HYDROGEL MEMBRANES OF PVP (POLY-VINYL PYRROLIDONE) AND BACTERIAL CELLULOSE

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Production of cellulose film by alcoholic fermentation was accomplished utilizing *mosto* composed by fresh pineapple juice with 0.4% initial acidity as bacterial culture medium. The cellulose film produced by this method was washed with distilled water and dried during 24h at 40°C. Characterization were performed against commercial pure cellulose as control by thermal gravimetric analysis (TGA), X-ray diffraction, scanning electron microscopy (SEM) and energy dispersive spectroscopy (EDS). Cellulose film triturated in water and dried films were used to prepare hydrogel membranes of PVP and obtained membranes showed improved property presenting 100% of swelling capacity within 50 minutes.

Introduction

Cellulose is one of the oldest, most abundant natural, renewable, biodegradable, and biocompatible polymers [1]. It has established a long record of applications in various areas such as the textile industry, the paper industry, and the biomedical field as tissue engineering materials due to their good biocompatibility, mechanical properties similar to those of hard and soft tissue and easy fabrication into a variety of shapes with adjustable interconnecting porosity [2-4-]. Recently, bacterial cellulose (BC) has been investigated as a potential scafold for tissue engineering [5-6].

Gluconacetobaqcter xylinus (old nomenclature, *Acetobacter xylinum*) is a cellulose secreting bacteria that is propelled by the secretion of BC (bacterial cellulose) [7]. Cellulosic microfibrils are produced extracellularly by bacteria of the genus *Gluconacetobacter* cultivated in a culture medium containing carbon sources such as sugar and nitrogen sources [8]. The microfibrils are in ribbon form composed of subfibrils. The microfibrils have good physical properties different from those of plant cellulose and therefore are expected to be a new functional material [9-10].

The cellulose synthesized by *Gluconacetobacter xylinus* (*G. xylinus*) is identical to that made by plants in respect to molecular structure; however, the secreted polysaccharide is free from lignin, pectin and hemicelluloses as well as other biogenic products, which are associated with plant cellulose. Additionally, extracellularly synthesized BC microfibrils differ from plant cellulose with respect to its high crystallinity, highwater absorption capacity and mechanical strength in the wet state [11]. The microfibrils are in ribbon form composed of subfibrils. The loci of the biosynthesis of subfibrils are formed on the surface of bacterium as a linearly order array of terminal complexes [8].

The aim of this work was to improve the mechanical properties of PVP hydrogel membranes by addition of bacterial cellulose in the formulation.

Experimental

BC obtained by alcoholic fermentation was examined by thermal gravimetric analysis, X-ray diffractometry (XRD), scanning electron microscopy (SEM) and energy dispersive spectroscopy (EDS). Hydrogel membranes of PVP and BC were analyzed by their sweelling capacity.

Thermal gravimetric analysis (TGA):

The samples were analyzed to the speed of 10°C.min^{-1}, in the interval from 30 to 650°C, using sample mass around 10mg and atmosphere of N_2 (20mL.min^{-1}).

X-ray diffraction:

X-ray diffraction data were collected in the 2θ interval from 5 to 90°, with a step time of 3s and step size equals to 0.02°, using a SIEMENS D5000

diffractometer with copper radiation monocromatized by a graphite crystal. The diffractometer was set at 40kV and 30mA.

Scanning Electron Microscopy (SEM) and Energy Dispersive Spectroscopy (EDS):

The samples were nitrogen frozen and broken. The broken superficial area of celluloses samples were prepared and analyzed by Philips XL microscope at 5000 and 7000x amplitude.

Hydrogel membranes preparation:

Different formulations of hydrogel membranes were prepared with PVP from BASF, poli ethylene glycol (PEG) from Oxiteno agar from Oxoid and bacterial cellulose obtained by alcoholic fermentation, as described in Table 1.

Table 1 – Hydrogel membranes formulations

Sample number	PVP K-90 (%)	PVP K-30 (%)	PEG 300 (%)	Agar (%)	Cellulose (%)
1	6	-	1.5	0.5	-
2	5	0.5	-	0.5	-
3	6	-	1.5	0.5	0.022
4	5	0.5	-	0.5	0.022
5	6	-	1.5	-	Film
6	5	0.5	-	-	Film

Weighted PVP and PEG added to water stayed overnight at room temperature. The addition and dissolution of agar was in a boiling solution and after it was added 0.022% of triturated cellulose in samples 3 and 4. The prepared formulations were cooled to about 45°C and poured into 60mm Petri dishes to prepare membranes. Samples 5 and 6 received the hydrogel formulations over a cellulose film. After that the samples were wrapped and sent to irradiation in a gamma rays Co-60 source at 25kGy dose.

Results and Discussion

The obtained EDS spectrum in Fig.1 shows the main element of the BC and these elements were identified as hydrogen and carbon.

