Fashion and Textiles

RESEARCH Open Access

Development of cellulosic-based hemostatic dressing with antibacterial activity

Mostafa Goodarz[1], Amir Behzadnia[2] and Hassan Mohammadi[3*]

*Correspondence:
hassan.mohammadi@tx.iut.ac.ir

[1] Department of Textile Engineering, Amirkabir University of Technology, Tehran, Iran
[2] Textile Department, Science and Research Branch, Islamic Azad University, Tehran, Iran
[3] Department of Handicrafts, Isfahan University of Art, Isfahan, Iran

Abstract

A cotton-based hemostatic dressing featuring antibacterial properties was developed with the potential of being used in traffic accidents to control hemorrhage. Cotton gauze was oxidized initially in an acidic medium and then coated by PVA nanofibers and/or PVA nanofibers loaded with Ciprofloxacin. Fabricated dressings were characterized by FTIR analysis and SEM images. The FTIR spectrum showed the existence of carboxyl groups on the oxidized cotton gauze's surface. The carboxyl groups content was estimated to be 17.3 ± 0.3 for the oxidized sample with a mixture of nitric acid and phosphoric acid for 24 h (OCF-Mixed acid24). Moreover, the effect of the exposure duration of cotton gauze in the acidic medium on the blood coagulation activity was assessed. It was observed that the OCF-Mixed acid24 sample exhibited an agreeable hemostatic activity (BCIs = 10). The antibacterial activity against *E. coli* and *S. aureus* bacteria was also captured for the coated cotton gauze by the PVA nanofibers loaded with Ciprofloxacin.

Keywords: Cotton gauze, Hemostatic, Nanofibers, Antibacterial activity, Electrospinning

Introduction

Profuse bleeding from the wound is one of the most prevalent causes of death in traffic accidents. It has been reported that uncontrolled bleeding causes more than 30% of traumatic deaths (Yu & Zhong, 2021; Zhao et al., 2021). If effective and timely measures could be taken to control bleeding at the early stages of wound healing, the mortality rate would be significantly reduced (Zhao et al., 2021).

An ideal hemostatic dressing must have some desirable characteristics, such as acceleration of clot formation, rapid interruption of various arteriovenous hemorrhages, adequate durability, a barrier to further microbial contamination, and applicability in extreme environments (Edwards et al., 2021; Zhao et al., 2021).

Natural polysaccharides, such as cellulose materials, are widely used for hemostatic products due to their unique properties, such as good biocompatibility, non-toxicity, non-stimulation, easy processing and low cost (Zhu et al., 2021).

Hemostatic dressings with accelerating properties for surface hemostasis have been developed in many different structures such as hydrogels, powers, fabrics, sponges,

membranes, and so on (Chen et al., 2018; Liang et al., 2016; Xia et al., 2015; Yu et al., 2019; Zhao et al., 2018). Fabric hemostatic dressings have advantages over other structures, such as strong mechanical properties, the possibility of wrapping the wound in the form of a bandage, and easy removal after treatment (Li et al., 2019; Yang et al., 2019; Zhu et al., 2018).

Although many efficient hemostatic agents have been manufactured and used clinically in the last two decades, the traditional hemostatic bandage, cotton gauze, is still used for compressible and non-compressible wounds due to its desirable properties such as safety, non-allergenicity, low cost, conformability, breathability, stability, blood absorption capacity, and ease of application (Leonhardt et al., 2019; Li et al., 2017; Shin et al., 2017; Wang et al., 2019; Yu et al., 2019; Yuk et al., 2019).

Electrospinning is a simple and comparatively versatile method for producing nanometer-scale fibers; it can be used to fabricate ultrafine 1D nanofibers, 2D nonwoven membranes and 3D scaffolds (Li et al., 2021).

Electrospun nanofibers with such unique qualities like high surface-to-volume ratio and considerable porosity have a great potential for diverse applications; these include scaffolds for tissue engineering, carriers for drug delivery and wound dressings (Asadi et al., 2020; Garg & Bowlin, 2011; Islam et al., 2019; Li et al., 2021; Memic et al., 2019).

Polyvinyl alcohol (PVA) is an appropriate material for electrospinning due to its good viscosity, noteworthy electrical conductivity and surface tension. It is widely used in medical applications due to its hydrophilicity as well as polar nature, making it biocompatible as well as biodegradable (Rafieian et al., 2021).

The antibacterial property of a wound dressing is an essential parameter, since microbial infections would lead to inflammation (Li et al., 2016). Therefore, Ciprofloxacin (Cipro) has been used as a model drug herein, because it is a quinolone antibiotic with a wide range of activity against both gram-negative and gram-positive pathogens; it is also widely used in the treatment of various bacterial infections on both humans and animals (Cao et al., 2019; Cormier et al., 2012; Thairin & Wutticharoenmongkol, 2021).

Different works have been done on the preparation of cotton-based hemostatic dressing for bleeding control (Zhang et al., 2020; Zheng et al., 2021); this has been done through various methods such as oxidation with HNO3/H3PO4-NaNO2 (Gajdziok & Vetchý, 2015) and carboxymethylation (Wang et al., 2020). In other studies, hemostatic dressings containing antibacterial agents like zinc oxide have been used to produce antibacterial hemostatic dressings (Shefa et al., 2019). However, as far as we know, the surface coating of cotton-based hemostatic dressing by nanofibers containing antibacterial agents had not been investigated before. In this research study, therefore, a cellulose-based hemostatic dressing with antibacterial activity was developed. For this purpose, a cotton gauze as a base material was used and two methods (HNO3/H3PO4-NaNO2 oxidized cotton gauze and carboxymethylated cotton gauze) were utilized to make hemostatic wound dressing; subsequently, coating was done with PVA nanofibers (with and without Ciprofloxacin). The composite dressing's hemostatic ability and antibacterial activity were then evaluated by the blood coagulation test under in vitro conditions and the Disk Diffusion Susceptibility Test method, respectively.

Methods

Materials

Medical absorbent cotton gauze (yarn count: 32 Ne, weight: 33 gm^{-2}) was purchased from Sepehr co. (Esfahan, Iran). Sodium nitrite, 68% (w/w) nitric acid solution, 85% (w/w) phosphoric acid solution and acetone, PVA (Mw: 85,000–124,000 $gmol^{-1}$, 98% hydrolyzed), calcium acetate, and monochloroacetic acid were all purchased from Merck. Blood containing citrate (CB) was then purchased from IUT People's Hospital (Esfahan, Iran). All applied chemicals for the investigations in this study had the analytical grade.

