Poly(*N*-Acryloyl Ciprofloxacin-*Co*-Acrylic Acid)-Incorporated Waterborne Polyurethane Leather Coating with Long-lasting Antimicrobial Property

by

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Abstract

To overcome the leaching problem associated with low-molecular-weight antimicrobial agents, a water soluble antimicrobial polymer, poly(*N*-acryloyl ciprofloxacin-*co*-acrylic acid), was designed and synthesized *via* acryloylation of the secondary amine in *7*-piperazinyl substituent of ciprofloxacin and subsequent copolymerization with acrylic acid. The structure of the copolymer was characterized by ^{1}H nuclear magnetic resonance (NMR), Fourier transform infrared (FTIR) spectra, X-ray photoelectron spectroscopy (XPS), and gel permeation chromatography (GPC). The copolymer was compatible with waterborne polyurethane for leather finishing, and remained antimicrobial even though the parent structure of ciprofloxacin molecule was altered. When incorporated into waterborne polyurethane leather coating as an antimicrobial agent, poly(*N*-acryloyl ciprofloxacin-*co*-acrylic acid) proved seldom leached out from the coating even after being rinsed for ten times. Based on these results, the water soluble antimicrobial copolymer designed in this study may prevent depletion of antimicrobial function of waterborne polyurethane leather coating, extending the lifetime of the leather products and enhancing their hygiene properties.

Introduction

Finishing, which includes a series of mechanical operations and, particularly, the application of a polymeric coating, aims at improving the gloss/handle/physical properties and hiding any defects or irregular appearance of the crust leather. Of multiple alternatives, waterborne polyurethane (WPU) has become one of the most prevalent coating-forming materials for leather finishing in modern leather industry. This versatile and environmentally-friendly leather coating material exhibits many unique properties, including superior handle, adhesive strength, improved water vapor permeability, and excellent low temperature flexibility.[1-3] However, polyurethane is inherently vulnerable to microbial attack, which is mainly dictated by its microbially-unstable soft segments (polyether and/or polyester) and various susceptible additives incorporated, such as lignocelluloses, stabilizers, and colorants.[4] Once the temperature and humidity become favorable, microorganisms will proliferate readily on WPU leather coating, leading to degradation, discoloring, and smell, which shortens the lifespan of the leather products.[5,6] In addition, baneful toxins excreted by micro-organisms are also considered pathogenesis of many hygiene-related diseases, posing health threats to consumers.[7,8]

For hygiene and extended lifetime, the normally employed strategy is to confer antimicrobial function to WPU leather coating by incorporating low-molecular-weight antimicrobial agents, such as phenols, chloro-phenols, organic-mercury salts, organic-tin salts, etc.[9] Despite the efficiency of this approach, antimicrobial agents of low molecular weight are generally vulnerable to leaching when the leather coating contacts with water, leading to short duration of action and limited effectiveness which may eventually be depleted. In addition, many traditional low-molecular-weight antimicrobials are toxic, thus leaching of which may also contaminate the environment. On the other hand, it has been recognized that leaching of antimicrobial agents into the environment can result in sub inhibitory concentrations that may give rise to antimicrobial resistance, a progressively severe issue making antimicrobial treatment of leather coating more difficult.[1,9]

Over the past decades, antimicrobial polymers capable of killing microorganisms or inhibiting their growth have gained considerable interests for both academic research and industrial applications. Until now, typical strategies to prepare antimicrobial polymers include covalently conjugating antimicrobial moieties into the polymeric backbone or as

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pendant groups, or preparing a polymerizable monomer with antimicrobial moieties and subsequently polymerizing with other monomers. Compared with low-molecular-weight antimicrobial agents, antimicrobial polymers are preferable, because they are generally non-volatile, chemically stable and carry a high density of antimicrobial moieties, promising prolonged activity as well as high bactericidal efficiency.[10,11] In particular, antimicrobial polymers are generally less susceptible to leaching, which not only guarantees their long-lasting efficiency but also reduces the risks of environmental contamination and development of antimicrobial resistance.

