

# ANTIMICROBIAL COTTON FIBERS PREPARED BY TEMPO-MEDIATED OXIDATION AND SUBSEQUENT SILVER DEPOSITION

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In this paper, we merge the unique properties of cotton and silver in order to obtain antimicrobial cotton fibers. The sorption properties of cotton were improved by selective TEMPO-mediated oxidation, *i.e.* oxidation with sodium hypochlorite, sodium bromide and 2,2,6,6-tetramethylpiperidine-1-oxyl radical (TEMPO), and evaluated by determination of moisture sorption and water retention. Increases of up to 1.55 and 4.7 times were obtained for moisture sorption and water retention value, respectively. The antimicrobial properties were achieved by incorporation of silver into TEMPO-oxidized cotton by combination of  $\text{Ag}^+$  chemisorption from aqueous  $\text{AgNO}_3$  and reduction of  $\text{Ag}^+$  to  $\text{Ag}^0$  by cellulose itself. The amount of incorporated silver was in the range from 0.102 to 0.806 mmol/g fibers. The antimicrobial activity of the obtained fibers against pathogens *C. albicans*, *S. aureus* and *E. coli* was evaluated. An attempt has been made to explain the roles of the introduced functionalities and changes in the sorption properties of the TEMPO-oxidized cotton on silver deposition.

**Keywords:** TEMPO-mediated oxidation, cotton fibers, functional groups, sorption properties, silver deposition, antimicrobial activity

## INTRODUCTION

Cotton fibers are deemed as the backbone of the textile industry, and considered as the foremost commercially important natural fiber in the world. Predictions are that the world's cotton consumption for 2014/15 will be of 24.5 million tons.<sup>1</sup> Due to its excellent properties, such as softness, affinity to skin, hygroscopic property, regeneration, biodegradation, dyeability and strength, cotton is widely used in clothing fields.<sup>2,3</sup> As a raw material, cotton fibers are used for preparing numerous products on the basis of cellulose, such as paper, chemicals, food additives, some cosmetic ingredients *etc.*<sup>4-6</sup> In some medical applications, the use of cotton fibers goes back to ancient times, and even today cotton fibrous materials cannot be replaced by any others. For specific applications of cotton fibers in the medical field or health services, the use of cotton fibers on organic tissue surface (external/non-implantable), such as for antimicrobial materials, gauzes, wound dressing,

bandages, surgical covers, nappies, tampons *etc.*, it is necessary to obtain fibers with improved sorption properties.<sup>7,8</sup>

In this work, the sorption properties of cotton fibers were improved by selective TEMPO-mediated oxidation, *i.e.* oxidation with sodium hypochlorite and a catalytic amount of sodium bromide and 2,2,6,6-tetramethylpiperidine-1-oxyl radical (TEMPO). According to the literature,<sup>9-14</sup> catalytic TEMPO-mediated oxidation, using stable and water soluble nitroxyl radicals, such as 2,2,6,6-tetramethylpiperidine-1-oxyl radical (TEMPO), under aqueous conditions, has been proposed as one of the most promising methods for cellulose functionalization, where aldehyde and carboxyl functional groups can be effectively introduced into solid cellulose under aqueous and mild conditions.

Together with the introduction of aldehyde and carboxyl groups into cellulose, by the TEMPO-mediated oxidation, the fibrous morphology of

cellulose fibers has been changed, depending on the oxidation conditions.<sup>9-14,15</sup> Changes in the chemical composition, crystallinity and void system (diameter, volume and inner surface of voids)<sup>16</sup> of cellulose during TEMPO-oxidation affect the sorption properties. In this paper, the sorption properties of TEMPO-oxidized cotton fibers were evaluated by determining their moisture sorption and water retention value.

The antimicrobial properties were achieved by the incorporation of silver into oxidized cotton fibers by chemisorption from aqueous silver nitrate solution. Silver was selected as an antimicrobial agent due to its strong inhibitory and bactericidal effects, as well as its broad spectrum of antimicrobial activities.<sup>17</sup> Furthermore, silver is one of a few antimicrobial agents, which possess both antibacterial and antifungal activity, and bacteria are not able to develop resistance to silver, as in the case of antibiotics.<sup>18-20</sup> The antifungal activity of the TEMPO-oxidized cotton fibers with incorporated silver was tested against fungi from the *Candida* family, *C. albicans* (ATCC 24,433), and the antibacterial activity was assessed for two strains: *S. aureus* (ATCC 25,923) and *E. coli* (ATCC 25,922).

## EXPERIMENTAL

### Materials

Cotton fibers: Russian, I class, 32/33 mm. TEMPO, sodium bromide and 13% sodium hypochlorite solution, and other chemicals obtained from commercial sources were p.a. grade.

### Methods

#### *Preparation of TEMPO-oxidized cotton fibers*

The oxidation procedure was based on the methodology described in the literature.<sup>14,21</sup> In brief, cotton fibers (10 g) were suspended in water (750 mL) containing TEMPO (0.025 g) and sodium bromide (0.25 g). Subsequently, a definite amount of NaClO solution containing 13% available chlorine, corresponding to 0.30, 2.42, 4.84 and 9.67 mmol/g fibers, was added to the cellulose slurry under continuous stirring. The pH of the slurry was maintained at 10.5 at room temperature by adding 0.5M NaOH for 1-4 h. After stirring for a definite time, the oxidation was quenched by adding ethanol (*ca.* 5 mL). The oxidized cotton fibers were washed thoroughly with water and then ethanol on a filter paper set in a Buchner funnel, and dried at room temperature for 48 h.

