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Development of Antimicrobial Polyester Fabric by a Green *In-Situ* Synthesis of Copper Nanoparticles Mediated from Chitosan and Ascorbic Acid

Behnaz Mehravani[1,a], Ana Isabel Ribeiro[1,b], Majid Montazer[2,c] and Andrea Zille[1,d*]

[1]2C2T-Centro de Ciência e Tecnologia Têxtil, Campus de Azurém, Universidade do Minho 4800-058 Guimarães, Portugal

[2]Amirkabir University of Technology (Tehran Polytechnic), Hafez Avenue, Tehran, Iran

[a]behnaz.mehravani@yahoo.com, [b]afr@2c2t.uminho.pt, [c]tex5mm@aut.ac.ir, [d]azille@2c2t.uminho.pt

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Abstract. The antimicrobial functionalization of polyester fabrics (PES) is useful to provide protection from pathogens and reducing odors. Copper nanoparticles (CuNPs) have been widely applied due to their antimicrobial properties and higher biocompatibility compared with other metal nanoparticles. However, the inherent instability of CuNPs under atmospheric conditions and the use of harmful chemicals during their synthesis limit their use. Thus, the development of efficient and safe methods for the CuNPs synthesis and their stabilization onto surfaces present high interest. In this work, PES was functionalized with CuNPs via *in situ* synthesis using cost-effective and safe chemicals in the presence and absence of chitosan. In sample without chitosan, the CuNPs showed a suitable stabilization onto PES due to the doubled stabilization of ascorbic acid (AA) and cetyl trimethyl ammonium bromide (CTAB). In sample with chitosan, less CuNPs were retained by the PES but also less CuNPs agglomeration was observed. Both samples presented excellent antibacterial effect against *Staphylococcus aureus* (*S. aureus)* and *Escherichia coli* (*E. coli)* as well as laundering durability.

Introduction

Recently, nanotechnology has been incorporated in textiles through various techniques such as coating and padding to induce some properties that are not inherent in textiles. Properties such as water repellency, antistatic, photocatalytic properties, electrical conductivity, thermal stability, flame retardancy and antibacterial can be improved by the surface functionalization of textiles [1,2]. Nanomaterials have attracted a lot of attention due to their unique physical, biological, and chemical properties. They display different properties at atomic levels due to the large surface area to volume ratio [3,4]. Nanotechnology has been used in many applications that include medicine, biotechnology, electronics, energy, and environment. Introducing nanoparticles in the fabric, called nano finishing, various properties will be provided such as UV blocking, antimicrobial, flame retardant, wrinkle resistant, antistatic, water repellent and self-cleaning properties [5,6]. Since the beginning of civilization, copper was considered a hygienic material and, during the last two decades, copper was proved to have good antibacterial properties in several research works [7]. Among metal nanoparticles, copper nanoparticles have been accepted in large due to their unique structure, good mechanical and thermal stability and ideal optical, magnetic, and catalytic properties [5]. However, oxidation is a noted demerit with the usage of copper and hence suitable stabilizing agents should be used while preparation of CuNPs [8]. Among several methods, the chemical reduction method is more often used because of cost effectiveness, high yields, and simplicity [9]. The main role of the reducing agent in the chemical reduction method is to reduce the precursor solution and avoid the oxidation of copper nanoparticles [10]. The most often used reductants that are used in the synthesis of CuNPs include hydrazine (N_2H_4), sodium borohydride ($NaBH_4$) and *L*-ascorbic acid (AA). However, those chemicals are expensive and quite toxic and hence it is essential to search for more