In this study, a low-molecular-weight antimicrobial agent, ciprofloxacin, was acryloylated by reaction of the secondary amine in *7*-piperazinyl substituent with acryloyl chloride, and then copolymerized with acrylic acid to design a water-soluble antimicrobial polymer, poly(*N*-acryloyl ciprofloxacin-*co*-acrylic acid). The chemical structure of this copolymer was characterized by multiple techniques. Then, this copolymer was incorporated into WPU for leather finishing. In comparison with control that contained low-molecular-weight ciprofloxacin only, poly(*N*-acryloyl ciprofloxacin-*co*-acrylic acid) proved compatible with WPU, and more importantly, seldom leached out from the WPU leather coating, which promised long-lasting antimicrobial property. Based on these results, the water soluble antimicrobial copolymer designed in this study may prevent depletion of antimicrobial function of waterborne polyurethane leather coating, extending the lifetime of the leather products and enhancing their hygiene properties.

Experimental Procedures

Materials

Acryloyl chloride (purity ≥96.0%) was supplied by Sigma-Aldrich Co. Ltd. (Shanghai, China). Acrylic acid (AA, recrystallized from chloroform before use, purity ≥99.5%), dichloromethane (DCM, chromatographic grade, purity ≥99.8%), anhydrous *N,N*-dimethylformamide (DMF), *n*-hexane, and diethyl ether were purchased from Kelong Chemical Engineering Co. Ltd. (Chengdu, China). Azoisobutyronitrile (AIBN, recrystallized from ethanol before use) and triethylamine (Et_3N, purity ≥99.0%) were provided by Shanghai Reagent Fourth Factory (Shanghai, China). Ciprofloxacin (CPF, purity≥98.0%) was obtained from the National Institute for the Control of Pharmaceutical and Biological Products (Beijing, China). LIVE/DEAD® BacLight™ Bacterial Viability Kit (catalog no. L-7012) containing SYTO 9 dye and propidium iodide for microscopy and quantitative assays was purchased from Thermo Fisher Scientific. (Waltham, MA, USA). WPU dispersion with a solid content of 30wt%, designed for leather finishing, was supplied by Wanhua chemical group co., LTD. (Yantai, China). It was synthesized from methylene isophorone diisocyanate (IPDI), polyetherester-based diol (composed of adipate acid, polyether polyol and *1,4*-butane diol, with a molecular weight of 2000 g/mol), and a mixed chain extender of ethylene glycol, dimethylol propionic acid and ethylenediamine. Cattle crusts with an average thickness of 1.0 mm were kindly donated by a local tannery (Chengdu, China).

Preparation of *N*-acryloyl Ciprofloxacin (NACPF)

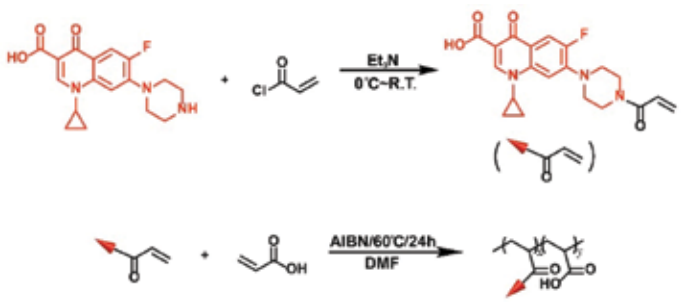
CPF powder (1.66 g, 5 mmol, 1 eq) and an excessive amount of Et_3N (0.66 g, 6.5 mmol, 1.3 eq) were first dissolved in 20 mL anhydrous DCM, which was stirred at 0°C under N_2 protection. After vigorously stirred for 30 min, acryloyl chloride (0.68g, 7.5 mmol, 1.5 eq) dissolved in 10 mL anhydrous DCM was added to the mixture dropwise over 15 min. After that, the mixture was stirred at ambient temperature for another 1 h. The resulting transparent yellow solution was then precipitated with *n*-hexane, and finally filtered to obtain the precipitates. Subsequently, the precipitates were washed with Milli-Q water (18.2 MΩ·cm at 25°C) repeatedly, and vacuum dried to yield *N*-acryloyl ciprofloxacin (NACPF, 1.43 g, 74%).

Synthesis of Poly(*N*-acryloyl ciprofloxacin-*co*-acrylic acid)

Poly(*N*-acryloyl ciprofloxacin-*co*-acrylic acid) was synthesized by free-radical copolymerization of acrylic acid (AA) and NACPF monomers, employing AIBN as an initiator. Briefly, NACPF (0.49 g; 1.27 mmol) and AA (5.56 g; 77.2 mmol) were dissolved in 28 ml anhydrous DMF in a Schlenk tube. The mixture was deoxygenated three times using a freeze-pump-thaw method, followed by adding AIBN (1 mol%) dissolved in 2.0 mL DMF *via* a syringe and then continuously stirring at 60°C under nitrogen atmosphere. After 24 h, the reaction was terminated by cooling down to ambient temperature. The co-polymer was precipitated repeatedly from diethyl ether, and the final solid was dried under vacuum to yield poly(*N*-acryloyl ciprofloxacin-*co*-acrylic acid), or poly(NACPF-*co*-AA). The synthesis procedure mentioned above was depicted in Scheme 1.

