

Research Article

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Modification of *Ceiba pentandra* cellulose for drug release applications

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Abstract: Tubular fibers (raw and wax-free) from *Ceiba pentandra* (CP) were cross-linked with butane-1,2,3,4-tetra-carboxylic acid (BTCA) at different concentrations to obtain a porous biodegradable medium for drug release applications. Chlorhexidine diacetate (CHX) was added to the cross-linked fibers for drug release studies. The Fourier transform infrared spectroscopy, thermogravimetric analysis, and scanning electron microscopy results indicated that the cross-linked fibers with a 5:1 fiber:BTCA ratio presented the higher cross-linking density. CHX was added at different concentrations (8% and 16% wt/wt); the elemental analysis indicated that CHX was loaded up to 7.99 wt%. *In vitro* studies showed a burst release of CHX within the first 3 h. CHX release kinetics was described using several models, with the Korsmeyer–Peppas equation, which adjusted better to the experimental data. The results indicated that the CP fibers are a feasible material for drug release applications.

Keywords: *Ceiba pentandra*, chlorhexidine diacetate, cellulose cross-linking, drug release

1 Introduction

The *Ceiba pentandra* (CP) is a widely distributed tree in Central America, South America, Southeast Asia,

and West Africa. The fruits contain seeds covered with long and woolly fibers, which are produced in large quantities (1). The main components of CP fibers are cellulose (35–44%) and lignin (13–35%), and other components are polysaccharides, wax, and oil (2,3).

The fibers (kapok) have a hollow tubular structure with a diameter of $14.5 \pm 2.4 \mu\text{m}$ (4), and their length ranges from 0.8 to 3 cm (1). Due to this morphological characteristic, CP fibers have been used in several application such as absorption of oils for pollution control (3,5,6), absorption of heavy metals (7), preparation of water- and moisture-resistant paper (8), and sound and thermal insulation (9,10) and used as textile materials (11). The fiber has been used in the preparation of activated carbon (12) and bioethanol (13) and as templates in the preparation of polymeric microtubes (14).

On the other hand, research in the drug release area involves the development of biodegradable, biocompatible, non-toxic materials, with different geometries that improve the transport of the drug to the site of action. The matrices for the release of drugs involve gels, microspheres, films, among others. In addition, there are studies reporting the use of microfibers and nanofibers from polymeric matrices exhibiting a morphology and structure similar to those presented by the CP fibers (15,16). Because the kapok fiber is a biocompatible and biodegradable material with a long and hollow tubular structure in micrometric dimensions, it could be employed for the encapsulation and release of drugs and designing of materials for the production of tissue scaffolding.

This study explores the possibility of using the cellulose tubular structure of CP to develop drug delivery systems using chlorhexidine diacetate (CHX) as a drug model because it is easily available, has thermal and chemical stability, and is easily detected by UV-vis spectrophotometry. Also, the effect of hydrophobic modification of CP fibers on drug release was studied using the raw fibers and pretreated fibers (free of wax and natural oils). In addition, the chemical cross-linking was performed to improve the physical properties of the fibers and improve their drug retention.

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To our knowledge, CP fibers have not been used for drug release applications so far.

2 Experimental

2.1 Materials

Kapok fruits were collected from Merida, Mexico, and their fibers were manually removed from the shells and seeds. Benzene, ethanol, sodium hypophosphite (SHP), butane-1,2,3,4-tetracarboxylic acid (BTCA), chlorhexidine acetate (CA), and phosphate-buffered saline (PBS) media were supplied by Sigma Aldrich Co. and were used as received.

2.2 Pretreatment

Kapok fibers are highly hydrophobic due to the presence of natural oils and waxes. The oily components and other impurities were removed by the TAPPI standard method T 204 (17) using a Soxhlet extractor with ethanol–benzene (1:2) for 4 h. Then, the extracted fibers were washed with deionized water and dried at 105°C.

2.3 Cellulose cross-linking

Five grams of the fibers (raw and pretreated) was dispersed for 5 min in an aqueous solution containing BTCA and SHP (50 wt% to BTCA) at different fiber:BTCA ratios (Table 1). The fibers were ground with a blender and dried in an oven at 102°C until constant weight was achieved. After drying, the fibers were cross-linked in an oven at 180°C for 5 min and then cooled to room temperature. Finally, the samples were washed with deionized water to remove the unreacted chemicals and

Table 1: Nomenclature and concentrations of the cross-linking agent and catalyst used in this research

Raw fiber	Pretreated fiber	Fiber:BTCA ratio
RF–BTCA-5:1		5:1 (5 g/1 g)
RF–BTCA-20:1		20:1 (5 g/0.25 g)
	PF–BTCA-5:1	5:1 (5 g/1 g)
	PF–BTCA-20:1	20:1 (5 g/0.25 g)

then dried in a vacuum oven at 65°C until constant weight was achieved.

