Drug Loaded Poly(Vinyl Alcohol) - Cellulose Composite Hydrogels for Wound Dressings

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Abstract

Biodegradable and biocompatible synthetic polymers like poly(vinyl alcohol) (PVA) have attracted much attention for wound dressing applications. However, PVA hydrogel possesses insufficient strength and very limited hydrophilicity characteristics which restrict its use as a wound dressing polymeric material. In the present work a suitable wound dressing hydrogel films were prepared with PVA-cellulose using freezing-thawing method. Standard antibiotics (ciprofloxacin and streptomycin) and natural herbals such as turmeric and *Tridax procumbens* plant extract were incorporated in PVA-cellulose hydrogels and its wound healing properties were evaluated. The synthesized hydrogels were characterized using scanning electron microscopy (SEM), X-ray powder diffraction (XRD) and fourier transform infra-red (FT-IR) spectroscopy techniques. The mechanical strength, in-vitro swelling studies, water vapour transmission rate, gel fraction test, degradation and antibacterial tests were carried out to study its wound dressing characteristics. The PVA cellulose hydrogels with *Tridax procumbens* showed good swelling ability and optimum antibacterial effect against pathogenic organisms as similar to ciprofloxacin loaded hydrogels. From this study, it is evident that PVA cellulose hydrogels with addition of *Tridax procumbens* could be used as a potential wound dressing material.

Keywords: Wound healing; hydrogels; PVA-cellulose composites; natural drugs

1. Introduction

Wound dressings play a crucial role in the management and promotion of wound healing. They should be attributed with ideal characteristics such as thermal insulation, absorption and retention of wound fluids and exudates. The wound dressings should also provide protection from further infections, prevent dehydration of wound, allow exchange of gases and on removal it should not cause any pain or trauma [1]. Studies have reported that hydrogels are credited to satisfy desirable wound dressing characteristics [2]. However, the use of hydrogel is often limited owing to their poor mechanical strength [3]. In order to overcome this problem polymer composite hydrogels are usually used which have sufficient mechanical strength without causing any significant change in their properties. Poly(vinyl alcohol) (PVA) is a synthetic polymer formed by free radical polymerization of vinyl acetate. Due to good water-solubility, biodegradability, non-carcinogenicity and biocompatibility PVA is used to blend with natural and synthetic polymers to enhance wound dressing applications [4]. Cellulose is a polysaccharide consisting of a linear chain of glucose molecules held together by 1, 4 β-glucosidic linkages. It has been widely used in medical materials. These membranes are highly nano-porous in nature which enables transfer of drugs into the wound site as well as serve as an effective barrier against microbial infections [5]. The unique nanostructure also provides excellent mechanical strength to the hydrogels. Streptomycin is a broad spectrum bactericidal antibiotic, derived from *Streptomyces griseus.* It acts against mycobacteria and belongs to the class of amino glycoside drugs. It is used to for sterilization of wound present on the surfaces prior to skin grafting [6]. Ciprofloxacin is also a broad spectrum antibiotic of the fluoroquinolone class [7]. Tridax (*Tridax procumbens)* is a known medicinal herb and is commonly used for its anti-inflammatory and wound healing activities. The leaf extract of tridax is used to arrest bleeding from bruises and cuts [8]. Turmeric *(Curcuma longa)* contains curcumin that is responsible for the antibacterial and anti-inflammatory activities of turmeric [9]. There have been a few studies where PVA-cellulose composite hydrogels were synthesized [10-12]. In the present study, PVA-cellulose hydrogels with streptomycin, ciprofloxacin, tridax and turmeric were prepared by freeze-thawing. The prepared hydrogels were characterized and evaluated for wound dressing behavior by conducting swelling, water vapor transmission rate, gel fraction, tensile studies and antibacterial studies.

2. Materials and methods

2.1. Materials

Polyvinyl alcohol (PVA, hot water soluble) and cellulose were purchased from Himedia. Streptomycin and ciprofloxacin were purchased from SRL, India. Wild *Tridax procumbens* was obtained from the locality and commercially available turmeric powder was procured.

2.2. Preparation of PVA-cellulose hydrogels loaded with drugs

Tridax procumbens leaves were collected from the Rourkela region, Sundargarh District, Orissa and extracted using Soxhlet extractor with 95% ethanol at 50˚C. Accordingly PVA (10% w/v) was dissolved in distilled water with continuous stirring at 90°C. After 2h of stirring 5% w/v of cellulose was added and stirred. Then, 0.1% w/v of drug was mixed. The prepared solution was freezed and thawed for three times.

2.3. Characterization

The surface morphology of the synthesized composite hydrogels was observed using scanning electron microscopy (SEM JSM 6480LV, USA). The X-ray diffraction (XRD) analysis was performed with a diffractometer (Rigaku Ultima IV Diffractometer, Japan) from 15° to 60°, with a scan speed of 5°/min and step size of 0.05°. FTIR (Perkin Elmer) spectroscopy was performed over the scanning range of 400 – 4000 cm^{-1}.