Characterization of Poly(NACPF-*co*-AA)

^1H nuclear magnetic resonance (NMR) spectra were recorded on a Bruker AVANCE III NMR (500 MHz) instrument (Bruker, Switzerland). Solid sample of approximately 5 mg was dissolved



Scheme 1. Synthesis procedure and structure of poly(NACPF-*co*-AA).

in 1 ml deuterated dimethyl sulfoxide (DMSO-d_6, 99.9 atom% D) containing 0.03% (v/v) tetramethylsilane (TMS). The mixture was sealed in a Pyrex NMR tube of 6 mm in diameter prior to analysis. The spectra reported were a sum of 32 scans, and the delay time between each scan was 5 s. Chemical shifts (δ) were expressed in parts per million (ppm), relative to the TMS signal as an internal standard.

Fourier transform infrared (FTIR) spectra were collected at ambient temperature using a Nicolet iS10 FTIR spectrometer (Thermo Scientific, USA) over a wavenumber range from 400 to 4000 cm^{-1} after 32 scans at 4 cm^{-1} resolution. KBr pellet technique was employed for sample preparation. For NACPF monomer in powder form, approximately 1 mg sample was ground with 200 mg spectroscopic grade KBr that had been pre-dehydrated at 200 °C for 24 h. Subsequently, 90 mg of the mixture was compressed in an evacuated stainless steel die under a pressure of 526 MPa to form a pellet of about 1 mm thick and 13 mm in diameter. For poly(NACPF-*co*-AA), it was dissolved in methanol, dropped onto a pure KBr pellet and dried under infrared lamp for 1 h before the measurement. A pure KBr pellet was employed as reference and its spectrum was subtracted from the sample spectra to suppress any spectral artifact caused by impurities and/or water.

X-ray photoelectron spectroscopy (XPS) was utilized to qualitatively analyze the elemental composition of poly(NACPF-*co*-AA). The measurements were carried out on a KRATOS XSAM800 X-ray photoelectron spectrometer with a monochromatic Al Kα X-ray source (1486.6eV), operating at 20 kV with a current of 10 mA. C 1s, N 1s, O 2s and F 1s spectra were detected with an air pressure of 2×10^{-7} Pa in the analysis chamber. The binding energy (BE) scale was regulated by setting the C 1s transition at 284.8 eV. The PeakFit Version 4.12 software (SPSS Inc., Chicago, IL, USA) was employed to deconvolve the peaks using the Shirley-type baseline to separate different species of the same element.

Number-average molecular weight (M_n) of poly(NACPF-*co*-AA) was determined using a gel permeation chromatography (GPC) system equipped with a Waters 2690D separation module and a Waters 2410 refractive index detector. The sample was dissolved in 0.1 M sodium nitrate to form a 1 g L^{-1} solution (pH=7.4). After filtration, 20 µl of the prepared solution was injected into the GPC system and eluted at 30°C with a flow rate of 1 ml min^{-1}. Waters millennium module software was used to calculate the molecular weight, based on a universal calibration curve generated by polyacrylic acid standard with a narrow molecular weight distribution.

Crust Leather Finishing by Poly(NACPF-*co*-AA)-incorporated WPU

Formulation of the finishes was tabulated in Table I. The crust was finished by applying the formulated finishes *via* spraying method. After spraying the first coat, the leather was dried at ambient temperature for 10 h, and then plated at 100°C, 80 atm for 30 s. Subsequently, the second coat was sprayed, and the finished leather was cured at 80°C for 5 h. The thickness of the WPU coating was about 0.1-0.15 mm.

Scanning Electron Microscope (SEM) Observation

Scanning electron microscope (SEM, Phenom ProX, Phenom-World BV, Netherlands) was employed to observe the surface morphologies of the crust leather and the poly(NACPF-*co*-AA)-incorporated WPU finished leather. The samples were subject to SEM-observation under the following operating condition: accelerating voltage 10.00 kV; emission current 40 µA; working distance 5.5 mm.