economical and safer reducing agents. Several works have been reported using sodium hypophosphite (SHP) or ascorbic acid (AA) as reducing agent in the preparation of CuNPs [9,11,12]. Most of the recent works are focusing on green synthesis to eliminate the usage of harmful chemical substances and as a part of this green synthesis, extracts from plants such as *Terminalia arjuna* bark, Magnolia leaf, *Datura metel* leaf and microorganisms like *Pseudomonas stutzeri*, *Morganella morganii* are used [13]. Besides the reducing agents, several capping agents have been used to control the NPs shape and protect them from agglomeration. In this group, cetyl trimethylamonium bromide (CTAB) and sodium dodecyl sulfate (SDS) have been widely used [14]. The addition of polymers can also be used for the stabilization of metal nanoparticles as the polymer functional groups may promote electronic interaction with the metal nanoparticles. After the reduction process, the larger protecting polymers cover or encapsulate the metal particles and there is a stabilization effect [15]. Various works discussed the usage of Chitosan, Starch ($C_6H_{10}O_5$), Polyacrylic acid (PAA), Poly methacrylic acid (PMAA) and Polyvinylpyrrolidone (PVP) as stabilizing agents [13,14]. Chitosan is a linear polysaccharide that is produced by the deacetylation of chitin. Its good biocompatibility, biodegradability, antimicrobial activity, wound healing property and antitumor effect has made it a candidate for having their application in various fields such as filtration, drug-delivery, wound dressing, cell culture, tissue engineering, cosmetic, ophthalmology, solid-state batteries and textiles [16]. In recent times, CuNPs have been gathering lot of attention owing to the ease of mixing with polymers [13]. It was reported in the literature that the presence of hydrophilic side chains in chitosan plays an important role in the stabilization of CuNPs and helps to reduce agglomeration [17]. Polyester is used in many applications due to its promising mechanical, chemical and thermal properties [18,19]. The fabrics produced by polyester are one of the most popular in the textile industry. However, problems such as low hydrophilicity, low dye-ability and the lack of functional groups in their polymeric chains make them difficult to induce any functionalities and hygienic problems emerge [20,21]. Therefore, inducing durable antibacterial properties in the polyester fabric presents high interest [22].

In this work, it was explored the functionalization of polyester fabric with CuNPs by an *in-situ* method using the naturally available chitosan as stabilizing agent. Here, SHP and ascorbic acid were used as reducing agents. Besides preventing oxidation, CTAB, it was also added to control the size of nanoparticles. Thereafter, the antibacterial and cytotoxicity of these fabrics were studied to investigate the potentiality of using them in biomedical applications.

Experimental

Materials

The chemical reagents were used without any purification. Copper (II) sulphate pentahydrate ($CuSO_4.5H_2O$) and cetyl trimethyl ammonium bromide (CTAB) were purchased from Merck Co. (Germany); Ascorbic acid (AA, $C_6H_8O_6$) from BDH (England); Sodium hypophosphite (SHP, $NaPO_2H_2$) from Acros Co. (United States) and chitosan with a bulk density of 0.26 $g.mL^{-1}$ and has a viscosity of 78 mPa.s. with a deacetylation degree of 90 % from Chitotech Co. (Iran). Commercial polyester fabric with weight per unit area of 105 $g.m^{-2}$ was obtained from Broojerd Textile Co. (Iran).

Fabric Wash

The polyester fabric was primarily washed with 2 $g.L^{-1}$ non-ionic detergent with liquor to ratio of L:G=50:1 at 80°C for 30 min to remove any impurities from the fabric surface. Then, the fabric was rinsed with distilled water and dried at room temperature.

Synthesis of Copper Nanoparticles

Two methods were used to synthesize CuNPs, one in the presence and another in absence of chitosan. In S1, the $CuSO_4.5H_2O$ (1 g), AA (1 g) and SHP (0.6 g) were dissolved in 100 mL of distilled water at 50°C with magnetic stirring at 375 rpm. Next, PES was immersed in the solution. CTAB (0.5 g) was added when the solution reached 85 °C. For S2, an initial solution of Cu salt (0.1 g) and AA(0.5

g) and SHP (0.6 g) was prepared in 80 mL of distilled water at 50°C with magnetic stirring. The PES was immersed in the solution. Chitosan (0.5 g) with AA (0.5 g) was dissolved in in 20 mL of distilled water and the total volume was mixed with the initial solution at 70°C. The mixture was heated to 85 °C and CTAB (0.5 g) was added. Both solutions were kept at 85 °C under stirring for 1h. The procedure is shown graphically using Fig. 1.