Preparation of HNO_3/H_3PO_4–$NaNO_2$ oxidized cotton gauze

Mixing of nitric acid and phosphoric acid was initially done based on a ratio of 2:1 (v/v). A cotton gauze sample with the 1:30 (w/v) liquor ratio was then immersed in the mixture solution; this was followed by adding 1.4% (w/v) sodium nitrite. The covering of the reaction vessel was done by applying a petri dish for the purpose of preventing reddish-brown vapors from becoming airborne. The mixture solution was gently swirled for 1, 8 and 24 h under the exclusion of light and at room temperature. Then, the cotton gauze was thoroughly rinsed with deionized water and then subjected to soaking in a 0.5% (w/w) propanetriol solution for a period of 20 min and at the ambient temperature for the purpose of removing the oxidant. Eventually, the washing of the oxidized fabrics was done using acetone; this was followed by air-drying at 60 °C for about 30 min and cooling to room temperature (Xu et al., 2014b). Oxidized cotton gauze samples, using this method for 1, 8 and 24 h, were then designated by OCF-Mixed acid1, OCF-Mixed acid8 and OCF-Mixed acid24, respectively.

Preparation of the carboxymethylated cotton gauze

A cotton gauze sample with the 1:40 (w/v) liquor ratio of was subjected to immersion for 30 min at room temperature in an alkaline solution. The alkaline solution (with a concentration of 2.5% (w/v)) was composed of sodium hydroxide as the solute and 85% ethanol as the solvent. Then a monochloroacetic acid solution (20 mL) with a concentration of 10 wt% was added to the previous solution (80 mL) and the reaction was continued at the temperature of 70 °C for a period of 3 h. The samples were removed; the excess alkali on the fabric was neutralized with a hydrochloric acid solution (0.1 mol/L). Finally, carboxymethylated cotton gauzes (MCA) were washed several times with 85% ethanol solution and dried in an oven at 80 °C (Wang et al., 2020a, b). Oxidized cotton gauze sample, using this method, was designated by OCF-MCA in the following steps.

Preparation of the coated sample with the PVA nanofibers

To coat the oxidized cotton gauze with the PVA nanofibers, a PVA solution with a concentration of 8% (w/v) was developed at the beginning through the dissolution of PVA in a hot distilled water (90 °C). In addition, Ciprofloxacin (as an antibiotic) with the concentration of 2% (w/w) was added to the PVA solution to fabricate drug-loaded PVA nanofibers. Electrospinning was then performed at room temperature

with 30% humidity; this was done with a positive voltage of 18 kV and a syringe pump with a volumetric flow rate of 0.125 mL h^{-1}, with a 15 cm distance between the needle and the collector, i.e., oxidized cotton gauze. In the following, OCF-PVA and OCF-PVA-Cipro refer to the coated OCF sample with PVA nanofibers and the coated OCF sample with Ciprofloxacin-loaded PVA nanofibers, respectively.

Determining the carboxyl content

Determination of the carboxyl groups content in the oxidized samples was done based on the method described in the United States Pharmacopeia (USP, 1995).

Whenever the carboxyl groups of the oxidized samples and calcium acetate (a salt of the weaker acid) react with each other, an oxidized sample salt and a corresponding amount of the weaker acid will be formed. Accordingly, the following procedure was developed to determine the carboxyl content in the oxidized cotton gauze. Treatment of the oxidized samples (0.5 g) was done with 0.01M HCl for 1 h and then washing was carried out with deionized water. After that, a calcium acetate solution (50 mL) with a concentration of 2% (w/w) was added to the oxidized samples. After vibrating by ultrasound for a period of 30 min to ease exchange, the titration of the mixture was done using 0.1M standardized sodium hydroxide by applying phenolphthalein to serve as an indicator. The carboxyl content of the samples was estimated (for three replicates) by the following equation:

$$-COOH(\%) = \frac{0.1M \times V_{NaOH} \times MW_{COOH}}{m \times \left(1 - \frac{w}{100}\right)} \times 100$$

Here, 0.1M refers the NaOH's normal concentration, V_{NaOH} indicates the volume (mL) of the NaOH solution applied in the titration, m represents the oxidized samples' weight (mg), and w stands for the moisture content of the samples (%) (Xu et al., 2014a, b).

Characterization of the prepared samples

The morphology of the prepared samples was studied by applying SEM (Serontechnologies Korean, AIS2100). The coating of the samples was done using an ultrathin gold layer (thickness 20 nm) by a sputter coater (SC7620) for 180 S. The analysis of the resulting images was done by applying the Digimizer software to determine the electrospun nanofibers mean diameters (100 fibers). A Fourier transform infrared spectrometer (FT/IR-4100(JASCO Inc.)) was then used for the chemical analysis of cotton gauze (CF), oxidized cotton gauze (OCF), pure PVA and PVA-Cipro nanofibers. The recording of all spectra was done at the 4000–500 cm^{-1} wavelength with the resolution of 4 cm^{-1}.

Water retention ratio

To determine the water retention capacity of cotton gauze (CF), oxidized cotton gauze (OCF), and coated OCF with the PVA nanofibers (OCF-PVA), at first, the weight of the samples (2×2 cm^2) was recorded (m_{dry}); then they were immersed in deionized water for 30 min and at the ambient temperature (25 ± 2 °C). After that, the removal of the samples was done and the remaining deionized water on the surface of the samples was absorbed by a filter paper; then the wet weight (m_{wet}) was recorded. The described

procedure was repeated 3 times for each type of sample and the water retention ratio (W_r) was calculated using the following equation:

$$W_r = \left[(m_{wet} - m_{dry})/m_{dry}\right] \times 100$$

Tensile properties of the oxidized samples

A thickness gauge (Rees Sanj, Iran) with the accuracy of 0.01 mm and pressure of 1 kPa was used in at least 5 random positions of the sample to determine its thickness. The sample's thickness mean values were applied to calculate the mechanical properties. The use was then made of a tensile tester (Zwick Universal Testing Machine-1446 60, Germany) to determine the tensile strength of cotton gauze and oxidized cotton gauze according to the Standard Method for Breaking Force and Elongation of Textile Fabrics (ASTM D 5035–95) based on a constant elongation rate (CRE method) (Edwards et al., 2001). The test speed and gauge length were 100 mm/min and 70.0 mm, respectively. Each of the samples with the 10×2.5 cm^2 dimensions were tested ten times and the mean values were reported. All these measurements were done under standard conditions (at the temperature of 20 °C and relative humidity of 65.0%).

Assessment of bacterial inhibition

The antibacterial activity of the PVA-coated OCF samples against *E. coli* as a gram-negative bacterium and *S. aureus* as a gram-positive bacterium was determined by the Disk Diffusion Susceptibility Test method. Accordingly, samples with a diameter of 6.4 mm were placed in the culture medium overnight at 37 °C. The zone of inhibition was observed and measured three times after 24 h of incubation (Lemraski et al., 2021).