#### *Silver deposition on TEMPO-oxidized cotton fibers*

Silver was incorporated into previously TEMPO-oxidized cotton fibers under conditions that were

optimized earlier.<sup>22</sup> The change in the concentration of silver after sorption was determined by NH<sub>4</sub>SCN titrations, employing Fe(NH<sub>4</sub>)(SO<sub>4</sub>)<sub>2</sub> as an indicator according to Volhard's method.<sup>23,24</sup>

#### *Determination of carboxyl group content in TEMPO-oxidized cotton fibers*

For determining the carboxyl content in oxidized cotton fibers, the calcium-acetate method described by Kumar and Yang,<sup>25</sup> and Yackel and Kenyon,<sup>26</sup> and recently modified by Praskalo *et al.*,<sup>14</sup> was applied.

#### *Determination of aldehyde group content in TEMPO-oxidized cotton fibers*

The aldehyde content in water-insoluble fractions was measured according to the method described in the literature.<sup>21,27</sup>

#### *Determination of moisture sorption*

Moisture sorption of oxidized cellulose fibers was determined according to standards (ASTM D, 2654-76, 1976).<sup>28</sup> Fibers were exposed to standard atmosphere: 20 ± 2 °C, 65 ± 2% relative humidity, for 24 h (ASTM D, 1776-74, 1974).<sup>29</sup> Moisture sorption was calculated as weight percentage of absolute dry material. Reported values are the mean values of three separate determinations. Variation among analyses was less than 8% in all cases.

#### *Determination of water retention value*

Water retention of cellulose fibers was determined in triplicate by the standard centrifuge method (ASTM D, 2402-78, 1978).<sup>30</sup> The variation coefficients were at the level of 10%.

#### *Surface morphology and elemental composition – scanning electron microscopy with energy dispersive X-ray analysis (SEM-EDX)*

Fiber morphology was investigated by scanning electron microscopy using a JEOL JSM 6610LV microscope, operating at 20 kV, after sputtering the samples with gold. The elemental composition was analyzed using an INCA Energy 350 Dispersive X-ray Microanalysis System.

#### *Determination of antimicrobial activity of TEMPO-oxidized cotton fibers with incorporated silver*

The agar diffusion test<sup>31,32</sup> was used to assess the antimicrobial activity of the TEMPO-oxidized cotton fibers with incorporated silver. Three test organisms were used: Gram-positive *S. aureus* (ATCC 25923), Gram-negative *E. coli* (ATCC 25922), and the yeast *C. albicans* (ATCC 24433).

## RESULTS AND DISCUSSION

It is known that catalytic oxidation using water-soluble and stable nitroxyl radicals, such as 2,2,6,6-tetramethylpiperidine-1-oxyl radical

(TEMPO), under aqueous conditions, has become one of the interesting routes to introduce carboxyl functionalities into polysaccharides.<sup>9-15,33</sup> The TEMPO-mediated oxidation of cellulose was described as causing selective oxidation at C6 of the anhydroglucose units to carboxyl groups via the intermediate aldehyde stage. The nitroxyl radical affects the oxidation from the alcohol to the aldehyde oxidation state, while the hypobromide generated *in situ* from hypochlorite and bromide performs the further oxidation of the aldehyde to the carboxyl groups.<sup>10,13</sup> Thus, oxidized cellulose contains both aldehyde and carboxyl groups.

The introduced carboxyl groups were used as reactive sites, *i.e.* the hydrogen atom present in carboxyl groups could be easily replaced with another cation, in this case, with silver ions (Fig. 1). Silver was incorporated into previously TEMPO-oxidized cotton fibers by chemisorption from aqueous silver nitrate solution, and after the ion-exchange process,<sup>34</sup> the reduction of the silver salt on the surface of the TEMPO-oxidized cotton fibers can lead to the formation of fine silver particles.<sup>35</sup>

In our previous article,<sup>14</sup> correlations between NaClO concentration, oxidation time and

carboxyl groups introduced into TEMPO-oxidized cotton fibers were shown in detail. In this paper, we discuss the relationship between oxidation conditions and silver sorption capacity, as well as the effect of structural changes, *i.e.* introduced functional groups, on the silver sorption capacity.

The effect of oxidation time and amount of the primary oxidant (NaClO) on silver sorption capacity of the TEMPO-oxidized cotton fibers is shown in Figure 2. The unmodified cotton fibers sorbed the lowest amount of silver (0.024 mmol/g fibers). In all other cases, where TEMPO-mediated oxidation was applied, the increase in the silver sorption by modified fibers was obtained, ranging from 0.102 to 0.806 mmol/g fibers. The obtained results show that silver sorption by modified cotton fibers increased with increasing either primary oxidant concentration or the duration of oxidation. The effect of reaction time on the silver content is more pronounced than the effect of the concentration of primary oxidant, especially in the case of higher NaClO concentrations (4.84 and 9.67 mmol NaClO/g fibers).

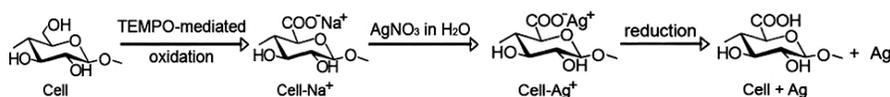


Figure 1: Scheme of TEMPO-mediated oxidation and synthesis of silver particles on TEMPO-oxidized cotton fibers<sup>35</sup>

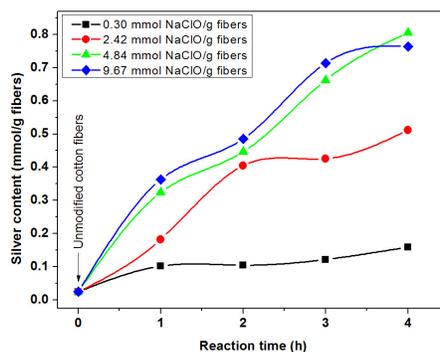


Figure 2: Relationship between oxidation time (h) and silver content (mmol/g fibers) of TEMPO-oxidized cotton fibers, where 0.30, 2.42, 4.84, and 9.67 mmol NaClO (per gram of fibers) was applied to the cellulose slurry, at RT and pH 10.5