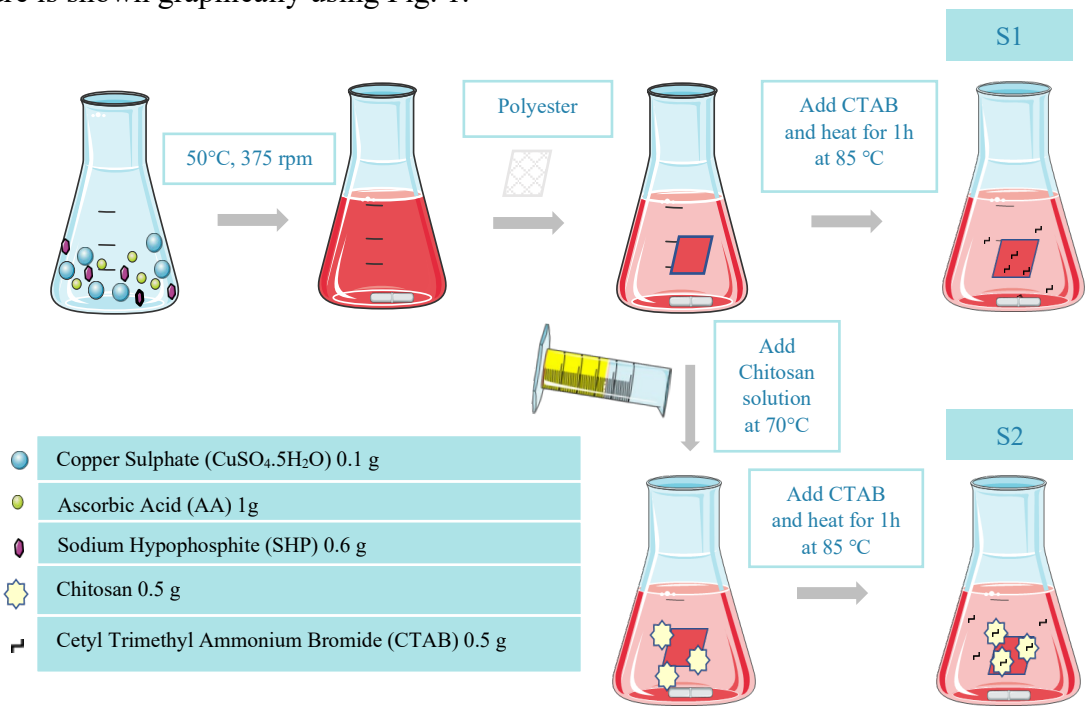

Fig. 1 Graphical Representation of Synthesis of CuNPs

UV-vis reflectance spectra

It is used to identify the confirmation of the formation of the copper nanoparticles on the fabric surface and the values are taken in the range of 200 to 800 nm. UV-Vis spectroscopy (Shimadzu UV-1800 spectrophotometer) was used to estimate the maximum absorbance of the copper nanoparticles.

X-Ray Diffraction (XRD)

X-ray diffraction patterns were obtained with an X-ray diffractometer using Cu K radiation (λ=1.54 nm) PW3040/60 from Panalitical Co. (Germany) to study the structure of the material. This equipment can analyse in the range of 5-150° with 0.001° resolution. The crystal size was calculated using the Debye-Scherrer equation according to Eq. 1.

$$\text{D (crystal size)} = \frac{k\lambda}{\beta \cos\theta} \quad (1)$$

In this formula, K is a constant equal to 0.9 and λ is X-ray wavelength and β is the peak bandwidth at half maximum height [23].

Field Emission Scanning Electron Microscopy (SEM)

FESEM was carried out in the laboratory for materials characterization services of the University of Minho. Morphological analyses of polyester fabrics functionalised with copper nanoparticles were carried out with an ultra-high resolution field emission Scanning Electron Microscope (FEG-SEM), NOVA 200 Nano SEM, FEI Company with integrated microanalysis X-ray systems (EDS-energy dispersive spectrometer) and Electron Backscatter Diffraction (EBSD). Secondary electron images and Backscattering electron images were achieved with an acceleration voltage at 5 kV and 15 kV respectively. Samples were covered with a film of Au-Pd (80-20 weight %) in a high-resolution sputter coater, 208HR Cressington Company, coupled to a MTM-20 Cressington High Resolution

Thickness Controller. EDAX Si (Li) detector was used to observe atomic compositions of the samples with an acceleration voltage of 5 kV.