In vitro Hemostatic test

To evaluate the hemostasis effect of the samples in vitro, blood clotting indices (BCI) were measured. Toward this goal, the following steps were carried out. First, CB (100 µL) was dispersed in deionized water (25 mL) and its absorbance was measured at 540 nm as a negative control. Then, samples with a size of 1×1 cm^2 were placed in a culture dish and 100 µL of CB was dropped on each of them. After that, 10 µL of calcium chloride solution (0.2 mol L^{-1}) was dropped on the samples surface. The incubation of all samples was done at the temperature of 37 °C for a period of 5 min. Then, addition of deionized water (25 mL) to the culture dish was done without disturbing the clotted clots; the incubation of the samples was done again for a period of 10 min. Finally, the resulting solution's absorbance was measured at 540 nm by applying a visible light spectrophotometer (Phiztech, Iran). The described process was repeated 3 times for each of the samples and calculation of BCI was done by employing the following equation:

$$BCI = \frac{A_{RS}}{A_{NC}},$$

where ARS refers to the resulting solution's absorbance for different samples at 540 nm and ANC is the absorbance of CB (100 µL) dispersed in deionized water (25 mL) at 540 nm (negative control).

Result and Discussion

Morphology of the dressing

SEM images were captured from the surfaces of the cotton gauze prior to and after the oxidization process to assess the surface morphology. Figure 1 shows the surfaces of the untreated cotton gauze and oxidized cotton gauze with a mixture consisting of phosphoric acid and nitric acid. A smooth surface was observed for the untreated cotton gauze (CF), while the oxidized cotton gauze (OCF) showed a roughened surface due to the oxidation process (Wang et al., 2020a, b).

Antibacterial property was added to the oxidized cotton gauzes through electrospinning PVA nanofibers (with or without Ciprofloxacin) on their surfaces. Figure 2 shows the electrospun PVA nanofibers on the CF substrate. No bead formation was observed and PVA nanofibers had a mean diameter of 261 nm. However, the mean diameter of the PVA nanofibers was reduced about 25% (196 nm) when loaded with Ciprofloxacin (i.e., an antibacterial agent). This could be attributed to the polar groups of Ciprofloxacin, which might have increased the electrical conductivity of the electrospinning solution. Thus, with the increase of the electrical conductivity of the polymer solution, the electrostatic repulsion overcame the viscosity of the polymer jet, resulting in finer fibers (Uhljar et al., 2021).

Carboxyl content in the oxidized cotton gauze

The carboxyl group content is a useful index to gain insights into the successful oxidation procedure of cellulose substrates as well as the preparation of hemostatic dressings. Therefore, two procedures were considered to oxidize the cotton gauze (i.e., mixed acid and MCA methods). Based on the results, higher oxidation efficiency (see Table 1) was obtained for the mixed acid method. It was also observed that long-term

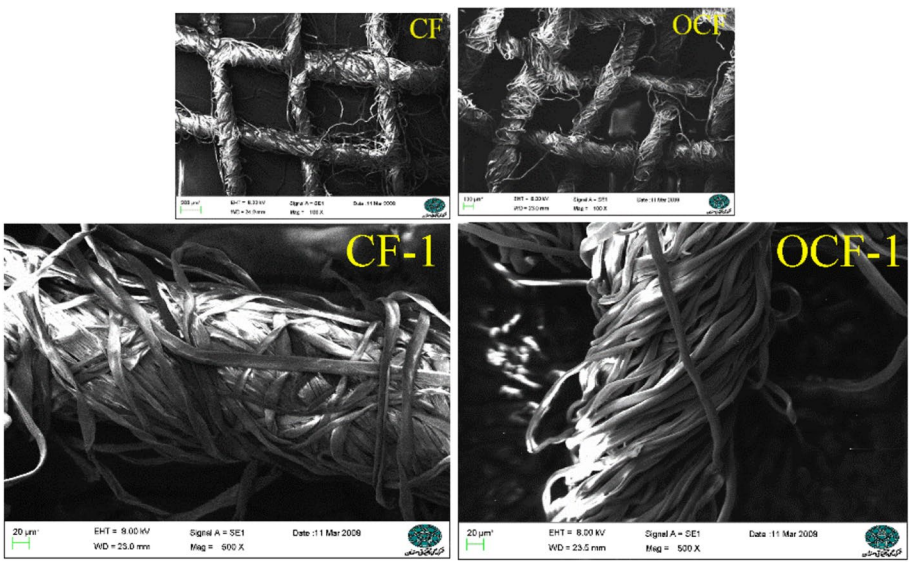

Fig. 1 Scanning Electron Microscope (SEM) images of Cotton Gauze 100 × Magnified (**a**), 500 × Magnified (**b**) and Oxidized Cotton Gauze 100 × Magnified (**c**), and 500 × Magnified (**d**)

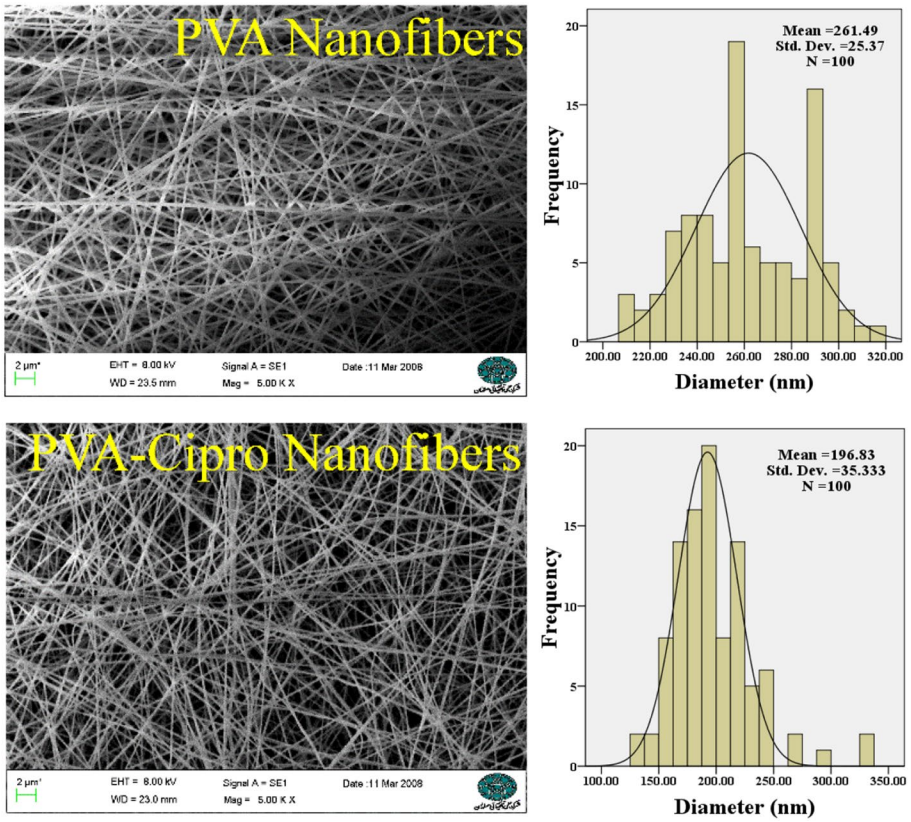

Fig. 2 Electrospun PVA nanofibers (**a**) and ciprofloxacin loaded electrospun PVA nanofibers (**b**) coated on the surface of oxidized cotton gauze

Table 1 Carboxyl content of oxidized cotton gauze

Oxidation method				
Mixed acid			MCA	
1 h	8 h	24 h		
2.7 ± 0.5	9.6 ± 0.3	17.3 ± 0.3	8.3 ± 1.1	Carboxyl content (%)

exposure to mixed acid medium (24 h) led to the higher content of carboxyl groups (Xu et al., 2014a, b).