To better understand the obtained results, we tried to determine the relationship between

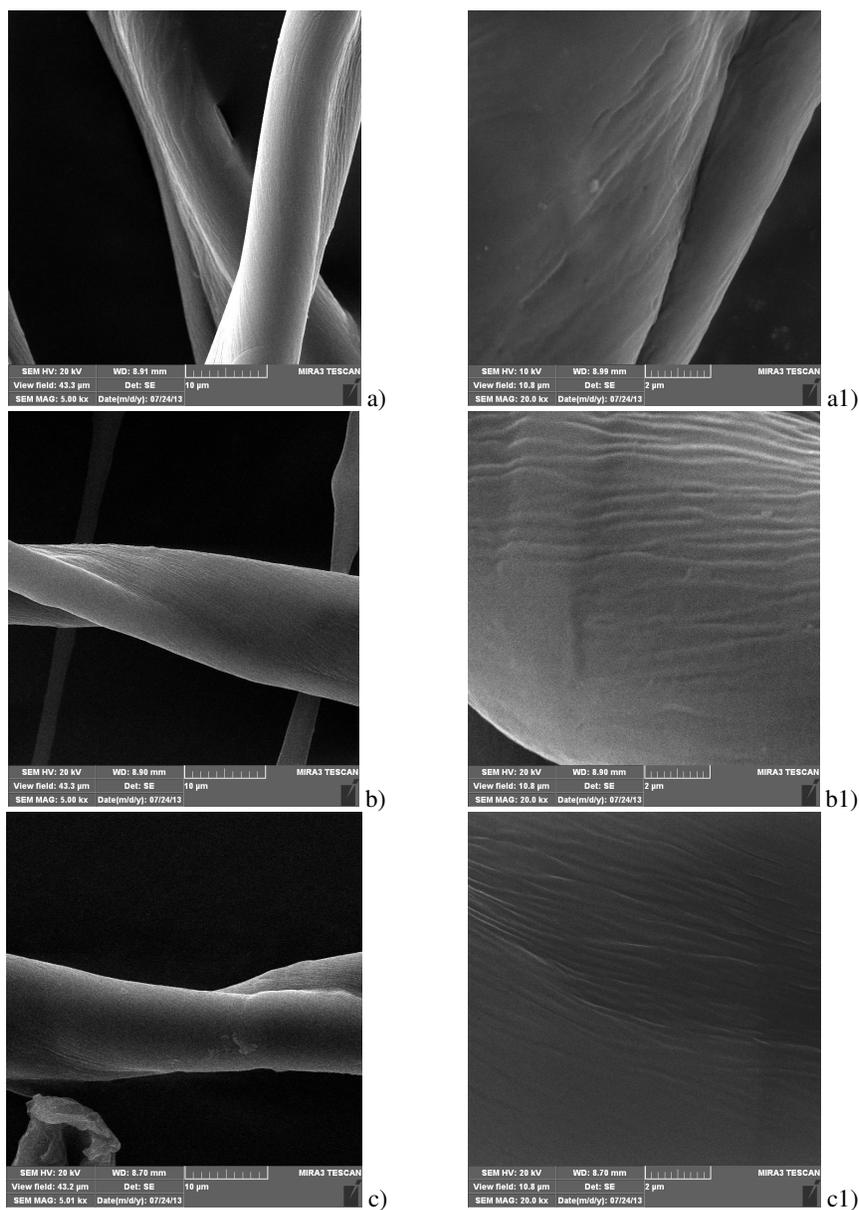
structural changes, introduced functional groups and silver sorption capacity of TEMPO-oxidized

cotton fibers. Depending on the oxidation conditions of TEMPO-mediated oxidation, together with the introduction of functional groups, the chemical composition and fibrous morphology of cotton fibers were changed.<sup>14,21</sup>

The structural changes between unmodified cotton fibers (a) and those modified by varying amounts of oxidative agent (b-e) may be seen in the SEM images shown in Figure 3. With increasing oxidative agent concentration, on the surface of modified fibers more pronounced ridges and cracks are observed, and the surface

presents signs of fibrillation (c, d). Rigorous oxidative conditions lead to damage, harm and fiber breakage (d).

The obtained morphological changes of the TEMPO-oxidized cotton fibers, together with the introduced hydrophilic carboxyl groups, cause changes in their sorption properties, which are very important for further fiber modification and/or application, in our case for silver sorption and possible application of oxidized fibers with sorbed silver in wound treatment.