Antimicrobial Test

Suspension method was used to study the antibacterial properties of the samples according to the AATCC 100-2004 standard. This is a quantitative method, more time-consuming in comparison with the agar plate. 1 mL of bacteria was considered for the bacteria growth which will be absorbed by the textile, and then the textile was soaked in the inoculant substance cultured in the closed-door plates at 37 °C for 24 h and finally the number of the bacteria on the surface of the textile was evaluated by consecutive dilution. The antibacterial action can be calculated as a reduced percentage by using the formula given in Eq. 2. The control sample (blank) without finishing was used for comparing the bacteria growth with finished samples. Two bacteria including one Gram-positive bacteria, *Staphylococcus aureus* (*S. aureus,* ATCC 25923), and one Gram-negative bacteria, *Escherichia coli* (*E. coli,* ATCC 25922), were used. Washing cycles for 10 times has been performed on the samples for 30 mins at 40 ℃ according to the AATCC Test Method 61(2A)-1996.

$$\text{R \% (bacterial efficiency)} = \frac{B-A}{B} \times 100 \quad (2)$$

where A is the number of bacteria recovered after incubation for 24 hours and B is the number of bacteria recovered at the 0 contact time [16].

Results and Discussion

During the preparation of CuNPs it was observed that the color was changed from blue to red after the addition of ascorbic acid and SHP in both methods (S1 and S2) that indicated the emergence of CuNPs. This kind of observation has been reported as an indicator of the reduction of copper ions to metallic copper [24]. Then, the dispersion was further characterized by UV-Vis. UV-vis results were studied to confirm the presence of CuNPs on dispersion and it is known from the literature that the CuNPs used to present an absorbance peak between 500 and 600 nm [25]. The UV-vis spectra of the dispersions S1 and S2 showed the absorbance peak at 590 and 610 nm, respectively (Fig. 2). Hence, the presence of CuNPs was confirmed in both samples. The observations from the study were in coincidence with reports from the literature [25][26].

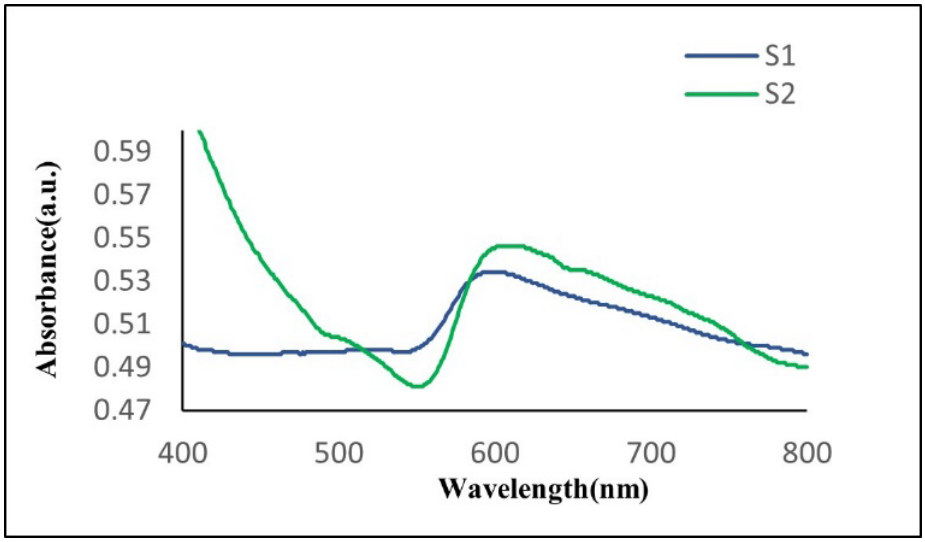

Fig. 2 UV-Vis spectra from S1 and S2 samples.