FT-IR analysis

FT-IR spectra of the grey cotton gauze and the oxidized cotton gauze for 8 and 24 h are shown in Fig. 3. All samples displayed some broad peak in the 3100 to 3500 cm^{-1} range as a result of OH stretching vibration. Regarding the oxidized samples, it was observed that the absorption peak of the OH stretching vibration was shifted to a higher wavenumber as the duration of the oxidation process was increased. This might due to the weakened hydrogen bonds between the cellulose chains. Thus, the cotton yarns' crystalline structure was weakened by the long-term oxidation. Some sharp absorption peak of the C=O stretching vibration of the carbonyl group appeared clearly at 1739 cm^{-1}. The

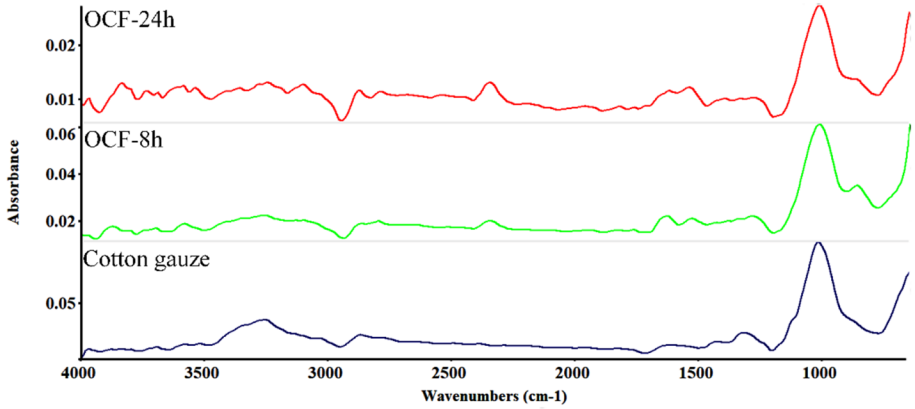

Fig. 3 FTIR spectrum of untreated cotton gauze and oxidized cotton gauze with mixture of nitric acid and phosphoric acid for 8 h (OCF-8 h) and 24 h (OCF-24 h)

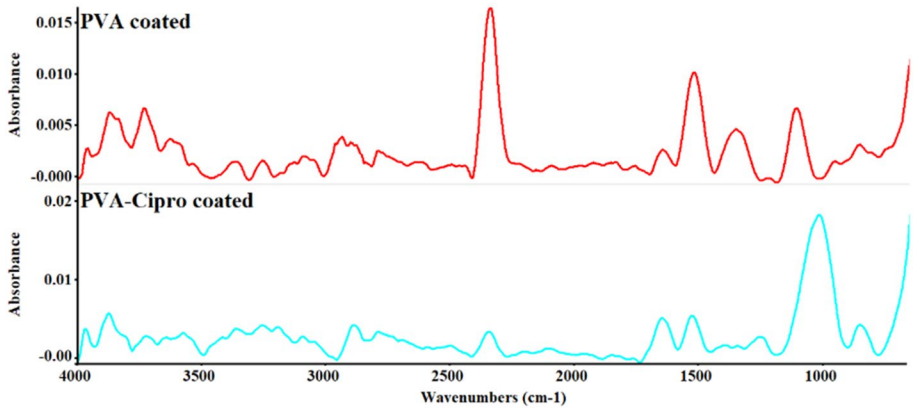

Fig. 4 FTIR spectrum of PVA and Ciprofloxacin loaded PVA nanofibers coated cotton gauze

intensity of this absorption peak was raised with increasing the duration of the oxidation process. This corresponded to the increase of the carboxyl groups in the oxidized fibers (Xu et al., 2014a, b).

To confirm the presence of both PVA nanofibers and Ciprofloxacin on the surface of the coated OCF sample, the FTIR spectra were examined in regard to the surface of the coated OCF sample with PVA (referred to as PVA-coated) and PVA nanofibers loaded with Ciprofloxacin (referred to as PVA-Cipro coated) (Fig. 4). The peak in the 3400 cm^{-1} region was associated to the stretching vibration of the hydroxyl group of PVA. In the FTIR spectrum of the coated OCF sample with PVA-Cipro, there was a prominent characteristic peak between 3500 and 3450 cm^{-1}. This peak resulted from the stretching vibration of the OH group, which could be attributed to intermolecular hydrogen bonding. There was another peak at 3000–2950 cm^{-1} that corresponded to the stretching of alkenes and aromatic C-H. The presence of a peak at 1750 to 1700 cm^{-1} indicated the C=O stretching of the carbonyl group. Furthermore, the peak at 1450 to 1400 cm^{-1} resulted from the C–O group and the one at 1300 to 1250 cm^{-1} corresponded to the bending vibration of the O–H group, thus indicating the carboxylic acid presence. Moreover, there was a strong adsorption peak between 1050 and 1000 cm^{-1} for the

Table 2 Mechanical properties of cotton gauze before and after oxidation by mixture of nitric acid/phosphoric acid (mixed acid) and monoacro acetic acid (MCA)

Sample	Stress		Strain		Young's modulus	
	MPa	SD	%	SD	MPa	SD
Cotton gauze	6.83	2.11	7.25	1.17	62.32	11.24
Mixed acid	0.84	0.33	20.18	4.12	2.83	1.15
MCA	2.37	0.43	24.98	7.38	0.53	0.62

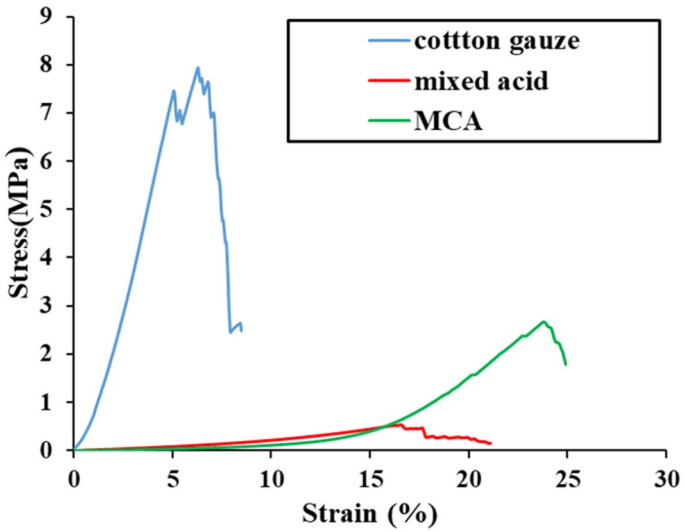

Fig. 5 Stress–strain diagram of cotton gauze before and after oxidation by mixture of nitric acid / phosphoric acid (mixed acid) and monoacro acetic acid (MCA)

C-F group, thus confirming the presence of Ciprofloxacin in the electrospun nanofibers (Sahoo et al., 2011).