Bactericidal Test

Staphylococcus aureus (*S. aureus*, ATCC 25923) and *Escherichia coli* (*E. coli*, ATCC 25922) were selected as the indicator microorganisms to test the antibacterial activity of poly(NACPF-*co*-AA)-incorporated WPU leather coating. The strains were aerobically precultured in Müller-Hinton (MH) broth

Table I
Formulation of the finishes.

WPU (w/w)	Black N932 Pigment[a] (w/w)	FL400 filler[b] (w/w)	BYK333 slip agent[c] (w/w)	Water (w/w)	poly(NACPF-*co*-AA)(w/w)
25	10	2	0.1	55	7.9

[a] Donated by Dowell Science & Technology Inc. (Sichuan, China).
[b] Purchased from DFCL Import and export trade co., LTD. (Guangzhou, China).
[c] Supplied by BYK Additives & Instruments. (Wesel, Deutschland).

(containing 2.0 g L^{-1} beef extract powder, 17.5 g L^{-1} acid digest of casein and 1.5 g L^{-1} starch, pH=7.4) at 37 °C for 12 h, and then harvested by centrifugation (5000 rmp for 10 min) and washing with phosphate buffer saline (PBS) three times. Before use, the bacteria cultivated were diluted to a concentration of 1×10^8 colony-forming units (CFU) mL^{-1} with PBS.

The antibacterial activity of poly(NACPF-*co*-AA) was qualitatively assessed by a standard Bauer–Kirby disk susceptibility test. A disk of filter paper, impregnated with 1.0 mg mL^{-1} poly(NACPF-*co*-AA) aqueous solution (pH=7.4), was placed in a Petri plate containing Müller-Hinton (MH) agar previously seeded with 1×10^8 CFU mL^{-1} of *E. coli*. After incubation at 37 °C for 24 h, the size of the inhibition zone of bacterial growth around the disk was measured.

To visualize the bacterial cells on the WPU leather coating surface, a LIVE/DEAD Baclight bacterial viability kit with two staining agents was used. This viability assay involves two fluorescent dyes that can bind to nucleic acids: the green stain SYTO 9 and the red stain propidium iodide. SYTO 9 labels both live and dead cells, whereas propidium iodide can penetrate only cells with compromised or damaged membranes. Moreover, the red propidium iodide abolishes the green fluorescence of SYTO 9 in damaged cells by displacing it from complexes with nucleic acids. Consequently, bacteria with intact membrane fluoresce green, while those with damaged cell membrane fluoresce red [12]. A suspension of *S. aureus* in PBS (10^5 CFU mL^{-1}, 50 µL) was seeded on the WPU leather coating, followed by incubation at 35°C for 24 h. After incubation, the leather coating surface was washed with PBS buffer three times, and soaked in the dye solution (4 µL SYTO 9 and 6 µL propidium iodide in 8 mL PBS) at room temperature in dark for 15 min. The stained bacterial cells were observed using an IX-71 Inversed Fluorescent Microscope (Olympus America Inc., Melville, NY). The open-source software *Image J* (NIH, Bethesda, USA) was utilized to process the fluorescent images obtained above.

The long-lasting antibacterial property of poly(NACPF-*co*-AA)-incorporated WPU leather coating were evaluated according to the following process. First, a suspension of *S. aureus* or *E. coli* in PBS (10^5 CFU mL^{-1}, 50 µL) was seeded on the poly(NACPF-*co*-AA)-incorporated WPU coating, followed by incubation at 35°C for 24 h. After that, all the bacteria on the coating surface were transferred and re-suspended in PBS, and the bacteria concentration survived in the PBS solution was counted by agar plate counting method. The antibacterial efficiency was quantified by the percentage reduction in the bacteria concentration. Subsequently, the coating was immersed in Mili-Q water, and the mixture was shook on an agitation shaker with the speed of 150 r min^{-1} at 25°C for 20 min. After that, the coating was dried at 30°C until a constant weight, and the *S. aureus* or *E. coli* suspension was seeded again to test the antibacterial activity of the rinsed coating. This rinsing process was repeated for ten times. WPU coating incorporated with CPF was used as control in all these trials.

Statistics

The reported data were means±standard deviation of quintuplicate samples for each measurement. Statistical analysis was performed with Student's *t*-test. $P < 0.05$ was accepted as statistically significant.