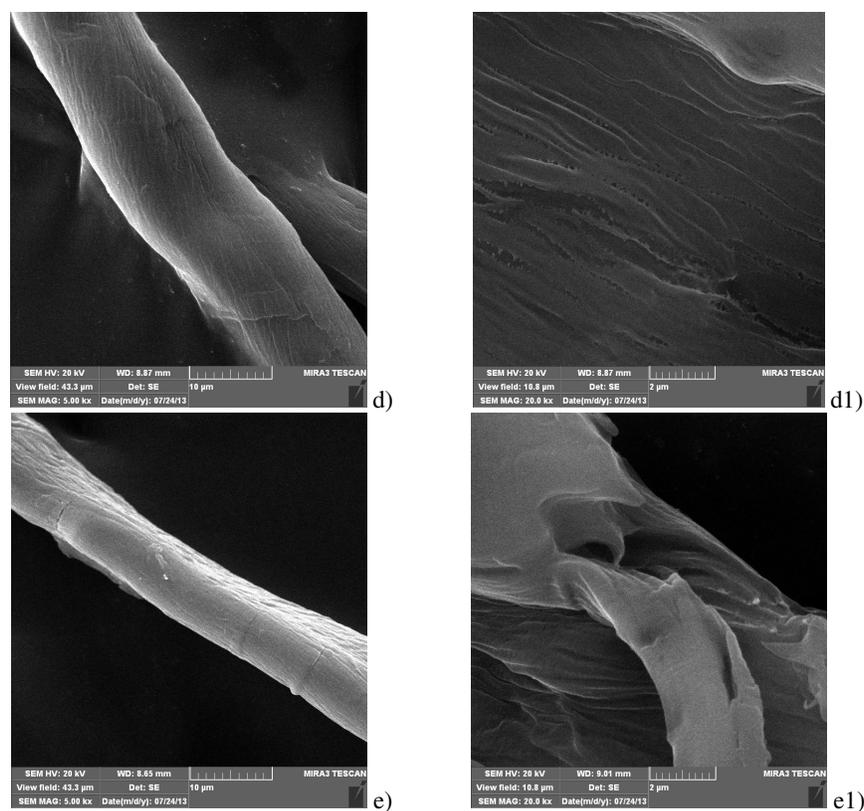


Figure 3: SEM images of a) unmodified and TEMPO-oxidized cotton fibers with b) 0.30; c) 2.42; d) 4.84; and e) 9.67 mmol NaClO (per gram of fibers), during 4 h, at RT and pH 10.5; Bar: 10  $\mu$ m and 2  $\mu$ m

The sorption properties of oxidized cotton fibers were evaluated by determining the moisture sorption and water retention value since water penetrates into the fiber in the form of vapor or in liquid state. The introduced carboxyl groups and free hydroxyl groups in the cotton fiber amorphous regions and on the crystallites' surfaces are responsible for the moisture sorption at 20 °C and up to 65% relative humidity, while higher relative humidity causes physical sorption due to capillary condensation. The changes in the moisture sorption of TEMPO-oxidized cotton fibers (for details see literature<sup>14</sup>) reflect changes in their chemical composition, crystallinity and pore structure,<sup>16,36</sup> all of them influencing the silver sorption capacity of oxidized cotton fibers (Fig. 4a). From the results shown, it is obvious that a linear dependence between the moisture sorption values (MSV) and the content of introduced silver in the TEMPO-oxidized cotton fibers is obtained, with the exception of the samples modified with the lowest concentration of NaClO, *i.e.* with 0.30 mmol NaClO/g fibers. As we concluded previously, with an increase of

the severity of the oxidation treatment, the moisture sorption value also increases,<sup>14</sup> and as a consequence of the increase of MSV, together with other changes that occurred during the TEMPO-mediated oxidation, the amount of sorbed silver increased too.

Water retention value is a measure of the swelling ability and accessibility of fibers to aqueous baths during wet finishing processes.<sup>37</sup> All water absorbing and holding surfaces, cracks and cavities are included in the water retention measurement. From the results shown in Figure 4b, it is evident that, with increased severity of the treatment, an increase in both water retention value and silver sorption is obtained. This can be explained by the fact that when fibers absorb water, they swell more and the fiber structures become more accessible for silver sorption. Additionally, during TEMPO-mediated oxidation, an effective decomposition of hydrophobic impurities and natural pigments in cotton fibers occurs,<sup>37,38</sup> which, along with morphological changes of the fibers (see Fig. 3d, e), leads to additional water and silver sorption into the fibers

from the aqueous solution. The water retention value increased from 21.55% to 85.36%, while the amount of sorbed silver, as mentioned above, increased from 0.102 mmol/g of fibers up to 0.806 mmol/g of fibers. The sample oxidized with 4.84 mmol NaClO/g fibers, for 4 h, which shows the maximum amount of sorbed silver (0.806 mmol/g fibers), sorbed 84% of water.

In order to better understand the influence of TEMPO-mediated oxidation on silver sorption capacity, we sought a deeper insight into the relationship between the introduced functional groups and the amount of silver deposited in the TEMPO-oxidized cotton fibers. Depending on oxidative conditions, TEMPO-oxidized cotton fibers contain both carboxyl and aldehyde groups.<sup>14</sup> The carboxyl groups along the entire surface of the modified cotton fibers can be used as reactive hooks to quantitatively introduce silver metal ions by an ion-exchange reaction,<sup>35</sup> while the aldehyde groups on the microfibrils serve as a reducing agent for silver ions.<sup>39</sup>

The expected result, that one carboxyl group reacts with one silver ion by the ion exchange mechanism in stoichiometry of 1:1,<sup>34,40</sup> was

obtained for the TEMPO-oxidized cotton fibers with a lower amount of carboxyl groups. In the case of TEMPO-oxidized cotton fibers with a higher amount of COOH groups, the amount of sorbed silver is smaller than the amount of carboxyl groups, with the exception of the sample oxidized with 4.84 mmol NaClO/g fibers, for 4 h, which showed the maximum amount of sorbed silver (0.806 mmol silver per gram of fibers) (Fig. 5a). This can be explained by the difference in affinity of carboxyl groups of cellulose towards various metal cations, which was noted by Heymann and Rabinov, and examined in detail by Davison.<sup>41</sup> The order of increasing affinity found by this author was  $N(CH_3)_4^+ < Li^+ < Na^+ < K^+ < TI^+ < Ag^+ < Ca^{2+}, Ba^{2+}$ . The applied Ca-acetate method for the determination of COOH groups and the stronger affinity of COOH groups for  $Ca^{2+}$  than for  $Ag^+$  could be the reason for the obtained results, *i.e.* the content of sorbed silver was smaller than the content of carboxyl groups. Furthermore, the obtained results are in agreement with the results obtained for silver sorption by oxidized hemp fibers.<sup>42</sup>