2.4 Chlorhexidine loading

Five hundred milligrams of the cross-linked fibers were immersed in 100 mL of distilled water using a magnetic stirrer for 30 min. CHX (8 and 16 wt% to fiber mass) was dissolved in 15 mL of distilled water using an ultrasonic bath for 30 min at room temperature and then mixed with the immersed fibers using a magnetic stirrer at room temperature for 24 h (the pH was adjusted to 4.1 with H₃PO₄). Finally, the fibers were filtered, washed with deionized water, and dried in a vacuum oven at 60°C until constant weight was achieved (18,19).

2.5 Drug release studies

In vitro studies on CHX release from the kapok fibers were carried out by placing 30 mg of fibers in 40 mL of PBS media (pH = 7.4) and slow magnetic stirring for 24 h at 37°C. Five milliliters of the dissolution medium was withdrawn at regular time intervals and replaced with a fresh solution medium to maintain sink conditions. The withdrawn aliquots were centrifuged, and the CA content in the supernatants was determined by UV spectrophotometer ($\lambda = 264$ nm) (18,20,21). The tests were performed in duplicate for each fiber samples.

2.6 Characterization

2.6.1 Scanning electron microscopy

Scanning electron microscopy (SEM) analysis was performed using an SEM JEOL JSM 6360LV with an acceleration voltage of 20 kV; the samples were coated with gold.

2.6.2 Fourier transform infrared spectroscopy

Fourier transform infrared spectroscopy (FTIR) analyses were conducted with a Thermo Scientific Nicolet 8700 equipment; the attenuated total reflectance technique was employed; the scanning range was 4,000–600 cm⁻¹

wavenumber with a resolution of 4 cm^{-1} ; the spectra were obtained averaging 50 scans and normalized against the highest intensity peak.

2.6.3 X-ray photoelectron spectrometry

The X-ray photoelectron spectrometry (XPS) analysis was performed with a Thermo Scientific K Alpha Surface Analysis equipment, at 40 W and 12 kV. The signal deconvolution was carried out by AAnalyzer® software and the Gaussian–Lorentzian method.

2.6.4 Elemental analysis

The elemental analysis of the CP fibers was carried out with a Thermo Scientific Flash 2000 series CHNS-O, organic elemental analyzer.

2.6.5 Thermogravimetric analysis

The thermogravimetric analysis (TGA) data were collected from 50 to 650°C at a rate of 10°C/min under nitrogen atmosphere by using a Perkin Elmer TGA 8000. The sample weight was 5 mg.

3 Results and discussions

3.1 Scanning electron microscopy

SEM micrographs of the cross-linked fibers (raw and pretreated) are shown in Figure 1. The fiber shows a cylindrical shape, with a hollow structure and smooth thin walls. The typical diameter is $\sim 18\ \mu\text{m}$ and the length was over a few centimeters. The fiber structure was preserved even after the solvent pretreatment and cross-linking process. Some fibers appear to be open or torn (marked in circles) as a result of the grinding process; however, the broken fibers were fewer than the intact fibers.

3.2 Fourier transform infrared spectroscopy

After the ethanol–benzene treatment, the average extractives content of the fibers was determined to be 2 wt% (five measurements were performed). This result is comparable to that reported by Hori *et al.* (3), who determined the extractives content as 2.2% and 1.8% using the same methodology; on the other hand, Draman *et al.* (22) and Abdullah *et al.* (23) reported