Results and Discussion

In general, low-molecular-weight antimicrobial agents suffer from leaching, which compromises the long-lasting antimicrobial property of the leather coating. In the present study, a broad-spectrum hydrophobic antimicrobial, ciprofloxacin, was acryloylated by the reaction of the secondary amine in *7*-piperazinyl substituent with acryloyl chloride, and converted into a polymerizable monomer. Such reaction exerted no influence on the *3*-carboxyl and *4*-keto in ciprofloxacin, both essential for hydrogen-bonding with microbial DNA to elicit antimicrobial activity[13,12]. It also imposed no influence upon the bicyclic heteroaromatic quinolone pharmacophore, π-π ring stacking of which permitted flexibility in adjusting position of ciprofloxacin for optimal hydrogen-bonding configuration. Thus, the resulting *N*-acryloyl ciprofloxacin remained microbiologically active even though the peripheral substituent of the parent antimicrobial was altered.[14-17] Herein, acrylic acid was utilized to copolymerize with *N*-acryloyl ciprofloxacin. This was because ciprofloxacin contains a quinolone nucleus of hydrophobicity and multiple hydrophobic substitutes, leading to limited aqueous solubility. Copolymerization with acrylic acid was intended to improve the water solubility of the antimicrobial polymer, and thus its compatibility with WPU for leather finishing.

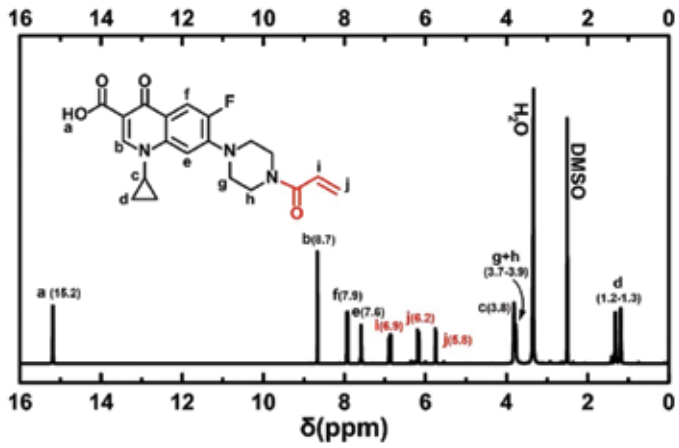


Figure 1. ^{1}H NMR spectrum of NACPF.

Herein, successful acryloylation of ciprofloxacin could be confirmed by analyzing the ^{1}H NMR spectrum of NACPF displayed in Figure 1. Corresponding to those chemically nonequivalent protons located in CPF moiety, a series of resonance peaks at around 1.2-1.3, 3.7-3.9, 3.8, 7.6, 7.9, 8.7, and 15.2 ppm were observed in NACPF. The position and relative intensity of these peaks were found consistent with previous reports on pure CPF.[12,18] In addition, three extra resonance signals peaking at around 5.8, 6.2, and 6.9 ppm appeared after the CPF molecule was acryloylated. They could be ascribed to the protons in vinyl group, which provided possibility for polymerization with acrylic acid.

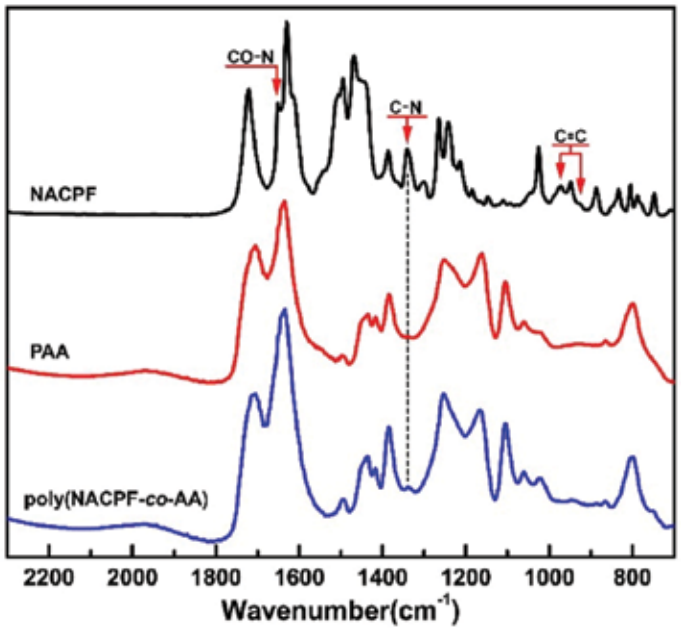


Figure 2. FTIR spectra of NACPF, PAA, and poly(NACPF-*co*-AA).