Tensile strength results

As a medical dressing, cotton gauze must have a reasonable tensile strength. Due to the use of modifiers, the degree and method of modification, the strength of the oxidized cotton gauze was decreased significantly, thus making post-processing and application more difficult (Dai et al., 2013).

To address the impact of the oxidation methods on the cotton gauze strength, the tensile strength of the samples was measured before and after oxidation processes. Table 2 and Fig. 5 show the tensile properties of the untreated and oxidized cotton gauze. It should be noted that reported data were the mean of three replicates. Tensile strength of the cotton gauze was decreased due to the oxidation processes. It was also found that oxidizing cotton gauze with a mixture of acid (nitric acid and phosphoric acid) resulted in inferior tensile properties, as compared to doing this by monochloroacetic acid. Such inferior tensile properties could be due to the longer duration of oxidization process with a mixture of acid. The amorphous region of the cotton fiber was decreased whenever an oxidation process was performed at highly acidic conditions. Thus, the penetration of

acid into the amorphous region, as well as etherification, could be hindered, thus resulting in the deterioration of tensile properties.

Water retention ability of the produced dressing

Hydrophilicity is one of the most important properties for blood clotting activity. Hemostatic materials must be highly absorbent, since blood contains a large amount of water (Cheng et al., 2018; Li et al., 2016). The water retention ratio of the uncoated samples and the coated sample with PVA nanofibers can be seen in Fig. 6. As illustrated, water retention of the cotton gauze was improved with the oxidization process; the oxidization duration was also increased. This improvement was caused by the increase of carboxyl groups. Test results related to the carboxyl groups content also confirmed it. Moreover, it was found that the water retention ratio of the oxidized cotton gauze samples was enhanced by increasing the hydroxyl groups of the dressing and applying a hydrophilic coating, i.e., PVA nanofibers, to the surface.

Antibacterial activity

As expected, antibacterial test results exhibited no sign of antibacterial activity for the coated OCF sample with PVA nanofibers. Clearly, neither the oxidized cotton gauze nor the PVA featured antibacterial properties. This inactivity could arise from the absence of halo around the coated OCF sample with the PVA nanofibers. Figure 7 shows the inhibition of halo around the samples against two types of gram-negative (*E. coli*) and gram-positive (*S. aureus*) bacteria. The addition of Ciprofloxacin as an antibiotic to treat wound infections imparted the antibacterial activity to the PVA nanofibers. Table 3 presents the diameter (mm) of the inhibition zone around the samples. The antibacterial activity of the coated OCF sample with drug-loaded PVA nanofibers against both gram-positive and gram-negative bacteria could be observed. Consequently, the prepared dressing could be used as a wound dressing in traffic accidents or wherever there is a high risk of infection.

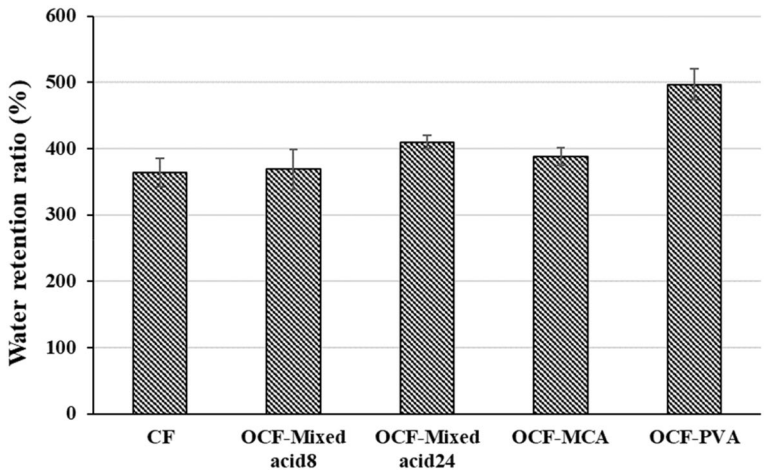

Fig. 6 Water retention ratio of cotton gauze (CF), oxidized cotton gauze with monocloro acetic acid (OCF-MCA) and mixture of nitric acid and phosphoric acid for 8 (OCF-Mixed acid8) and 24 (OCF-Mixed acid24) hours, and oxidized cotton gauze coated by PVA nanofibers (OCF-PVA)

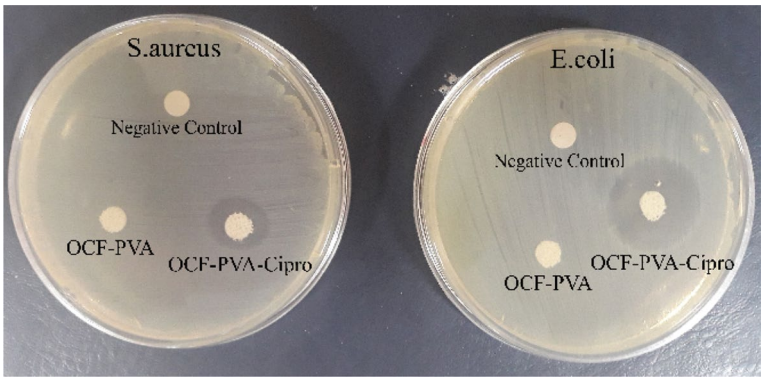

Fig. 7 Inhibition halo surrounding for oxidized cotton gauze coated with PVA nanofibers (OCF-PVA) and Ciprofloxacin loaded PVA nanofibers (OCF-PVA-Cipro) against E. coli and S. aureus

Table 3 Inhibition zone (mm) of oxidized cotton gauze coated with PVA nanofibers (OCF-PVA) and Ciprofloxacin loaded PVA nanofibers (OCF-PVA-Cipro) against *E. coli* and *S. aureus*

Diameter of inhibition zone (mm)	Sample	Bacteria
0	Negative Control	*E. coli*
0	OCF-PVA	
23.5	OCF-PVA-Cipro	
0	Negative Control	*S. aureus*
0	OCF-PVA	
14	OCF-PVA-Cipro	

In vitro hemostatic activities

Hemostatic activity of the oxidized cotton gauze samples was assessed in regard to the effects of oxidization methods and duration of the oxidization process. To this end, 100 µL of CB was placed on the samples with the dimensions of 1×1 cm^2 and its diffusivity on the samples was evaluated. According to Fig. 8 and observations, diffusivity of blood was higher for the samples OCF-MCA, OCF-Mixed acid1, and OCF-Mixed acid8, as compared to the sample OCF-Mixed acid24. Comparing the color shades of bloodstain showed that the sample "OCF-Mixed acid24" exhibited a darker shade, as compared to the others. Thus, it can be inferred that the sample "OCF-Mixed acid24" outperformed the others in terms of blood coagulation.