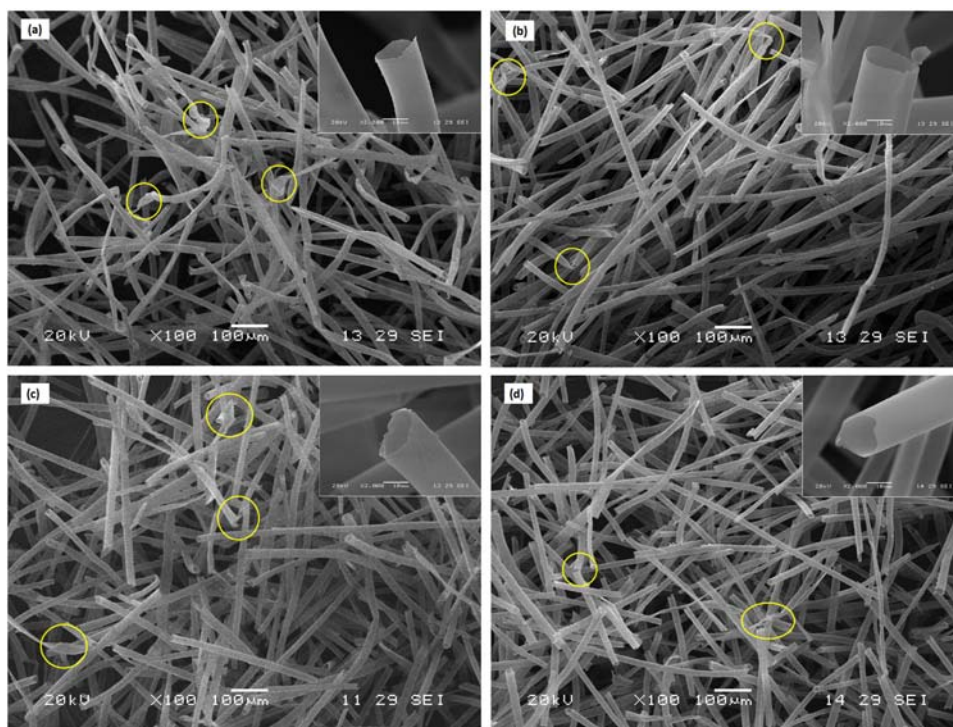


Figure 1: SEM micrographs of kapok fibers crosslinked with BTCA at different ratios: (a) raw fiber 20:1; (b) pretreated fiber 20:1; (c) raw fiber 5:1 and (d) pretreated fiber 5:1.

5.3% and 3% of wax content using chloroform. The same authors stated that the wax consists of fatty acids, long-chain alkenes, aldehydes, ketones, and small portions of alcohols and esters.

Figure 2 shows the FTIR spectra of the raw and pretreated kapok fibers. The intensity of the band at $3,340\text{ cm}^{-1}$, corresponding to -OH groups of cellulose, is higher for pretreated fibers (spectra were normalized to an absorbance of 1 at $1,037\text{ cm}^{-1}$ wavenumber); -OH groups could be more exposed due to the removal of wax and oil from the fiber surface. The bands around $2,980$ and $2,830\text{ cm}^{-1}$ are related to the asymmetric and symmetric stretching vibrations of the C-H bonds of cellulose (24,25); the band at $1,732\text{ cm}^{-1}$ corresponds to the ester bonds of the carbonyl groups in the hemicellulose and lignin (26,27). The peaks at $1,505$ and $1,594\text{ cm}^{-1}$ correspond to C=C aromatic skeletal vibration of lignin. The band at $1,425\text{ cm}^{-1}$ is attributed to the bending vibrations of C-H from cellulose and lignin. The peaks at $1,370$, $1,161$, and $1,035\text{ cm}^{-1}$ correspond to cellulose, hemicellulose, and lignin, respectively; the first one for C-H bending vibration, the second one for C-O-C asymmetric stretching, and the third one for stretching of the C-O bonded to the aromatic ring of lignin (22,28,29). Finally, the band at 899 cm^{-1} corresponds to the presence of β -glycosidic linkage between glucose units in cellulose (30).

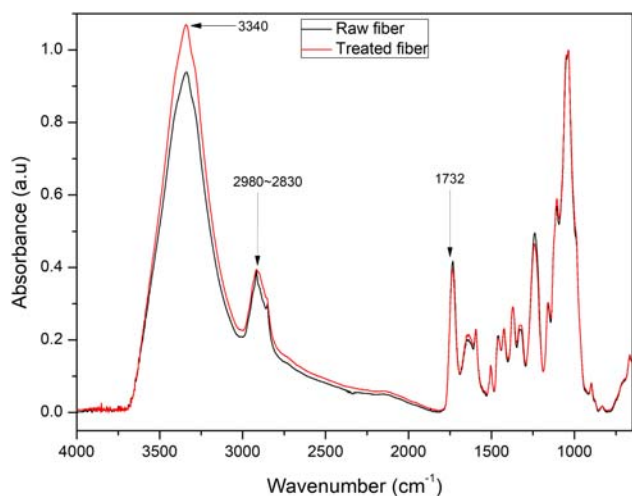


Figure 2: FTIR spectra of raw and pretreated fibers.