XRD analysis was used to analyse the crystal size of the CuNPs on the samples synthesized with and without the chitosan (S1and S2). XRD patterns of the CuNPs were found in S1 and S2. The characteristic peaks are related to the plane index of (111) (200) (220), where in S1 the peaks appeared at 2θ = 43.354°, 50.479° and 74.152°, and in S2 at 2θ = 43.371°, 50.498° and 74.164°, respectively (Fig. 3). According to (JCPDS file no. 98-005-5321) the peaks of both samples are indexed to the pure copper. The average crystal size of the CuNPs was calculated by using the Debar Scherrer relation and was determined as 50.3 nm for S1 and 56.0 nm for S2. The crystal sizes of the CuNPs agreed with the values reported in the literature, where it was found that the crystal sizes of the copper nanoparticles are in the range of 35 to 75 nm [23]. It was observed from the XRD that the size of the copper nanoparticles with chitosan (56 nm) was larger than CuNPs in the absence of chitosan (50.33 nm). This draws to the conclusion that the presence of chitosan has resulted in the increment of the size of the CuNPs. This tendency can be due to the reason that chitosan covers the nanoparticles to avoid agglomeration during the synthesis process and hence improving their increase in size [27]. Another reason for the size enhancement with chitosan can be due to the coordination ability of the metallic ions towards the amino groups of the chitosan. They also reported that this could also make the polymer behave as a chelating agent. Hence these interactions will allow the metallic cations to act as templates and form chitosan nanoparticles [28].

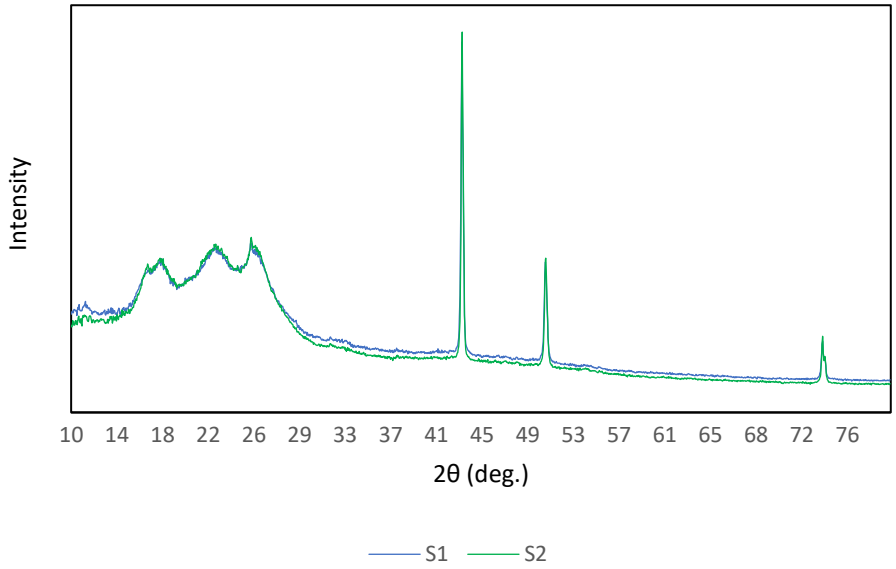

Fig. 3 XRD Pattern of Sample (Control Sample, S1 and S2)

SEM images were collected to analyse the morphology of CuNPs onto PES in the different conditions. First, it was possible to confirm the loading of CuNPs in both samples. Despite the S1 sample showed more CuNPs onto PES surface, also some agglomeration was observed. The CuNPs in S2 were less but they showed to be better distributed, with less agglomeration (Fig. 4). The presence of both CTAB and chitosan might have performed the role of capping agents and have prevented the agglomeration of CuNPs. The EDS analysis proved the superior adhesion of CuNPs in S1 (7.71%) than in S2 (1.71%) (Table 1).