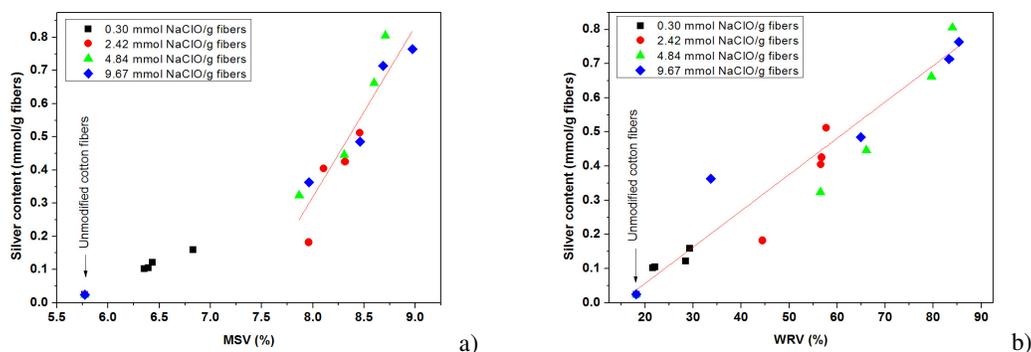


Figure 4: Relationships between: a) moisture sorption; b) water retention values; and silver content of unmodified and TEMPO-oxidized cotton fibers, during 1-4 h, at room temperature and pH 10.5

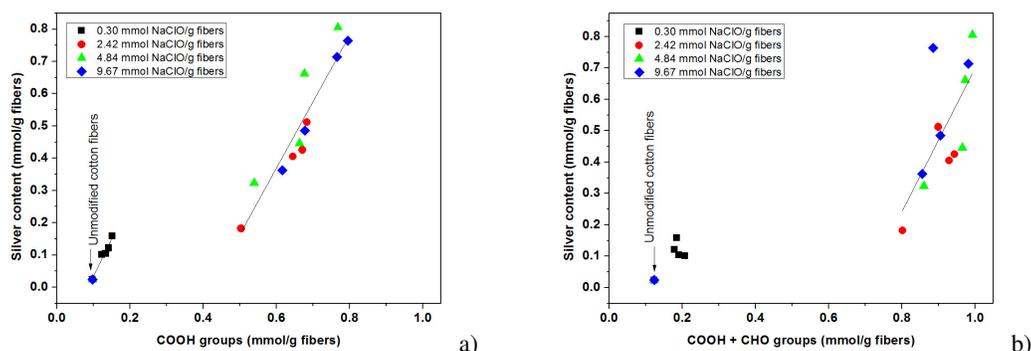


Figure 5: Relationships between a) COOH; b) COOH + CHO groups and silver content of unmodified and TEMPO-oxidized cotton fibers, during 1-4 h, at room temperature and pH 10.5

The influence of the aldehyde groups, naturally occurring in cellulose fibers and formed by TEMPO-mediated oxidation, on the amount of sorbed silver should not be neglected, since, as we mentioned, the aldehyde groups can serve as a reducing agent for silver ions.<sup>39</sup> The absence of correlation between aldehyde group content and deposited silver content (data not shown) can be explained by the combination of ion-exchange and reduction of  $\text{Ag}^+$  on cellulose fibers. In addition, silver exhibits propensity for auto-catalytic reduction, *i.e.*  $\text{Ag}^+$  reduction is accelerated in the presence of metallic silver, which acts as nuclei for further reduction of the ions.<sup>43-46</sup>

To summarize, the relationship between the sums of both groups, *i.e.* carboxyl and aldehyde groups, and the silver contents is shown in Figure 5b. Two groups of results have been obtained: the first one for the lowest concentration of oxidative agent  $\text{NaClO}$ , *i.e.* the content of functional groups less than 0.25 mmol/g, where the data are grouped without any regularity, and the second for the samples with the amount of functional groups higher than 0.5 mmol/g, for which linear dependence between the content of functional groups and the content of introduced silver in TEMPO-oxidized cotton fibers is obtained, but with a higher slope compared to the data presented in Figure 5a. The observed characteristic inflection step is evidence of mentioned combination of two processes: ion-exchange and reduction of  $\text{Ag}^+$  on cellulose fibers. However, it is difficult to determine the extent of  $\text{Ag}^+$  reduction since, in addition to  $\text{Ag}^+$  reduction during the sorption,<sup>47</sup> it proceeds further during and after fiber drying in air.<sup>35,48,49</sup>

The surface morphology of the TEMPO-oxidized cotton fibers with deposited silver was examined by SEM and the obtained microphotographs are presented in Figure 6. Microphotographs clearly show the effects of the ion-exchange process (Fig. 6a) and reduction of  $\text{Ag}^+$  to  $\text{Ag}^0$  by CHO groups (Fig. 6b) on silver deposition. Due to the presence of a high amount of COOH groups and a low amount of CHO groups, silver is present predominantly in the form of ions, *i.e.* silver particles are not observed on the surface of the fibers or they are present to a smaller extent, although the amount of incorporated silver is high. For example, the sample presented in Figure 6a has 0.7950 mmol

COOH/g fibers, 0.0907 mmol CHO/g fibers and a silver content of even 0.7640 mmol/g fibers, with just a few silver nanoparticles observed on the fiber surface. For the samples with a high content of aldehyde groups, the silver dominates in the form of nanoparticles (Fig. 6b). The elemental composition of the particles was identified by the SEM-EDX technique, and results show strong silver peaks in the spectra of the TEMPO-oxidized cotton fibers with deposited silver (data not shown). SEM images recorded under magnification of 100 000 times show silver particles (“white dots”), with isometric shapes and sizes ranging from 10 to 100 nm, which are quite uniformly distributed on the surface of the TEMPO-oxidized cotton fibers. Additionally, the silver particles are also present inside the fiber structure, as shown in Figure 7. The color of the TEMPO-oxidized cotton fibers with incorporated silver changed from cream yellow for oxidized fibers to brown in response to silver deposition. According to the literature,<sup>50</sup> the reduction of silver ions ( $\text{Ag}^+$ ) to silver metal ( $\text{Ag}^0$ ) is often achieved through the use of chemicals, such as sodium borohydride or hydrazine, which are rather hazardous chemicals.