During cross-linking process (Figure 3), ester bonds were formed among cellulose -OH groups and carboxylic groups from the cross-linking agent; as a result, there is a decrease in hydroxyl-to-ester ratio ($\text{Abs } 3,340\text{ cm}^{-1} / (\text{Abs } 1,732\text{ cm}^{-1})$) from 2.215 to 1.753 for raw fibers and from 2.388 to 1.786 for pretreated fibers, i.e., the hydroxyl bands tend to decrease and ester bands

tend to increase. In this sense, the fibers cross-linked with higher concentration of cross-linking agent (i.e., 5:1 fiber:BTCA ratio) exhibited -OH bands at $3,340\text{ cm}^{-1}$ with relative lower intensity than those using lower concentration of BTCA. Similarly, higher concentration of BTCA produced higher intensity bands at $1,732\text{ cm}^{-1}$, indicating a correlation between the concentration of cross-linking agent and the cross-linking degree.

3.3 X-ray photoelectron spectrometry

The surface elemental compositions of all kapok fibers are summarized in Table 2. The analysis of the survey spectra indicates that the fibers consist mainly of carbon (C1s) and oxygen (O1s). In general, oxygen content increased with the concentration of the cross-linking agent, mainly attributed to the introduction of O-containing groups. The pretreated sample exhibited higher oxygen content than the raw sample since the oils and waxes were removed.

A high-resolution scan of the XPS spectra of C1s showed a decomposition in four component peaks around 284.7, 286.1, 287.5, and 289.2 eV, resulting from C1 (carbon atoms bonded to carbon (C-C) and carbon atoms bonded to hydrogen atom (C-H)), C2 (carbon-oxygen bonds of alcohols and ethers (C-O)), C3 (carbon atoms bonded to two non-carbonyl oxygen (O-C-O) or to a single carbonyl oxygen atom (C=O)), and C4 (carbon atoms bonded to a carbonyl and a non-carbonyl oxygen (O-C=O)) (31,32).

In general, C2 and C3 carbon atoms decrease with the cross-linker content since OH groups were consumed during the esterification reaction. In this sense, the results indicate that the C4 carbon atom contribution increased with the cross-linker content regardless of the pretreatment process; the highest contribution was observed in the pretreated sample cross-linked with a 5:1 fiber:BTCA ratio, indicating that new ester bonds were formed. The FTIR results show that the sample with a higher degree of cross-linking is the pretreated fiber modified with a 5:1 fiber-BTCA ratio.

3.4 Elemental analysis

The elemental analysis results are presented in Table 3; it was noted that the nitrogen content increases in the pretreated fibers loaded with CHX. The low N values and the highly hydrophobic nature of the kapok fiber could

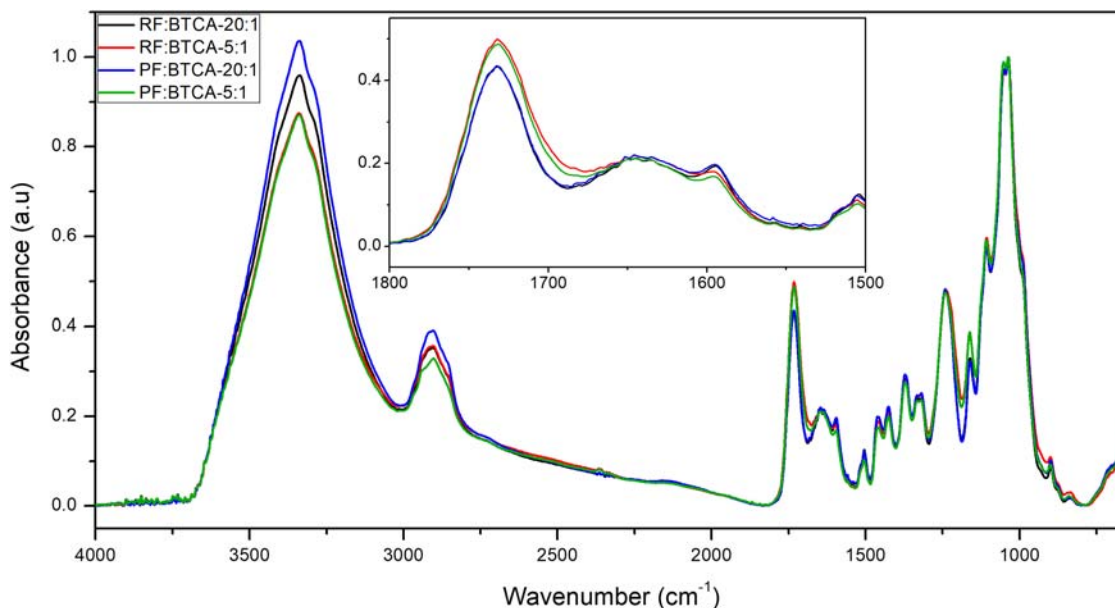


Figure 3: FTIR spectra for all fibers crosslinked at different BTCA ratios.