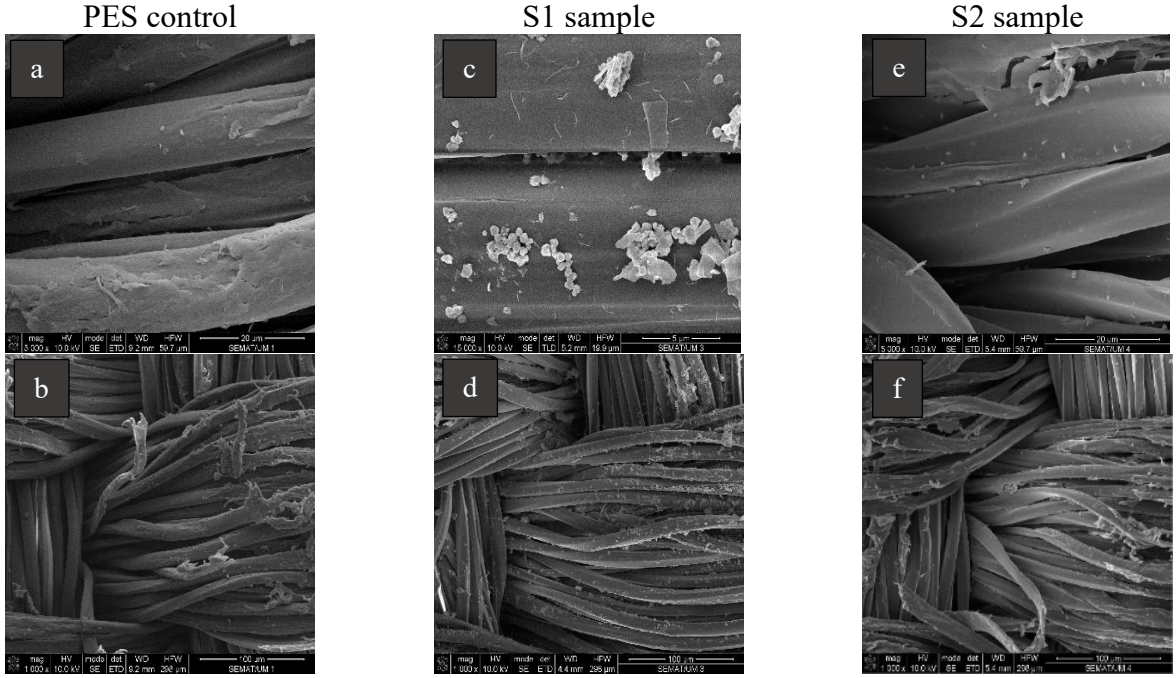

Fig. 4 SEM images of control sample (a and b), S1 (c and d) and S2 (e and f) at magnifications of 5000x and 1000x.

Table 1 EDS analysis of the PES (control), S1 and S2 samples.

Sample	C (W%)	O (W%)	Cu (W%)
Control	64.06	34.94	-
S1	66.77	25.25	7.71
S2	67.94	30.34	1.71

PES samples with CuNPs (S1 and S2) were tested against Gram-negative bacterial strain *E.coli* and Gram-positive strain *S. aureus*. In this test, the antimicrobial activities of S1 and S2 samples were compared with a blank sample, PES fabric without CuNPs (Table 2). The results showed a reduction percentage of 99.9% in both samples with CuNPs and any inhibition in the control sample. To understand the durability and the stability of the CuNPs onto the fabric, 10 washing cycles at 40°C for 30 minutes were performed. The antimicrobial efficacy after wash also showed 99.9% of bacterial reduction in both samples, with and without chitosan. Thus, it can be concluded that both samples have the potential to be used in medical applications. Despite the higher concentration of CuNPs in S1, proved on EDS results, the sample S2 also present equal antimicrobial efficacy. The stabilization of chitosan, ended with the same inhibitory activity towards Gram-negative and Gram-positive bacteria using a reduced concentration of CuNPs, which present several benefits namely in terms of the environmental contamination with metals, reduced cytotoxicity and economical aspects. The antimicrobial activity of CuNPs is commonly attributed to the ions released from the CuNPs. Interaction of the copper ions with the microbial membranes and production of radicals when the ions and nanoparticles are attached to the DNA molecules. It results in a disordering helical structure by cross-linking within and between the nucleic acid strands inducing the antimicrobial effect [29]. Chitosan may improve the antimicrobial effect due to its polycationic nature at pH under 6. It interacts with molecules with negatively charged components on the bacteria cell wall, forming an impermeable layer around the cell and changing the permeability of the cells which results in the blocking of transport into the cell [30].

Table 2. Antimicrobial activity of control, S1 and S2 samples before and after 10 washing cycles.