Safer chemicals, such as sugar or alcohol, can also be used to reduce  $\text{Ag}^+$  to  $\text{Ag}^0$ , but then the use of surfactants to stabilize the nanoparticles is required. However, using cellulose fibers as a matrix for *in situ* preparation and incorporation of silver nanoparticles has the advantage that cellulose acts both as a reducing agent, due to the presence of aldehyde groups, and as a stabilizing agent, due to the steric effect of the cellulose chains.

#### Antimicrobial activity of TEMPO-oxidized cotton fibers with deposited silver

The antibacterial activity of the TEMPO-oxidized cotton fibers with incorporated silver was tested against strains: gram (+) *Staphylococcus aureus* (ATCC 25,923) and gram (-) *Escherichia coli* (ATCC 25,922), and their antifungal activity – against fungi *Candida albicans* (ATCC 24,433).

The antimicrobial activity for unmodified cotton fibers, TEMPO-oxidized cotton fibers (2.42 mmol  $\text{NaClO}$ /g fibers, 4 h) and silver-loaded TEMPO-oxidized cotton fibers (0.30, 2.42, 4.84 and 9.67 mmol  $\text{NaClO}$ /g fibers, 4 h; + silver) is shown in Table 1. Obviously, no antimicrobial

activity was observed for untreated and TEMPO-oxidized cotton fibers, while the incorporation of silver in TEMPO-oxidized cotton fibers generally inhibited the growth of the tested microbes. Among the tested microorganisms, Gram-positive bacteria strain *S. aureus* is the most sensitive to the TEMPO-oxidized cotton fibers with deposited silver, and the TEMPO-oxidized cotton fibers containing 0.714 mmol silver/g fibers and 0.806 mmol silver/g fibers were the most effective. Most of the tested samples showed good antimicrobial activity against gram (-) strains *Escherichia coli* and the fungi *Candida albicans*. The obtained results showed that the quantity of bonded silver, in all cases, is enough to develop desirable antimicrobial activity in the TEMPO-oxidized cotton fibers with deposited silver, although, there is no clear dose dependent

antimicrobial activity. The reason for this is probably because silver does not attack microorganisms directly; it operates as a catalytic agent and moreover, it is not consumed in this process.<sup>51</sup> According to the literature<sup>19</sup> the silver acts in a complex manner, *i.e.* silver binds to electron donor receptors, notably disulphide, amino, imidazole, carbonyl and phosphate residues on membranes leading to intracellular absorption. Inactivation of membrane-related enzymes results in denaturation of the bacterial cell envelope, which ultimately leads to the lethal effect on a cell. This mode of antimicrobial action does not necessarily depend on the quantity of silver, but rather on the availability of silver in a cellulose matrix and its ability to reach and react with microorganism cells.

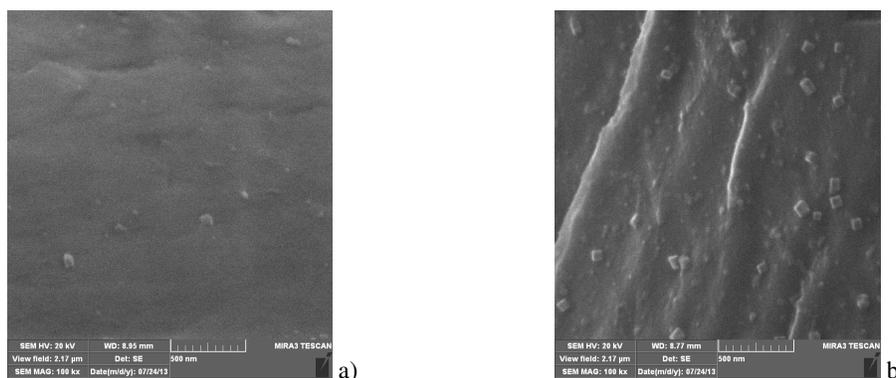


Figure 6: SEM images of TEMPO-oxidized cotton fibers with deposited silver showing the influence of two processes: a) ion exchange by COOH groups, in the presence of 0.7950 mmol COOH/g fibers, 0.0907 mmol CHO/g fibers and 0.7640 mmol Ag/g fibers; and b) reduction of  $\text{Ag}^+$  to  $\text{Ag}^0$  by CHO groups in the presence of 0.3211 mmol CHO/g fibers, 0.5392 mmol COOH/g fibers and 0.3236 mmol Ag/g fibers. Magnification and bar: 100 000x and 500 nm, respectively

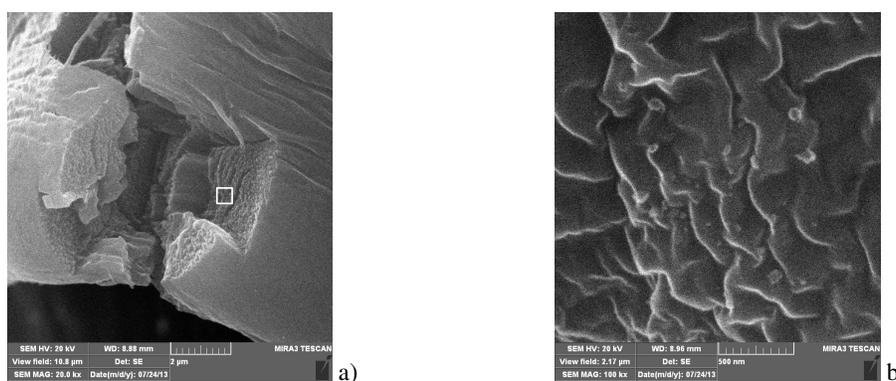


Figure 7: SEM images of TEMPO-oxidized cotton fibers with deposited silver showing the presence of silver inside the fiber structure, for the sample with 0.6769 mmol COOH/g fibers, 0.2964 mmol CHO/g fibers and 0.6620 mmol Ag/g fibers. Magnification and bar: a) 20 000x and 2 µm and b) 100 000x and 500 nm