Blood clotting indices (i.e., BCI) can be used as a general index for the purpose of evaluating the hemostatic potential of materials under in vitro conditions (Cheng et al., 2019). Accordingly, BCI was used herein to quantify the observations (i.e., hemostatic activities). A low BCI indicates the better blood clotting ability of the sample (Zheng et al., 2021). As shown in Fig. 9, the oxidized cotton gauze with a mixture of two acids for 24 h (OCF-Mixed acid24) exhibited a lower BCI in comparison to the other samples. This was due to the higher content of carboxyl groups in the OCF-Mixed acid24 samples. As could be observed in the SEM images of the oxidized cotton gauze (Fig. 1), the oxidation process roughened the surface of the cotton gauze. Therefore, the uneven surface of the "OCF-Mixed acid24" sample contributed to the sufficient fabric-to-blood contact and the capability of absorbing water, thus allowing it to absorb blood

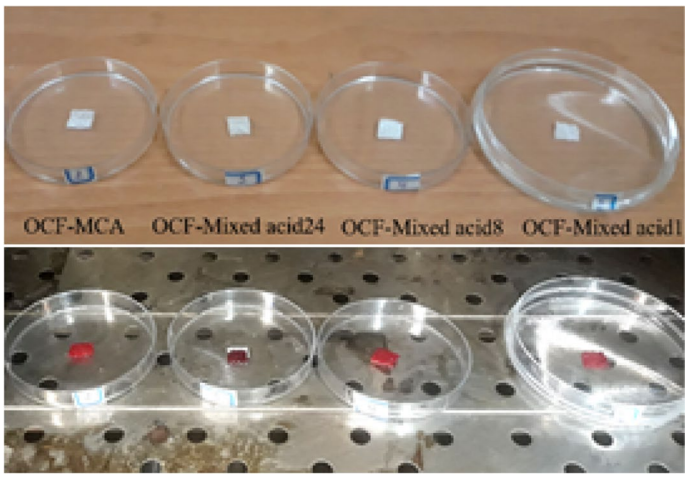

Fig. 8 Optical images of blood coagulation in vitro of the cotton gauze oxidized with momocholoro acetic acid (OCF-MCA), and mixture of nitric acid and phosphoric acid for 1 (OCF-Mixed acid1) 8 (OCF-Mixed acid8), and 24 (OCF-Mixed acid24) hours

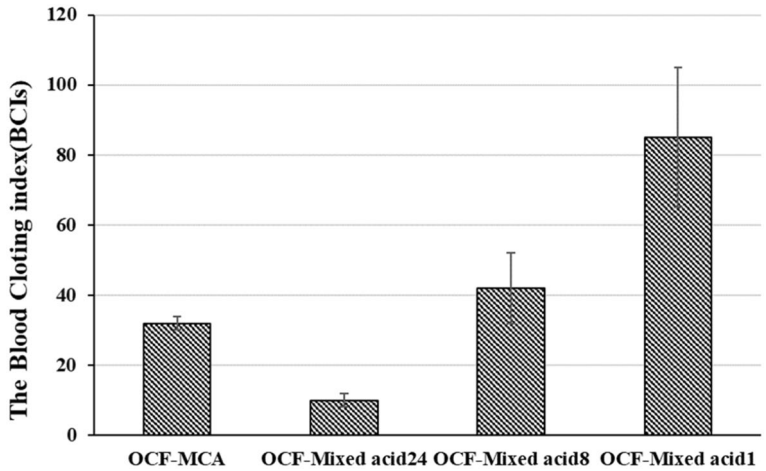

Fig. 9 Blood clotting indexes of the cotton gauze oxidized with momocholoro acetic acid (OCF-MCA), and mixture of nitric acid and phosphoric acid for 1 (OCF-Mixed acid1) 8 (OCF-Mixed acid8), and 24 (OCF-Mixed acid24) hours

and improve blood clotting. In addition, the prepared "OCF-Mixed acid24" sample can serve as a cloth, acting a physical barrier and blocking the blood pressure. Consequently, "OCF-Mixed acid24" sample can be used as a suitable base material for blood clotting in hemostatic dressings.

Conclusions

A cotton gauze was used in this study to produce a hemostatic dressing with antibacterial properties. Accordingly, two methods were employed to oxidize cotton gauze. The first method involved a mixture of nitric acid and phosphoric acid for different oxidation times, while the second one included monochloroacetic acid. SEM images showed the roughened surface of the cotton gauze after the oxidization process. The carboxyl groups

content of oxidized cotton gauzes using the first method (17.3 ± 0.3) was substantially higher, as compared to those oxidized using the second method (8.3 ± 1.1). It was also found that the carboxyl groups content was enhanced through increasing the duration of oxidation process. Based on the tensile test results, oxidation process had a negative impact on the tensile strength of cotton gauze. After the oxidization process, the oxidized cotton gauze with the highest content of carboxyl groups was further coated with PVA nanofibers (diameter of 196 nm) or PVA nanofibers loaded with Ciprofloxacin (diameter of 261 nm). Thus, according to the test results, a dressing featuring antibacterial activity against *E. coli* and *S. aureus* was obtained. Moreover, the findings indicated the improved water retention ratio of the coated samples in comparison to the uncoated ones. Besides this, an acceptable blood clotting activity with the BCI of 10 was observed for the oxidized cotton gauzes by using the first method (mixture of nitric acid and phosphoric acid) for 24 h. Overall, the developed hemostatic dressing exhibited promising features for use in traffic accidents.

Acknowledgements
Not applicable.

Author contributions
HM carried out the experiments, developed the ideas, interpretation of the results, and drafted the manuscript of the analysis. MG and AB supervised the project. All authors read and approved the final manuscript.

Authors' information
Mostafa Goodarz: Associate research professor at Amirkabir University of Technology, Iran.
Amir Behzadnia: Researcher at Islamic Azad University (Science and Research Branch), Iran.
Hassan Mohammadi: Sessional Lecturer at Art university of Isfahan, Iran.

Funding
Not applicable.

Availability of data and materials
Not applicable.

Declarations

Competing interests
The authors declare that they have no competing interests.