Sample	*S. aureus* (reduction %)	*E. coli* (reduction %)	*S. aureus* - washed (reduction %)	*E. coli* - washed (reduction %)
Control	0	0	0	0
1	99.9	99.9	99.9	99.9
2	99.9	99.9	99.9	99.9

Conclusion

The synthesis and deposition of CuNPs on PES were performed successfully using an *in-situ* method. Both samples demonstrated suitable antibacterial properties against Gram-positive and Gram-negative bacteria opening new opportunities for the development of efficient and safe-by-design antimicrobial PES fabrics.

References

[1] A. Salman, F.A. Metwally, M. Elbisi, G.A.M. Emara, Applications of nanotechnology and advancements in smart wearable textiles: An overview, Egypt. J. Chem. 63 (2020) 2177–2184.

[2] C.I. Idumah, Influence of nanotechnology in polymeric textiles, applications, and fight against COVID-19, J. Text. Inst. 0 (2020) 1–21.

[3] S. Riaz, M. Ashraf, T. Hussain, M.T. Hussain, A. Rehman, A. Javid, K. Iqbal, A. Basit, H. Aziz, Functional finishing and coloration of textiles with nanomaterials, Color. Technol. 134 (2018) 327–346.

[4] B. Mehravani, A.I. Ribeiro, A. Zille, Gold nanoparticles synthesis and antimicrobial effect on fibrous materials, Nanomaterials. 11 (2021) 1067.

[5] P. Sharma, S. Pant, P. Poonia, S. Kumari, V. Dave, S. Sharma, Green Synthesis of Colloidal Copper Nanoparticles Capped with Tinospora cordifolia and Its Application in Catalytic Degradation in Textile Dye: An Ecologically Sound Approach, J. Inorg. Organomet. Polym. Mater. 28 (2018) 2463–2472.

[6] A.I. Ribeiro, M. Modic, U. Cvelbar, G. Dinescu, B. Mitu, A. Nikiforov, C. Leys, I. Kuchakova, M. De Vrieze, H.P. Felgueiras, A.P. Souto, A. Zille, Effect of dispersion solvent on the deposition of PVP-silver nanoparticles onto DBD plasma-treated polyamide 6,6 fabric and its antimicrobial efficiency, Nanomaterials. 10 (2020) 607.

[7] S. Anita, T. Ramachandran, R. Rajendran, cv Koushik, M. Mahalakshmi, A study of the antimicrobial property of encapsulated copper oxide nanoparticles on cotton fabric, Text. Res. J. 81 (2011) 1081–1088.

[8] M.C. Sportelli, R.A. Picca, R. Ronco, E. Bonerba, G. Tantillo, M. Pollini, A. Sannino, A. Valentini, T.R.I. Cataldi, N. Cioffi, Investigation of industrial polyurethane foams modified with antimicrobial copper nanoparticles, Materials (Basel). 9 (2016) 1–13.

[9] D. Lai, T. Liu, G. Jiang, W. Chen, Synthesis of highly stable dispersions of copper nanoparticles using sodium hypophosphite, J. Appl. Polym. Sci. 128 (2013) 1443–1449.

[10] U. Kathad, h p Gajera, Synthesis of copper nanoparticles by two different methods and size comparision, Int. J. Pharma Bio Sci. 5 (2014) 533–540.

[11] T.M.D. Dang, T.T.T. Le, E. Fribourg-Blanc, M.C. Dang, Synthesis and optical properties of copper nanoparticles prepared by a chemical reduction method, Adv. Nat. Sci. Nanosci. Nanotechnol. 2 (2011).

[12] S. Jain, A. Jain, P. Kachhawah, V. Devra, Synthesis and size control of copper nanoparticles and their catalytic application, Trans. Nonferrous Met. Soc. China English Ed. 25 (2015) 3995–4000.

[13] M.I. Din, R. Rehan, Synthesis, Characterization, and Applications of Copper Nanoparticles, Anal. Lett. 50 (2017) 50–62.