Table 1  
Antimicrobial activity of TEMPO-oxidized cotton fibers with deposited silver

Oxidative conditions			Width of inhibition zone after 24 h (mm)		
NaClO, mmol/g fibers	Reaction time	Silver content (mmol/g fibers)	<i>S. aureus</i>	<i>E. coli</i>	<i>C. albicans</i>
0.00	0	0	0	0	0
2.42	4	0	0	0	0
0.00	0	0.024	<1.0, IC*	<1.0, IC*	<1.0, IC*
0.30	4.0	0.324	1.0 - 1.5	1.0	1.0 - 1.5
2.42	4.0	0.446	1.0 - 1.5	1.0 - 1.5	1.5
4.84	4.0	0.714	2.0 - 2.5	2.0 - 2.5	1.5
9.67	4.0	0.806	2.0 - 2.5	1.5	1.5

\*IC – inhibition in contact, with very small inhibition zone

## CONCLUSION

An attempt has been made to explain the roles of introduced functional groups and changes in the sorption properties of TEMPO-oxidized cotton on silver deposition.

Depending on oxidative conditions, TEMPO-oxidized cotton fibers contain both carboxyl and aldehyde groups. Carboxyl groups along the entire surface of modified cotton fibers can be used as reactive hooks to quantitatively introduce silver metal ions by an ion-exchange reaction, while the aldehyde groups on the microfibrils serve as a reducing agent for silver ions. Changes in fiber morphology, together with introduced hydrophilic carboxyl groups, cause changes in sorption properties of TEMPO-oxidized cotton fibers. Increases in water retention and moisture sorption positively affect subsequent silver sorption by TEMPO-oxidized cotton fibers.

Silver was incorporated into TEMPO-oxidized cotton fibers by a combination of  $\text{Ag}^+$  chemisorption from aqueous silver nitrate solution and reduction of  $\text{Ag}^+$  to  $\text{Ag}^0$  by cellulose itself. In the case of a high amount of COOH groups and a low amount of CHO groups, silver is incorporated predominantly in the form of ions, *i.e.* silver particles are not observed on the surface of the fibers or they are present to a smaller extent, although the amount of incorporated silver is high, while in the case of a high content of aldehyde groups, the silver is predominantly in the form of nanoparticles. Using cellulose fibers as a matrix for *in situ* preparation and incorporation of silver particles has the advantage that cellulose acts as both reducing agent and stabilizing agent. The amount of incorporated silver (in the form of  $\text{Ag}^+$  and  $\text{Ag}^0$ ) was in the range from 0.102 to 0.806 mmol/g fibers.

The antimicrobial activity of the silver loaded TEMPO-oxidized cotton fibers was confirmed against different pathogens: *C. albicans*, *S. aureus* and *E. coli*. The obtained results show that the quantities of incorporated silver, in all cases, are enough to develop desirable antimicrobial activity, although, there is no clear dose dependant antimicrobial activity.

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## REFERENCES

- <sup>1</sup> USDA ERS Cotton and Wool Outlook, December 2014, online at <http://www.ers.usda.gov/>, accessed 18 January, 2015.
- <sup>2</sup> J. Schurz, *Progr. Polym. Sci.*, **24**, 481 (1999).
- <sup>3</sup> D. Klemm, B. Heublein, H.-P. Fink and A. Bohn, *Chem. Int. Ed.*, **44**, 3358 (2005).
- <sup>4</sup> G. Cerchi and M. Tullio, European Patent No. 1676956 (2006).
- <sup>5</sup> E. J. McGinley and D. C. Tuason Jr., US Patent No. 5192569 (1993).
- <sup>6</sup> P. Kleinebudde, M. Jumaa and F. El Saleh, *AAPS Pharm. Sci.*, **2**, 5 (2000).
- <sup>7</sup> S. Strnad, O. Sauperl and L. Fras-Zemljic in "Biopolymers", edited by M. Elnashar, Sciyo, 2010, pp. 181-200.
- <sup>8</sup> R. L. Stilwell, M. G. Marks, L. Saferstein and D. M. Wiseman, in "Handbook of Biodegradable Polymers", edited by A. Doms, J. Kostand and D. Wiseman, Harwood Academic Publishers, Amsterdam, 1997, pp. 291-306.
- <sup>9</sup> A. E. de Nooy, A. C. Besemer and van H. Bekkum, *Carbohydr. Res.*, **269**, 89 (1995).