Received: 13 March 2022 Accepted: 2 June 2022
Published online: 25 September 2022

References

Asadi, N., Del Bakhshayesh, A. R., Davaran, S., & Akbarzadeh, A. (2020). Common biocompatible polymeric materials for tissue engineering and regenerative medicine. *Materials Chemistry and Physics, 242*, 122528. https://doi.org/10.1016/j.matchemphys.2019.122528.

Cao, X.-L., Zhang, Q.-H., Pan, X.-H., Chen, Z., & Lü, J. (2019). Mechanochemical synthesis of nano–ciprofloxacin with enhanced antibacterial activity. *Inorganic Chemistry Communications, 102*, 66–69. https://doi.org/10.1016/j.inoche.2019.02.015.

Chen, J., Cheng, W., Chen, S., Xu, W., Lin, J., Liu, H., & Chen, Q. (2018). Urushiol-functionalized mesoporous silica nanoparticles and their self-assembly into a Janus membrane as a highly efficient hemostatic material. *Nanoscale, 10*(48), 22818–22829. https://doi.org/10.1039/C8NR05882B.

Cheng, F., Liu, C., Li, H., Wei, X., Yan, T., Wang, Y., & Huang, Y. (2018). Carbon nanotube-modified oxidized regenerated cellulose gauzes for hemostatic applications. *Carbohydrate Polymers, 183*, 246–253. https://doi.org/10.1016/j.carbpol.2017.12.035.

Cheng, F., Wu, Y., Li, H., Yan, T., Wei, X., Wu, G., & Huang, Y. (2019). Biodegradable N, O-carboxymethyl chitosan/oxidized regenerated cellulose composite gauze as a barrier for preventing postoperative adhesion. *Carbohydrate Polymers, 207*, 180–190. https://doi.org/10.1016/j.carbpol.2018.10.077.

Cormier, R., Burda, W. N., Harrington, L., Edlinger, J., Kodigepalli, K. M., Thomas, J., & Turos, E. (2012). Studies on the antimicrobial properties of N-acylated ciprofloxacins. *Bioorganic & Medicinal Chemistry Letters, 22*(20), 6513–6520. https://doi.org/10.1016/j.bmcl.2012.05.026.

Dai, H. L., Tan, M. Y., & Guo, L. M. (2013). Study on absorbency and strength of the carboxymethyl medical cotton gauze. *Advanced Materials Research, 821*, 502–510. https://doi.org/10.4028/www.scientific.net/AMR.821-822.502.
Edwards, J. V., Prevost, N., Yager, D., Nam, S., Graves, E., Santiago, M., & Dacorta, J. (2021). Antimicrobial and hemostatic activities of cotton-based dressings designed to address prolonged field care applications. *Military Medicine, 186*(1), 116–121. https://doi.org/10.1093/milmed/usaa271.
Edwards, J. V., Yager, D. R., Cohen, I. K., Diegelmann, R. F., Montante, S., Bertoniere, N., & Bopp, A. F. (2001). Modified cotton gauze dressings that selectively absorb neutrophil elastase activity in solution. *Wound Repair and Regeneration, 9*(1), 50–58. https://doi.org/10.1046/j.1524-475x.2001.00050.x.
Mondal, M. I. H. (Ed.). (2015). *Cellulose and Cellulose Derivatives: Synthesis, Modification, and Applications*. Nova Publishers.
Garg, K., & Bowlin, G. L. (2011). Electrospinning jets and nanofibrous structures. *Biomicrofluidics, 5*(1), 013403. https://doi.org/10.1063/1.3567097.
Islam, M. S., Ang, B. C., Andriyana, A., & Afifi, A. M. (2019). A review on fabrication of nanofibers via electrospinning and their applications. *SN Applied Sciences, 1*(10), 1–16. https://doi.org/10.1007/s42452-019-1288-4.
Lemraski, E. G., Jahangirian, H., Dashti, M., Khajehali, E., Sharafinia, S., Rafiee-Moghaddam, R., & Webster, T. J. (2021). Antimicrobial double-layer wound dressing based on chitosan/polyvinyl alcohol/copper: In vitro and in vivo assessment. *International Journal of Nanomedicine, 16*, 223. https://doi.org/10.2147/IJN.S266692.
Leonhardt, E. E., Kang, N., Hamad, M. A., Wooley, K. L., & Elsabahy, M. (2019). Absorbable hemostatic hydrogels comprising composites of sacrificial templates and honeycomb-like nanofibrous mats of chitosan. *Nature Communications, 10*(1), 1–9. https://doi.org/10.1038/s41467-019-10290-1.
Li, J., Celiz, A., Yang, J., Yang, Q., Wamala, I., Whyte, W., & Suo, Z. (2017). Tough adhesives for diverse wet surfaces. *Science, 357*(6349), 378–381. https://doi.org/10.1126/science.aah6362.
Li, Q., Lu, F., Shang, S., Ye, H., Yu, K., Lu, B., & Lan, G. (2019). Biodegradable microporous starch with assembled thrombin for rapid induction of hemostasis. *ACS Sustainable Chemistry & Engineering, 7*(10), 9121–9132. https://doi.org/10.1021/acssuschemeng.8b05701.
Li, T.-T., Lou, C.-W., Chen, A.-P., Lee, M.-C., Ho, T.-F., Chen, Y.-S., & Lin, J.-H. (2016). Highly absorbent antibacterial hemostatic dressing for healing severe hemorrhagic wounds. *Materials, 9*(9), 793. https://doi.org/10.3390/ma9090793.
Li, Y., Zhu, J., Cheng, H., Li, G., Cho, H., Jiang, M., & Zhang, X. (2021). Developments of advanced electrospinning techniques: A critical review. *Advanced Materials Technologies, 34*, 2100410. https://doi.org/10.1002/admt.202100410.
Liang, D., Lu, Z., Yang, H., Gao, J., & Chen, R. (2016). Novel asymmetric wettable AgNPs/chitosan wound dressing: In vitro and in vivo evaluation. *ACS Applied Materials & Interfaces, 8*(6), 3958–3968. https://doi.org/10.1021/acsami.5b11160.
Memic, A., Abudula, T., Mohammed, H. S., Joshi Navare, K., Colombani, T., & Bencherif, S. A. (2019). Latest progress in electrospun nanofibers for wound healing applications. *ACS Applied Bio Materials, 2*(3), 952–969. https://doi.org/10.1021/acsabm.8b00637.
Rafieian, S., Mahdavi, H., & Masoumi, M. E. (2021). Improved mechanical, physical and biological properties of chitosan films using Aloe vera and electrospun PVA nanofibers for wound dressing applications. *Journal of Industrial Textiles, 50*(9), 1456–1474. https://doi.org/10.1177/1528083719866932.
Sahoo, S., Chakraborti, C., Naik, S., Mishra, S., & Nanda, U. (2011). Structural analysis of ciprofloxacin-carbopol polymeric composites by X-Ray diffraction and Fourier transform infra-red spectroscopy. *Tropical Journal of Pharmaceutical Research, 10*(3), 8. https://doi.org/10.4314/tjpr.v10i3.14.
Shefa, A. A., Taz, M., Hossain, M., Kim, Y. S., Lee, S. Y., & Lee, B. T. (2019). Investigation of efficiency of a novel, zinc oxide loaded TEMPO-oxidized cellulose nanofiber based hemostat for topical bleeding. *International Journal of Biological Macromolecules, 126*, 786–795. https://doi.org/10.1016/j.ijbiomac.2018.12.079.
Shin, M., Park, S.-G., Oh, B.-C., Kim, K., Jo, S., Lee, M. S., & Kim, K.-S. (2017). Complete prevention of blood loss with self-sealing haemostatic needles. *Nature Materials, 16*(1), 147–152. https://doi.org/10.1038/nmat4758.
Thairin, T., & Wutticharoenmongkol, P. (2021). Ciprofloxacin-loaded alginate/poly (vinyl alcohol)/gelatin electrospun nanofiber mats as antibacterial wound dressings. *Journal of Industrial Textiles, 3*, 1528083721997466. https://doi.org/10.1177/1528083721997466.
Uhljar, L. É., Kan, S. Y., Radacsi, N., Koutsos, V., Szabó-Révész, P., & Ambrus, R. (2021). In vitro drug release, permeability, and structural test of ciprofloxacin-loaded nanofibers. *Pharmaceutics, 13*(4), 556. https://doi.org/10.3390/pharmaceutics13040556.
USP (United States Pharmacopeia 23/National Formulary 18). (1995). Oxidized cellulose (p. 318).
Wang, Y., Xiao, D., Zhong, Y., Liu, Y., Zhang, L., Chen, Z., Sui, X., Wang, B., Feng, X., Xu, H., & Mao, Z. (2020b). Preparation and characterization of carboxymethylated cotton fabrics as hemostatic wound dressing. *International Journal of Biological Macromolecules, 160*, 18–25. https://doi.org/10.1016/j.ijbiomac.2020.05.099.
Wang, Y., Xiao, D., Zhong, Y., Zhang, L., Chen, Z., Sui, X., & Mao, Z. (2020a). Facile fabrication of carboxymethyl chitosan/paraffin coated carboxymethylated cotton fabric with asymmetric wettability for hemostatic wound dressing. *Cellulose, 27*(6), 3443–3453. https://doi.org/10.1007/s10570-020-02969-2.
Wang, Y., Zhou, P., Xiao, D., Zhu, Y., Zhong, Y., Zhang, J., & Mao, Z. (2019). Chitosan-bound carboxymethylated cotton fabric and its application as wound dressing. *Carbohydrate Polymers, 221*, 202–208. https://doi.org/10.1016/j.carbpol.2019.05.082.
Xia, Q., Liu, Z., Wang, C., Zhang, Z., Xu, S., & Han, C. C. (2015). A biodegradable trilayered barrier membrane composed of sponge and electrospun layers: Hemostasis and antiadhesion. *Biomacromolecules, 16*(9), 3083–3092. https://doi.org/10.1021/acs.biomac.5b01099.
Xu, Y., Liu, X., Liu, X., Tan, J., & Zhu, H. (2014). Influence of HNO3/H3PO4–NaNO2 mediated oxidation on the structure and properties of cellulose fibers. *Carbohydrate Polymers, 111*, 955–963. https://doi.org/10.1016/j.carbpol.2014.05.029.
Xu, Y., Qiu, C., Zhang, X., & Zhang, W. (2014). Crosslinking chitosan into H3PO4/HNO3–NANO2 oxidized cellulose fabrics as antibacterial-finished material. *Carbohydrate Polymers, 112*, 186–194. https://doi.org/10.1016/j.carbpol.2014.05.054.