2.4. In-vitro biological evaluation

2.4.1. Swelling Studies

Weighed dried samples (W_d) were soaked in 10 ml of PBS at 37°C. Then the samples were taken out at an interval of 1 h, surface dried and re-weighed (W_w). The swelling degree S_w (%) was calculated using the equation (1) [13]:

$$S_w(\%) = [(W_w - W_d)/W_d] \text{ X } 100 \qquad (1)$$

2.4.2. Water vapour transmission rate (WVTR)

Cylindrical plastic bottles were taken and 15ml of water was added to it. The samples were cut and used as a cap on the mouth of the bottle. Then the initial weight of the setup was taken (W_i) and kept in an incubator at 37ºC for 24 h. After 24 h the final weight of the setup is recorded (W_f). The following formula (2) was used to calculate WVTR [2]:

$$\text{WVTR} = 10^6 \times \left[\frac{(W_f - W_i)}{24 \times A}\right] \text{ g/m}^2\text{h} \qquad (2)$$

2.4.3. Gel fraction studies

Preweighed samples were dried under vacuum at room temperature until no change in its mass was observed. Another sample of identical weight was immersed into water for 4 days to rinse away unreacted species and dried at room temperature under vacuum until constant weight was attained. The gel fraction was calculated by using the following equation (3) [13]:

$$G(\%) = \frac{W_d}{W_i} X\ 100 \qquad (3)$$

Where W_i is the initial weight of dried samples and W_d is the dried insoluble weight of the samples.

2.4.4. Tensile strength measurement

Tensile strength of the developed composite scaffolds was carried out on a universal testing machine (ElectroPuls E1000, Instron, UK) using a load cell of 250 N and a strain rate of 2 mm/min.

2.4.5. Antibacterial study

Nutrient agar plates were prepared and inoculated with *S.aureus* (gram-positive) and *E.coli* (gram-negative). PVA-Cellulose samples with drugs were cut in circular shapes with a diameter of 1 cm and kept on agar plates.

3. Results and discussion

3.1. Scanning electron microscopy (SEM)

The SEM micrographs of the samples are shown in Figure 1. The SEM micrographs of all the composite samples showed uneven surfaces. Comparing all the samples, PCTri have demonstrated the highest amount of surface smoothness that may be due to proper mixing of tridax extract with the PVA-cellulose polymer matrix. The surfaces of PCC and PCS incorporated with chemical drugs showed an uneven drug distribution.

Figure 1 - Scanning electron micrographs of PVA-cellulose composites.

3.1. X-ray diffraction studies

The XRD patterns of drug incorporated PVA-cellulose composites are shown in Figure 2(a). PVA showed its characteristic diffraction peak at 2θ = 19.5˚ and 40.06˚. The peaks obtained at 2θ=23.1˚ and 2θ=34.9˚ corresponds to cellulose. There was no strong peak for drugs in composite hydrogels. This might be due to the lower concentration of AC as compared to PVA in the composites.

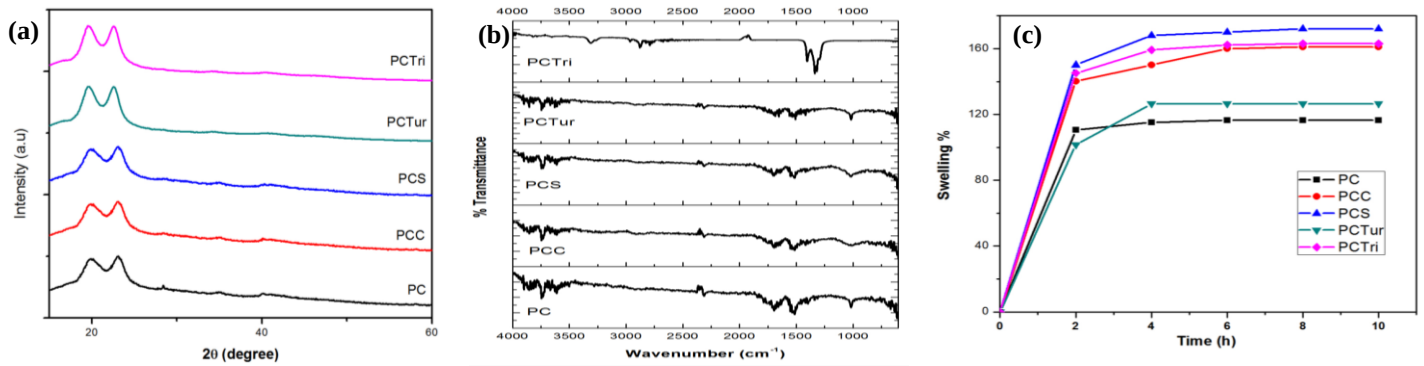

Figure 2 – (a)-XRD, (b) FTIR and (c) swelling patterns of PVA-cellulose composites

3.2. Fourier transform infrared spectroscopy (FTIR)

The FTIR spectra of the PVA-cellulose composite samples are shown in Figure 2(b). Several characteristic bands were obtained from the FTIR spectra of the samples corresponding to various functional groups and chemical bonds. The peaks around 3500-3900 cm^{-1} correspond to O-H stretching vibrations. The PCTri hydrogel showed a peak for $–CH_2$ stretching at 2904 cm^{-1}. A broad peak at 1725 cm^{-1} indicates the C=O stretching of the ester group in PVA. The absorption band associated with C=C stretching was observed around 1526 cm^{-1}. Another band around 1011 cm^{-1} represents C-O stretching vibrations of cellulose. In the PCTri sample peaks at 1326 cm^{-1} and 1410 cm^{-1} correspond to benzene ring and C-H stretching respectively.