[14] G. Granata, T. Yamaoka, F. Pagnanelli, A. Fuwa, Study of the synthesis of copper nanoparticles: the role of capping and kinetic towards control of particle size and stability, J. Nanoparticle Res. 18 (2016) 1–12.
[15] A. Travan, C. Pelillo, I. Donati, E. Marsich, M. Benincasa, T. Scarpa, S. Semeraro, G. Turco, R. Gennaro, S. Paoletti, Non-cytotoxic silver nanoparticle-polysaccharide nanocomposites with antimicrobial activity, Biomacromolecules. 10 (2009) 1429–1435.
[16] E. Hadipour-Goudarzi, M. Montazer, M. Latifi, A.A.G. Aghaji, Electrospinning of chitosan/sericin/PVA nanofibers incorporated with in situ synthesis of nano silver, Carbohydr. Polym. 113 (2014) 231–239.
[17] A.M. Muthukrishnan, Green Synthesis of Copper-Chitosan Nanoparticles and Study of its Antibacterial Activity, J. Nanomed. Nanotechnol. 06 (2015) 1–5.
[18] A. Bashiri, M. Montazer, One-step fabrication of fatty acids / nano copper / polyester shape-stable composite phase change material for thermal energy management and storage, Appl. Energy. 228 (2018) 1911–1920.
[19] A. Bashiri, M. Montazer, M. Mahmoudi, Photo and biocatalytic activities along with UV protection properties on polyester fabric through green in - situ synthesis of cauli fl ower-like CuO nanoparticles, J. Photochem. Photobiol. B Biol. 176 (2017) 100–111.
[20] A.B. Rezaie, M. Montazer, Amidohydroxylated polyester with biophotoactivity along with retarding alkali hydrolysis through in situ synthesis of Cu / Cu 2 O nanoparticles using diethanolamine, J. Appl. Polym. Sci. 44856 (2017) 1–15.
[21] M.M. Abdul-Bari, R.H. McQueen, H. Nguyen, W. V. Wismer, A.P. de la Mata, J.J. Harynuk, Synthetic Clothing and the Problem With Odor: Comparison of Nylon and Polyester Fabrics, Cloth. Text. Res. J. 36 (2018) 251–266.
[22] A. Majumdar, B.S. Butola, S. Thakur, Development and performance optimization of knitted antibacterial materials using polyester-silver nanocomposite fibres, Mater. Sci. Eng. C. 54 (2015) 26–31.
[23] M.S. Usman, N.A. Ibrahim, K. Shameli, N. Zainuddin, W.M.Z.W. Yunus, Copper nanoparticles mediated by chitosan: Synthesis and characterization via chemical methods, Molecules. 17 (2012) 14928–14936.
[24] S. Pradhan, R. Shrestha, K. Bhandari, Effect of Various Parameters on Bio-Synthesis of Copper Nanoparticles Using Citrus Medica Linn (Lemon) Extract and Its Antibacterial Activity, Amrit Resarch J. 1 (2020) 51–58.
[25] M. Biçer, I. Şişman, Controlled synthesis of copper nano/microstructures using ascorbic acid in aqueous CTAB solution, Powder Technol. 198 (2010) 279–284.
[26] Y. Suresh, S. Annapurna, a K. Singh, G. Bhikshamaiah, Green Synthesis and Characterization of Tea Decoction Stabilized Copper Nanoparticles, Int. J. Innov. Res. Sci. Eng. Technol. 3 (2014) 11265–11270.
[27] E. Tabesh, H.R. Salimijazi, M. Kharaziha, M. Mahmoudi, M. Hejazi, Development of an in-situ chitosan-copper nanoparticle coating by electrophoretic deposition, Surf. Coatings Technol. 364 (2019) 239–247.
[28] G.R. Domínguez, M.C. Rodríguez-argüelles, N. Gonzalez Ballesteors, G. Rodriguez Dominguez, M. Campanini, L. Nasi, I. Vazquez, C. Sieiro, Broad-Spectrumm Antimicrobial Activity of Silver Nanoparticles in Different Types of Chitosan Matrices, Chem. J. 1 (2015) 165–171.
[29] S.M. Dizaj, F. Lotfipour, M. Barzegar-Jalali, M.H. Zarrintan, K. Adibkia, Antimicrobial activity of the metals and metal oxide nanoparticles, Mater. Sci. Eng. C. 44 (2014) 278–284.
[30] C. keong Lim, A. sukari Halim, Biomedical-Grade Chitosan in Wound Management and Its Biocompatibility In Vitro, in: M.E. Magdy (Ed.), Biopolymers, InTech, 2010: pp. 19–33.