Table 2: Atomic percentages; O/C ratio; and C1, C2, C3, and C4 distribution of CP fibers

Sample	O%	C%	O/C	C1%	C2%	C3%	C4%
Raw fiber	7.43	95.57	0.08	69.06	17.69	9.14	4.11
RF-BTCA-20:1	12.21	87.79	0.14	63.34	21.52	10.56	4.58
RF-BTCA-5:1	15.53	84.47	0.18	68.40	18.38	8.40	4.81
Pretreated fiber	10.48	89.52	0.12	64.42	21.20	9.94	4.44
PF-BTCA-20:1	9.20	90.80	0.10	69.67	17.13	8.89	4.31
PF-BTCA-5:1	15.67	84.33	0.19	68.16	18.65	7.94	5.25

prevent the CHX diffusion into the hollow structure of the fiber. In the solvent pretreated fibers, the hydrophobicity of the fibers is reduced, allowing CHX loading more efficiently.

The nitrogen content was used to quantify the amount of drug loaded into the cross-linked fibers. The nitrogen content was calculated, considering that a sample of pure CHX, with

the chemical formula $C_{26}H_{38}Cl_2N_{10}O_4$ and a molecular weight of 625.5 g/mol, contains 27.70 wt% of nitrogen content. The amount of chlorhexidine in the final products ranged from 3.59 to 7.99 wt%. The concentration of chlorhexidine in the commercially available products ranges from 0.5% to 2.0% either in solution or topical forms; the results of this study suggest the use of kapok fibers as a feasible material for their future development in pharmaceuticals.

3.5 Thermogravimetric analysis

Figure 4a shows the TGA thermograms of the raw and pretreated fibers, revealing clearly three stages of weight loss: the first one from 50°C to 100°C, which is related to water from moisture, and the second one from 230°C to 360°C, which is ascribed to the thermal degradation of

Table 3: Elemental analysis of cross-linked fibers by CHNSO-analyzer and loading efficiency of CHX

Sample	Fiber:BTCA	CHX loaded (%)	Carbon (wt%)	Nitrogen (wt%)	Hydrogen (wt%)	Calculated CHX load (wt%)
Raw fiber	20:1	8	47.33 ± 0.12	1.17 ± 0.18	5.59 ± 0.03	4.24
		16	47.52 ± 0.13	1.00 ± 0.01	5.56 ± 0.02	3.59
	5:1	8	47.09 ± 0.01	1.99 ± 0.14	5.52 ± 0.03	7.20
		16	47.33 ± 0.13	1.82 ± 0.01	5.60 ± 0.07	6.58
Treated fiber	20:1	8	47.24 ± 0.06	2.21 ± 0.06	5.69 ± 0.07	7.99
		16	47.40 ± 0.31	2.04 ± 0.01	5.76 ± 0.01	7.36
	5:1	8	46.59 ± 0.03	2.07 ± 0.01	5.44 ± 0.07	7.49
		16	46.82 ± 0.08	2.20 ± 0.13	5.48 ± 0.03	7.96