- <sup>10</sup> P. S. Chang and J. F. Robyt, *J. Carbohydr. Chem.*, **15**, 819 (1996).
- <sup>11</sup> C. Tahiri and M. R. Vignon, *Cellulose*, **7**, 177 (2000).
- <sup>12</sup> Z. Dang, J. Zhang and A. Ragauskas, *Carbohydr. Polym.*, **70**, 310 (2007).
- <sup>13</sup> T. Saito, Y. Okita, T. T. Nge, J. Sugiyama and A. Isogai, *Carbohydr. Polym.*, **65**, 435 (2006).
- <sup>14</sup> J. Praskalo, M. Kostic, A. Potthast, G. Popov, B. Pejic *et al.*, *Carbohydr. Polym.*, **77**, 791 (2009)
- <sup>15</sup> A. E. de Nooy, A. C. Besemer and H. van Bekkum, *Recueil des Travaux Chimiques Pays-Bas*, **113**, 165 (1994).
- <sup>16</sup> T. Kreze, S. Jeler and S. Strnad, *Mater. Res. Innov.*, **5**, 277 (2002).
- <sup>17</sup> Q. L. Feng, J. Wu, G. Q. Chen, F. Z. Cui, T. N. Kim *et al.*, *J. Biomed. Mater. Res.*, **52**, 662 (2000).
- <sup>18</sup> U. Wollina, M. B. Abdel-Nase and S. Verma, in "Biofunctional Textiles and the Skin", edited by U.-C. Hipler and P. Elsner, Curr. Probl. Dermatol., Karger, Basel, 2006, vol. 33, p. 1
- <sup>19</sup> A. B. G. Lansdown, in "Biofunctional Textiles and the Skin", edited by U.-C. Hipler and P. Elsner, Curr. Probl. Dermatol., Karger, Basel, 2006, vol. 33, p. 17.
- <sup>20</sup> M. Heide, U. Mohring, R. Hansel, M. Stoll, U. Wollina *et al.*, in "Biofunctional Textiles and the Skin", edited by U.-C. Hipler and P. Elsner, Curr. Probl. Dermatol., Karger, Basel, 2006, vol. 33, pp. 179.
- <sup>21</sup> T. Saito and A. Isogai, *Biomacromolecules*, **5**, 1983 (2004).
- <sup>22</sup> M. M. Kostic, J. Z. Milanovic, M. V. Baljak, K. Mihajlovski and A. D. Kramar, *Fiber. Polym.*, **1**, 57 (2014).
- <sup>23</sup> Lj. Rajakovic, T. Djurkic and A. Peric-Grujic, in "Analytical Chemistry: Quantitative Chemical Analysis", (in Serbian), TMF, Belgrade, 1999, pp. 192.
- <sup>24</sup> P. Patnaik, in "Handbook of Environmental Analysis", Lewis Publishers, CRC Press, Boca Raton, London, New York, 1997, pp. 39.
- <sup>25</sup> V. Kumar and T. Yang, *Carbohydr. Polym.*, **48**, 403 (2002).
- <sup>26</sup> E. C. Yackel and W. O. Kenyon, *J. Am. Chem. Soc.*, **64**, 121 (1942).
- <sup>27</sup> E. J. Parks and R. L. Hebert, *Tappi J.*, **55**, 1510 (1972).
- <sup>28</sup> ASTM D 1776-74 (1974) Conditioning textiles and textile products for testing.
- <sup>29</sup> ASTM D 2402-78 (1978) Standard test method for water retention of fibers (centrifuge method).
- <sup>30</sup> ASTM D 2654-76 (1976) Moisture content and moisture regain of textiles.
- <sup>31</sup> B. G. Joiner, in "Bioactive Fibers and Polymers", edited by J. V. Edwards and L. T. Vigo, American Chemical Society, Washington, DC, 2001, pp. 201.
- <sup>32</sup> J. Buchenska, *J. Biomater. Sci. Polym.*, **12**, 55 (2001).
- <sup>33</sup> T. Saito, I. Shibata, A. Isogai, N. Suguri and N. Sumikawa, *Carbohydr. Polym.*, **61**, 414 (2005).
- <sup>34</sup> T. Saito and A. Isogai, *Carbohydr. Polym.*, **61**, 183 (2005).
- <sup>35</sup> S. Ifuku, M. Tsuji, M. Morimoto, H. Saimoto and H. Yano, *Biomacromolecules*, **10**, 2714 (2009).
- <sup>36</sup> B. Pejic, M. Kostic, P. Skundric and J. Praskalo, *Bioresour. Technol.*, **99**, 7152 (2008).
- <sup>37</sup> D. Fakin, V. Golob, K. S. Kleinschek and A. M. L. Marechal, *Textile Res. J.*, **76**, 448 (2006).
- <sup>38</sup> K. Stana-Kleinschek, S. Strnad and V. Ribitsch, *Polym. Eng. Sci.*, **39**, 1412 (1999).
- <sup>39</sup> M. Wu, S. Kuga and Y. Huang, *Langmuir*, **24**, 10494 (2008).
- <sup>40</sup> R. S. Simonson, J. A. Cuculo and L. C. Wadsworth, *Textile Res. J.*, **49**, 352 (1979).
- <sup>41</sup> A. M. Sookne and M. Harris, in "Cellulose and Cellulose Derivatives", edited by E. Ott, H. M. Spurlin and M. W. Grafflin, Interscience Publishers, New York, 1954, pp. 213.
- <sup>42</sup> J. Milanović, T. Mihailović, K. Popović and M. Kostić, *J. Serb. Chem. Soc.*, **77**, 1759 (2012).
- <sup>43</sup> M. Harada and E. Katagiri, *Langmuir*, **26**, 17896 (2010).
- <sup>44</sup> K. Kato, *Textile Res. J.*, **29**, 409 (1959).
- <sup>45</sup> T. Rabilloud, L. Vuillard, C. Gilly and J. J. Lawrence, *Cell. Mol. Biol.*, **40**, 57 (1994).
- <sup>46</sup> H. E. Emam, A. P. Manian, B. Široká, H. Duelli, B. Redl *et al.*, *J. Clean. Prod.*, **39**, 17 (2013).
- <sup>47</sup> N. E. Kotelnikova, M. Stoll, G. Wegener, E. Windeisen, E. Wenzkowski *et al.*, *Cellulose Chem. Technol.*, **37**, 51 (2003).
- <sup>48</sup> N. E. Kotelnikova, V. N. Deminov, G. Wegener, E. Windeisen and V. P. Kotelnikov, *Cellulose Chem. Technol.*, **37**, 225 (2003).
- <sup>49</sup> J. Cai, S. Kimura, M. Wada and S. Kuga, *Biomacromolecules*, **10**, 87 (2008).
- <sup>50</sup> S. T. Dubas, P. Kumlangdudsana and P. Potiyaraj, *Colloid. Surf., A: Physicochem. Eng. Aspects*, **289**, 105 (2006).
- <sup>51</sup> R. L. Davis and S. F. Etris, *Catal. Today*, **36**, 107 (1997).