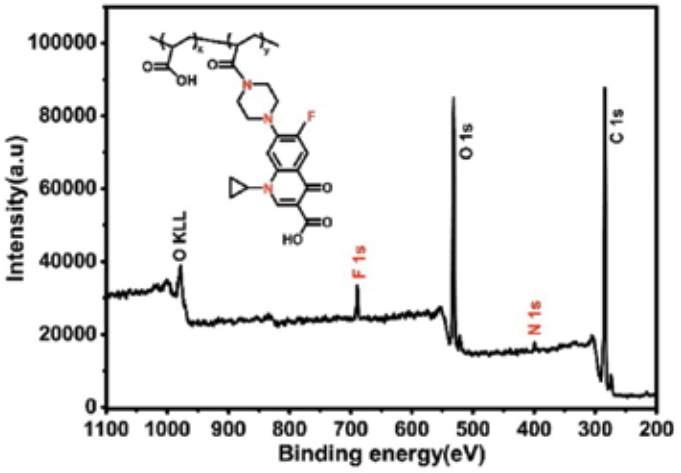


Figure 3. XPS survey spectrum of poly(NACPF-*co*-AA).

After copolymerization, the presence of CPF moieties in the copolymer could be verified by FTIR and XPS analysis. The FTIR spectra of NACPF and poly(NACPF-*co*-AA) were demonstrated in Figure 2. For comparison, the FTIR spectrum of pure PAA, synthesized under the same conditions, was also provided. As for NACPF, the characteristic absorption band peaking at 1650 cm^{-1} was associated with amide I band,[18] which was produced by the reaction of the secondary amine in CPF with acryloyl chloride. As a result, vinyl group was successfully introduced to CPF, manifesting by the presence of two infrared absorption bands peaking at 971 and 926 cm^{-1} in NACPF. After copolymerization, the presence of CPF moieties in poly(NACPF-

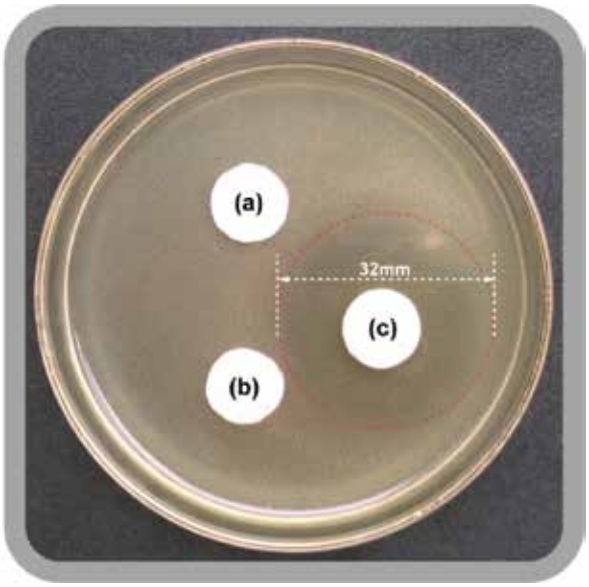


Figure 4. Photographs of inhibition zone against *E. coli* around disks of filter paper impregnated with (a) Milli-Q water, (b) 1.0 mg mL^{-1} PAA solution, and (c) 1.0 mg mL^{-1} poly(NACPF-*co*-AA) solution (pH=7.4).

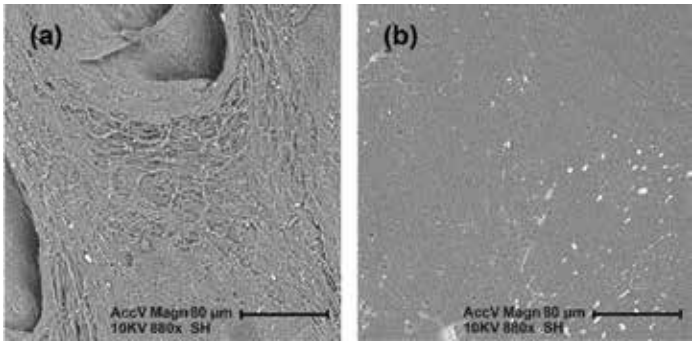


Figure 5. Scanning electron microscopic image of (a) crust leather surface and (b) poly(NACPF-*co*-AA)-incorporated WPU leather coating surface.

co-AA) could be discerned by the absorption bond peaking at 1338 cm^{-1}, associated with the vibration absorption of C-N.[19] This band could only be ascribed to CPF moieties because it could not be detected in pure PAA. The XPS survey spectrum for poly(NACPF-*co*-AA) was illustrated in Figure 3. In addition to C and O elements, F and N elements could also be detected in the copolymer. This observation also indicated the presence of CPF moieties in the copolymer.