Yang, X., Liu, W., Shi, Y., Xi, G., Wang, M., Liang, B., & Shi, C. (2019). Peptide-immobilized starch/PEG sponge with rapid shape recovery and dual-function for both uncontrolled and noncompressible hemorrhage. *Acta Biomaterialia, 99*, 220–235. https://doi.org/10.1016/j.actbio.2019.08.039.
Yu, L., Shang, X., Chen, H., Xiao, L., Zhu, Y., & Fan, J. (2019). A tightly-bonded and flexible mesoporous zeolite-cotton hybrid hemostat. *Nature Communications, 10*(1), 1–9. https://doi.org/10.1038/s41467-019-09849-9.
Yu, P., & Zhong, W. (2021). Hemostatic materials in wound care. *Burns & Trauma, 9*. https://doi.org/10.1093/burnst/tkab019
Yuk, H., Varela, C. E., Nabzdyk, C. S., Mao, X., Padera, R. F., Roche, E. T., & Zhao, X. (2019). Dry double-sided tape for adhesion of wet tissues and devices. *Nature, 575*(7781), 169–174. https://doi.org/10.1038/s41586-019-1710-5.
Zhang, S., Li, J., Chen, S., Zhang, X., Ma, J., & He, J. (2020). Oxidized cellulose-based hemostatic materials. *Carbohydrate Polymers, 230*, 115585. https://doi.org/10.1016/j.carbpol.2019.115585.
Zhao, X., Guo, B., Wu, H., Liang, Y., & Ma, P. X. (2018). Injectable antibacterial conductive nanocomposite cryogels with rapid shape recovery for noncompressible hemorrhage and wound healing. *Nature Communications, 9*(1), 1–17. https://doi.org/10.1038/s41467-018-04998-9.
Zhao, Y., Hao, J., Chen, Z., Li, M., Ren, J., & Fu, X. (2021). Blood-clotting model and simulation analysis of polyvinyl alcohol–chitosan composite hemostatic materials. *Journal of Materials Chemistry b., 9*(27), 5465–5475. https://doi.org/10.1039/D1TB00159K.
Zheng, W., Chen, C., Zhang, X., Wen, X., Xiao, Y., Li, L., & Liu, X. (2021). Layer-by-layer coating of carboxymethyl chitosan-gelatin-alginate on cotton gauze for hemostasis and wound healing. *Surface and Coatings Technology, 406*, 126644. https://doi.org/10.1016/j.surfcoat.2020.126644.
Zhu, L., Zhang, S., Zhang, H., Dong, L., Cong, Y., Sun, S., & Sun, X. (2021). Polysaccharides composite materials for rapid hemostasis. *Journal of Drug Delivery Science and Technology, 66*, 102890. https://doi.org/10.1016/j.jddst.2021.102890.
Zhu, T., Wu, J., Zhao, N., Cai, C., Qian, Z., Si, F., & Shao, L. (2018). Superhydrophobic/superhydrophilic janus fabrics reducing blood loss. *Advanced Healthcare Materials, 7*(7), 1701086. https://doi.org/10.1002/adhm.201701086.

Publisher's Note