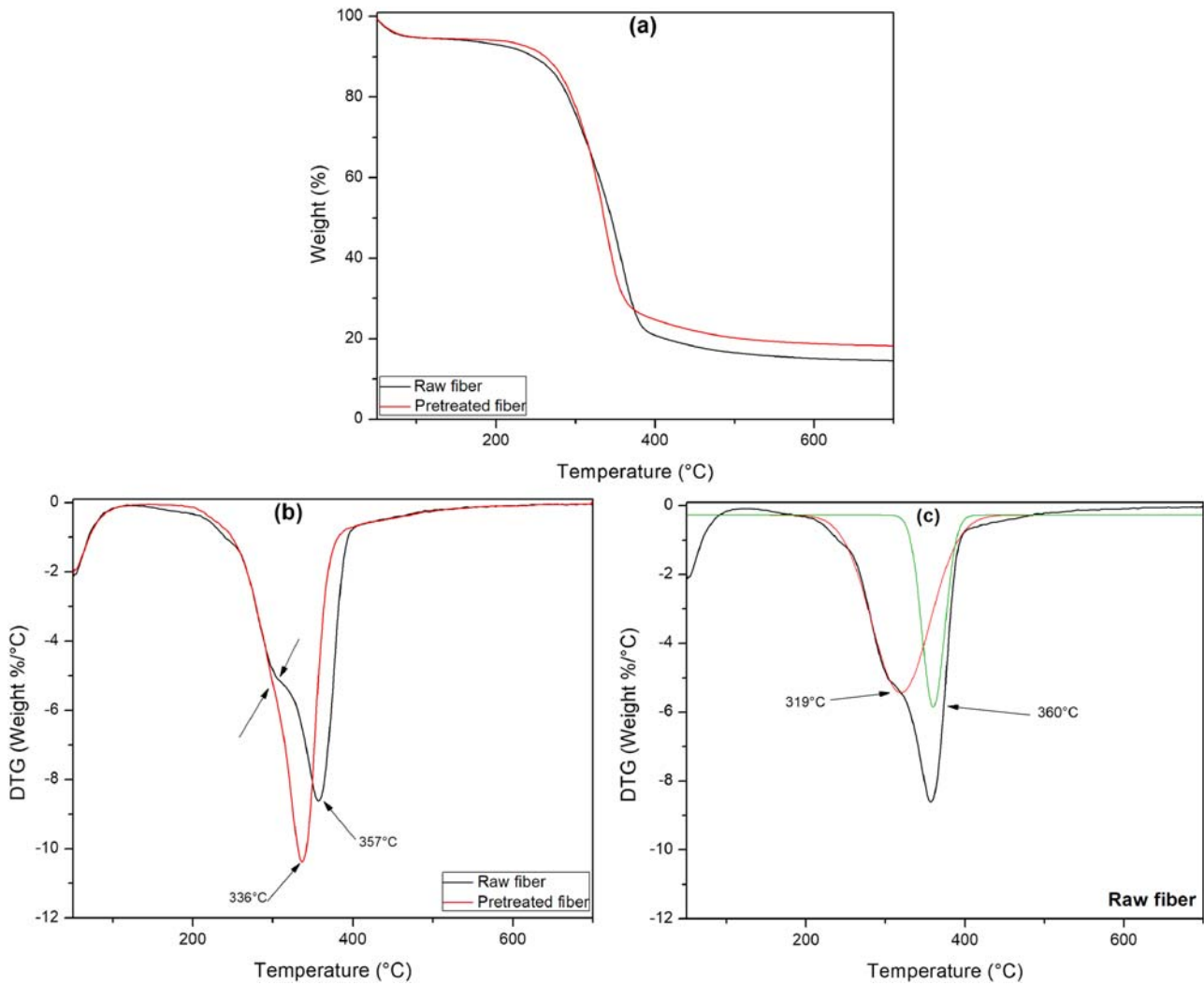


Figure 4: (a) TGA, (b) DTG of raw and pretreated fibers and (c) deconvolution of DTG curves of raw fiber.

cellulose and hemicellulose (22,33). The last weight loss is above 360°C related to lignin, which is reported to have a thermal degradation interval from 200°C to 900°C (28). At 700°C, the residual mass of the raw fiber is 14.62% and that of the pretreated fiber is 18.28%.

Figure 4b shows the DTG curves of the raw and pretreated fibers. It shows that the raw fibers not only exhibit a higher decomposition temperature than the pretreated fibers (357°C vs. 336°C, respectively) but also a shoulder around 290°C. These facts could be attributed to the elimination of oils and waxes during the solvent pretreatment, decreasing the thermal resistance (34). The deconvolution of the DTG curve (Figure 4c) reveals two overlapped decomposition peaks, corresponding to the degradation of the hemicellulose at 319°C and the degradation of the cellulose at 360°C.

Figure 5 shows the TGA thermograms of the cross-linked fibers with BTCA at different ratios. The temperature

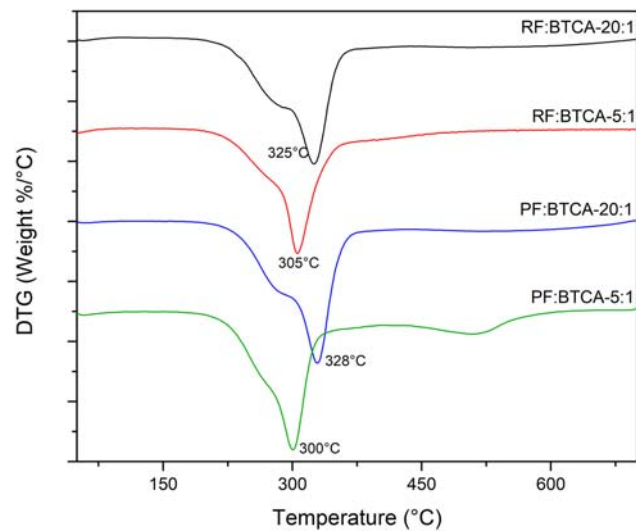


Figure 5: DTG curves of crosslinked fibers.