In the present study, Bauer-Kirby disk susceptibility test was employed to evaluate the antibacterial activity of poly(NACPF-*co*-AA) by using *E. coli* as the indicator. As displayed in Figure 4, robust growth of *E. coli* could be observed around control, a disk of filter paper impregnated with Milli-Q water. Similarly, *E. coli* colonies were found to be able to grow up to the edge of that disk impregnated with pure PAA, forming an opaque culture. After copolymerized with NACPF, a visible zone of inhibition with a diameter of approximately 32 mm appeared around the disk impregnated with poly(NACPF-*co*-AA), suggesting that the ciprofloxacin moieties in the copolymer remained biologically active.

The molecular weight of antimicrobial polymers plays an important role in determining their antimicrobial properties[9]. Although antimicrobial polymers usually carry a high density of antimicrobial moieties, promising high bactericidal efficiency, molecular weight larger than a threshold leads to a decrease in activity. This dependence on molecular weight can be attributed to the sequence of steps necessary for biocidal action. After all, polymers with an extremely large molecular weight have trouble diffusing through the bacterial cell wall and cytoplasm. By using GPC analysis, the number-average molecular weight of poly(NACPF-*co*-AA) was determined to be 74211 g mol^{-1}. As shown in Figure 4, poly(NACPF-*co*-AA) exhibited excellent antimicrobial activity against *E. coli*. In a follow-up study, the molecular weight-dependence of antimicrobial activity in poly(NACPF-*co*-AA) will be systematically investigated to determine its optimal molecular weight for maximized antimicrobial activity.

With the antimicrobial activity of CPF uncompromised in poly(NACPF-*co*-AA), the copolymer was employed as an antimicrobial agent in WPU for leather finishing. Figure 5 exhibited the surface morphology of cattle crust, and that finished by poly(NACPF-*co*-AA)-incorporated WPU. It could be observed that the collagen fibers and pores possessed by the crust leather were covered up after finishing, which resulted in a uniform and smooth surface. The antimicrobial polymer was completely compatible with WPU, and therefore no phase separation was observed.

After incubation of *S. aureus* on poly(NACPF-*co*-AA)-incorporated WPU leather coating, two fluorescent nucleic acid dyes, SYTO 9 and propidium iodide, were used to stain the DNA of the bacteria. As shown in Figure 6, there were few dead bacteria on pure WPU leather coating; most of the seeded bacteria remained viable, as indicated by the green fluorescence. This was because the polyether and/or polyester segments of WPU macromolecules, as well as other additives, could serve as carbon sources for microbial growth. However, on WPU leather coating incorporated with poly(NACPF-*co*-AA), almost all the bacteria were stained by propidium iodide, showing red fluorescence. The predominance of red fluorescence in this case

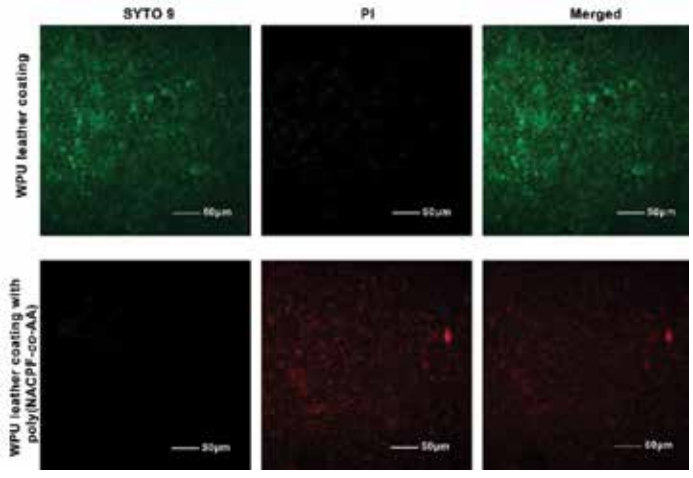


Figure 6. Fluorescent images showing *S. aureus* adhered on the surface of WPU leather coating and poly(NACPF-*co*-AA)-incorporated WPU leather coating (green, live bacteria; red, dead bacteria). The merged images represent the superposition of live and dead bacteria.

