shibata T., Mori K., Kudo M., Kanoh H., Tati M., 1995, Acute and subacute inhalation toxicity of diborane in male ICR mice, Arch Toxicol 69, 397-404.

Yallappa S., Manjanna J., Sindhe M.A., Satyanarayan N.D., Pramod S.N., Nagaraja K., 2013, Microwave assisted rapid synthesis and biological evaluation of stable copper nanoparticles using T. arjuna bark extract, Spectrochim. Acta Part A: Mol. Biomol. Spectrosc. 110, 108–115.