of maximum degradation peak decreases with the concentration of BTCA, suggesting that the cross-linking process reduces the thermal resistance. This agrees with the work reported by vom Stein *et al.* (35), who found that cellulose depolymerization can be catalyzed by carboxylic acids at temperatures above 160°C; also, it is possible that the cross-linking process reduces the hydrogen bonding of cellulose, diminishing the thermal stability of cellulose crystals.

3.6 Drug-release studies

The cumulative drug release from the cross-linked fibers is shown in Figure 6. In general, the

cross-linked fibers showed a burst release within the first hours; then, a slower release was observed. The cross-linked raw fibers showed a burst release in the first 3 h, then the drug release was slower in the next 21 h. On the other hand, the pretreated cross-linked fibers exhibited a burst release within the first 2 h, followed by a slower drug release process. In both cases, the initial burst release is associated with the CHX on the fiber surface, which can be easily released into the buffer solution, and the second behavior could be associated with the diffusion of CHX from the hollow fiber structure. The cross-linked fibers with a higher amount of BTCA showed higher CHX release; higher degree of cross-linking leads to a more porous material, increasing the probability for adsorbing more drug.

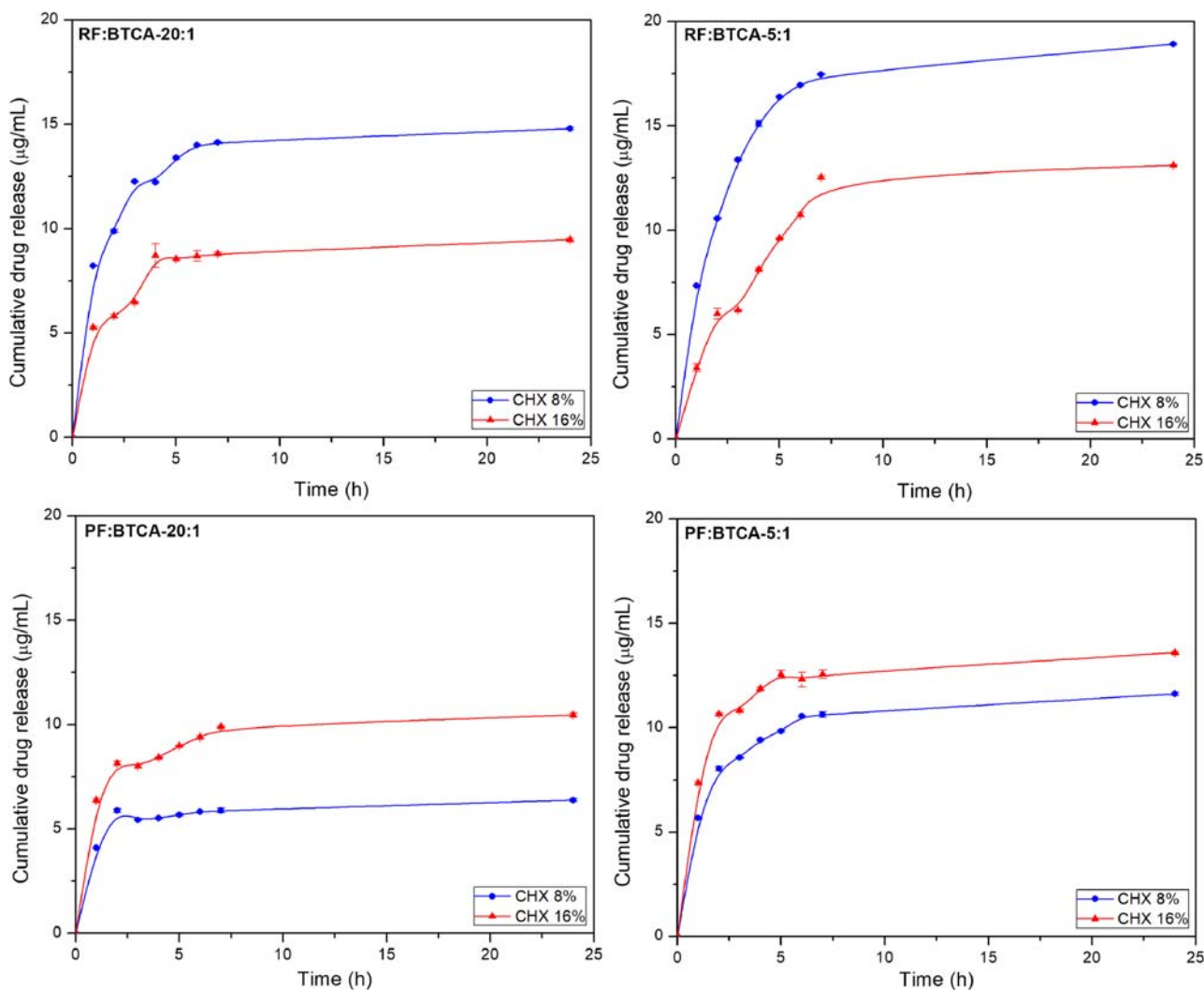


Figure 6: Cumulative release of chlorhexidine from *Ceiba pentandra*'s fibers. Raw fibers (top), pretreated fiber (bottom).

Table 4: R^2 and rate constant k for chlorhexidine release from cross-linked CP fibers

Sample	CHX (%)	Zero order	First order	Higuchi	Hixon–Crowell	Korsmeyer–Peppas
RF–BTCA-20:1	8	0.8540 5.08×10^{-5}	0.9677 1.49×10^{-4}	0.9667 7.19×10^{-3}	0.9292 3.22×10^{-5}	0.9972 8.13×10^{-2}
	16	0.9007 4.77×10^{-5}	0.9606 1.22×10^{-4}	0.9844 6.65×10^{-3}	0.9486 2.81×10^{-5}	0.9940 7.45×10^{-2}
RF–BTCA-5:1	8	0.9343 4.56×10^{-5}	0.9974 1.07×10^{-4}	0.9965 6.30×10^{-3}	0.9829 2.57×10^{-5}	0.9888 7.01×10^{-2}
	16	0.9809 4.03×10^{-5}	0.9229 9.12×10^{-5}	0.9911 5.44×10^{-3}	0.9786 2.19×10^{-5}	0.9536 5.97×10^{-2}