3.3. Swelling studies

Swelling percentage curves of the PVA-cellulose composites are shown in Figure 2(c). The equilibrium state of swelling was achieved within 10 h. The swelling profiles of the drug loaded samples clearly indicate a linear increase in the swelling percentages and got saturated after 4 h. The sample PCS showed highest swelling percentage whereas PC showed lowest swelling percentage. Thus the incorporation of both natural and chemical drugs increased the water absorbing capacity of the hydrogels. This could be due to decrease in crosslinking with addition of drugs [14].

3.4. Water Vapor Transmission Rate

The water vapor transmission rate (WVTR) values for normal skin and diseased skin have been reported to be 8.5 g/m^2 to 11.6 g/m^2 per hour [15]. The results were 6.5, 8.7, 7.7, 6.5 and 8.3 g/m^2h for PC, PCC, PCS, PCTur and PCTri respectivrly. The WVTR values of PCC and PCTri are in the range of required WVTR values followed by PCS. Hence, PCC, PCS, and PCTri hydrogel composites could be used in moderate exudative wounds.

3.5. Gel fraction studies

The results were 91.6, 77.1, 56, 85, 82.1 % for PC, PCC, PCS, PCTur and PCTri respectively. The results of the gel fraction study indicate PC with highest gel fraction and PCS with the lowest gel fraction among the samples. This indicates that PC was completely crosslinked. The drugs reduced the cross-linking interaction between polymers hence reducing the gel fraction of the composite hydrogels with drugs. The gel fraction of PCTri was found to be around 82% that is an ideal gel fraction value for wound dressing applications.

3.6. Mechanical testing

The results obtained from mechanical testing are shown in Table 1. A slight decrease in tensile strength and Young's modulus was observed with the addition of different drugs. The sample PCTri showed optimum values for gel fraction and hence, mechanical strength was also good. However, the hydrogels loaded with natural drugs showed better mechanical strength as compared to those with chemical drugs.

Table 1 Mechanical testing result of PVA-cellulose composites

Sample	Tensile strength (MPa)	Young's modulus (MPa)
PC	25.7±3.4	456.2±22.5
PCC	22.5±2.3	310.6±15.6
PCS	20.7±1.2	278.6±12.5
PCTur	23.6±1.6	422.5±17.8
PCTri	23.4±0.8	436.9±16.3

3.7. Antibacterial study

The results are shown in Table 2. On testing the samples against *S. aureus*, it was found that the PC has shown no zone of inhibition and PCC has shown a maximum zone of inhibition. The sample PCTri showed a significant zone of inhibition and PCTur showed a small zone of inhibition against *S. aureus.* A similar trend was observed when the samples were tested against *E. coli* with PCC, PCS and PCTri showing the maximum zone of inhibition. The PCTri have shown a significant zone of inhibition against both gram-positive *S. aureus* and gram-negative *E. coli* bacteria.

Table 2 Antibacterial activity of PVA-cellulose composites towards *S. aureus and E. Coli*

Sample	**Diameter of zone of inhibition (cm)**	
	S. aureus	***E. coli***
PC	0	0
PCC	3.5	3.4
PCS	2.5	2
PCTur	1.5	1.4
PCTri	2.1	1.8

Conclusion

PVA-cellulose composites incorporated with chemical and natural drugs were prepared by cyclic freeze thaw method and characterized using SEM, XRD and FTIR. The wound dressing characteristics were also evaluated by performing swelling studies, WVTR, antimicrobial studies, gel fraction studies and mechanical testing. Micrographs of all the samples have shown ample roughness which will help in good adherence of the samples to the wound. No peaks for drugs were obtained in XRD since they were used in a very scanty amount (0.1%). FTIR studies of the sample revealed the presence of several characteristic bands corresponding to various functional groups and chemical bonds present in the samples. The WVTR studies indicated that PCTri and PCC may be used in wound dressing application since their WVTR values lies in the range of normal skin and diseased skin. Swelling and gel fraction studies further confirmed that the PCTri could be used as wound dressing materials. The tensile strength measurement showed the sample PC and PCTri has good strength when compared to other samples. The antibacterial study also indicated that PCC, PCS and PCTri exhibited good antimicrobial activity and thus they can be used for wound dressing applications. According to the results the samples were found to satisfy wound dressing properties.

Acknowledgement

Authors acknowledge the logical support provided by the Department of Biotechnology and Medical Engineering, National Institute of Technology, Rourkela for the successful completion of this research study.

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