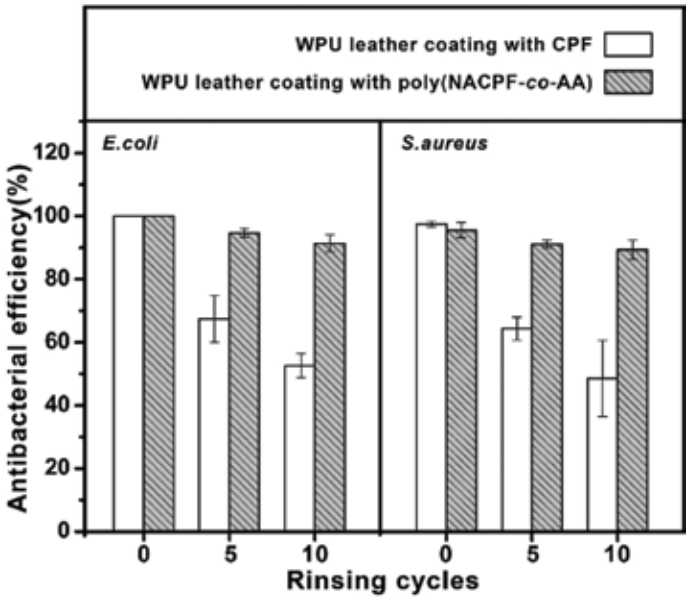


Figure 7. Antibacterial efficiency (%) of poly(NACPF-*co*-AA)-incorporated WPU leather coating against *E. coli* and *S. aureus versus* rinsing cycle.

indicated that the bacteria lost viability after contacting with the coating. This result suggested again that poly(NACPF-*co*-AA) remained biological activity even though the parent structure of CPF had been altered.

Compared with low-molecular-weight antimicrobial agent such as CPF, antimicrobial polymers are seldom leached out from the devices because of their large molecular weight, which promised their long-lasting function. In the present study, *S. aureus* and *E. coli* were selected as indicators to quantitatively investigate the long-lasting antibacterial property of poly(NACPF-*co*-AA)-incorporated WPU leather coating. As could be seen in Figure 7, when poly(NACPF-*co*-AA) was incorporated, the WPU leather coating displayed excellent antimicrobial property against both strains, manifesting as an antibacterial efficiency of 100% for *E. coli* and more than 95.6% for *S. aureus*. These results indicated that incorporation of poly(NACPF-*co*-AA) could confer good antimicrobial property to the WPU leather coating. However, it was found that the antibacterial activity originating from poly(NACPF-*co*-AA) could be conserved after rinsing for ten times, and the antibacterial efficiency against both strains remained above 89.8%. The slight decline could be ascribed to the loss of poly(NACPF-*co*-AA) on the coating surface, but most poly(NACPF-*co*-AA) molecules were trapped and reserved in the coating. On the contrary, the majority of low-molecular-weight CPF molecules were lost during the rinsing cycles, and therefore, the antibacterial efficiency of CPF-incorporated WPU leather coating against both strains decreased significantly. For example, after rinsed for five times, the antibacterial efficiency of CPF-incorporated WPU leather coating decreased from 100% to 68.1% for *E. coli*, and from 97.7% to 64.2% for *S. aureus*. After rinsed for ten times, the antibacterial efficiency decreased to 52.3% for *E. coli* and 49.1% for *S. aureus*, respectively. According to these results, it was found that poly(NACPF-*co*-AA), as an antimicrobial agent, was seldom leached out from WPU leather coating, exhibiting long-lasting antimicrobial function compared with low-molecular-weight CPF.

Conclusions

A water-soluble antibacterial polymer, poly(NACPF-*co*-AA), was successfully synthesized *via* initial acryloylation of the secondary amine in *7*-piperazinyl substituent of ciprofloxacin, and subsequent copolymerization with acrylic acid. The protocol for structural tailoring of ciprofloxacin molecules adopted herein did not perturb its biological activity. The poly(NACPF-*co*-AA)-incorporated WPU leather coating exhibited excellent antimicrobial activity even after being rinsed for ten times, indicating a long-lasting antimicrobial property. These results suggested potential application of poly(NACPF-*co*-AA) as an antibacterial agent for leather finishing, which promised long-lasting hygiene and extended lifetime.

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