PF–BTCA-20:1	8	0.8270 4.94×10^{-5}	0.8654 1.26×10^{-4}	0.9508 7.04×10^{-3}	0.8549 2.93×10^{-5}	0.9895 7.99×10^{-2}
	16	0.8673 4.76×10^{-5}	0.9631 1.17×10^{-4}	0.9718 6.71×10^{-3}	0.9336 2.75×10^{-5}	0.9979 7.58×10^{-2}
PF–BTCA-5:1	8	0.8988 4.64×10^{-5}	0.9815 1.08×10^{-4}	0.9866 6.49×10^{-3}	0.9573 2.61×10^{-5}	0.9981 7.28×10^{-2}
	16	0.8724 4.86×10^{-5}	0.9531 1.23×10^{-4}	0.9753 6.85×10^{-3}	0.9274 2.87×10^{-5}	0.9968 7.72×10^{-2}

3.7 Release kinetics data analysis

Drug release kinetics was modeled using several models (Table 4), with the Korsmeyer–Peppas equation (36), with an n value of 0.25, which describes better the drug release kinetics. According to the model, the n value of 0.25 has been associated with the presence of pores in the polymeric matrix (37) and the high R^2 values indicate that the drug release depends on the diffusion of the drug from the porous materials obtained by the chemical cross-linking of the fibers. Table 4 summarizes the R^2 and rate constant k for chlorhexidine release from the cross-linked CP fibers. The rate constant k of the samples are comparable regardless of the amount of CHX loaded. The extracted fibers exhibit higher k values, suggesting that the hydrophobic nature of the raw fibers prevents the out diffusion of CHX to the buffer solution.

4 Conclusions

This work explores for the first time the possibility of using CP fibers for drug release applications. The kapok fibers (raw and oil-free) were successfully cross-linked using BTCA as a cross-linking agent according to the FTIR and XPS analyses; the better cross-linked systems were obtained using a 5:1 fiber–BTCA ratio. Morphology and crystallinity were not changed during the cross-linking process.

CHX was successfully loaded to the cross-linked kapok fibers; the elemental analysis showed that the cross-linked fibers retained a maximum of 7.99 wt% of chlorhexidine, making them feasible materials for drug release applications. The drug release kinetics studies showed a fast burst release within the first 3 h, with the Korsmeyer–Peppas model, which describes better the drug release kinetics.

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