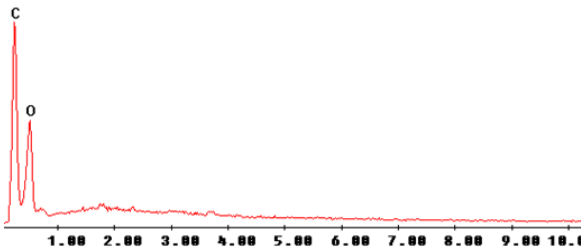

Figure 1. Microscopy EDS of the bacterial cellulose

XRD measurements were carried out in order to verify the cellulose phases present in the samples. Generally, native cellulose is composed by two allomorphs, a mixture of I_α and I_β phases, which ratio depends on the specie and source. For instance, I_α structure is prevalent in cellulose produced by algae and bacteria whereas I_β is dominant in cellulose obtained from woods [12-13]. Figure 2 shows the XRD patterns of commercial (Sigma Aldrich, 20µ) and bacterial celluloses together with the XRD patterns of native cellulose (PDF n° 50-2241). As it can be observed, only native cellulose was obtained. Broad diffraction peaks at 15 and 22.5° are assigned to the characteristic interplane distances of cellulose I_α and I_β phases ($100_{I\alpha}$, $110_{I\beta}$ and $010_{I\beta}$ planes at 15° and $110_{I\alpha}$ and $200_{I\beta}$ at 22.5°). The XRD of commercial wood cellulose presents a diffraction peak located at 34,5° which is ascribed to I_β phase [14]. The low intensity of this peak observed in the bacterial cellulose obtained in this work indicated that it is mainly composed by I_α native cellulose. These results are in agreement with Horii et al [15], who reported a native cellulose produced by *Acetobacter xylinum* composed by 64% of I_α and 36% of I_β phases.

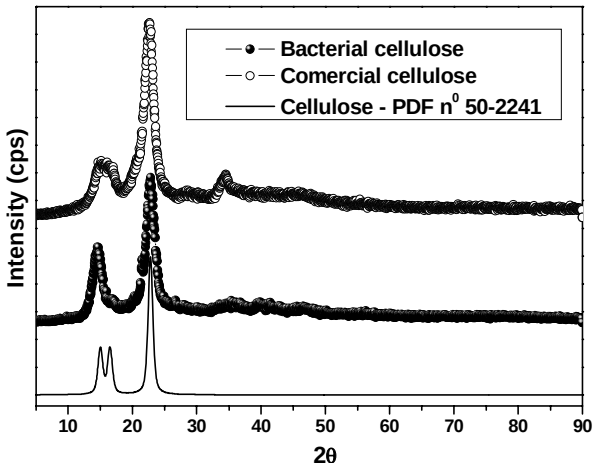

Figure 2. XRD patterns of commercial (-o-) and bacterial (-•-) celluloses together with the XRD patterns of native cellulose (PDF n° 50-2241).

Bacterial cellulose film samples analyzed by SEM were broken after frozen. The purpose of this analysis was to observe the heterogeneity of the surface texture. It can be observed in the Fig. 3 the presence of microfibrils in the bacterial cellulose film sample.

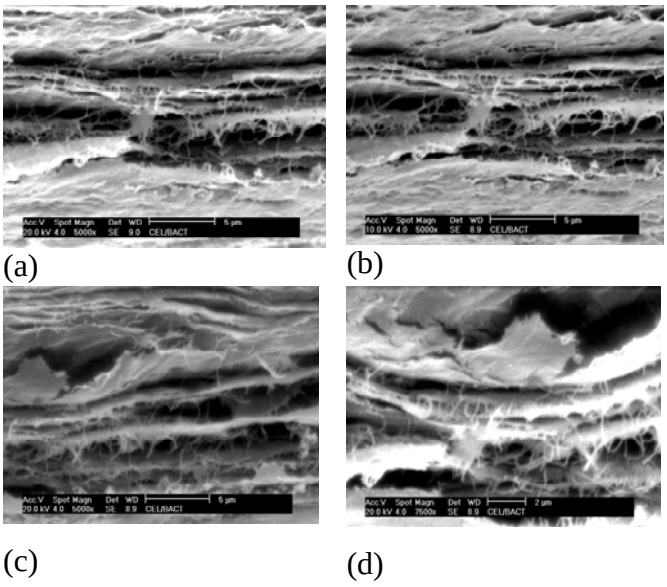

Figure 3. SEM of bacterial cellulose. The sample was broken after frozen. Amplitude of 5000x (a, b and c) and 7000x (d)

The microscopy of bacterial cellulose showed presence of microfibrils.

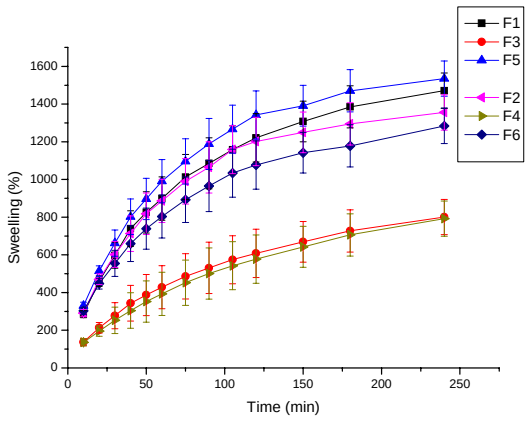

Figure 4. Sweelling curves of PVP hydrogels. F1 and F2 = PVP hydrogel; F3 and F4 = PVP hydrogel + triturated BC: F5 and F6 = PVP hydrogel + BC film.

All hydrogel formulations showed the same behavior (Fig. 4)with 100% capacity of water absorption within 50 min. The results of hydrogels formulated with fiber of bacterial cellulose (F3 and F4), showed a clear decrease, about 50%, as compared with PVP hydrogel and PVP plus cellulose film hydrogel.

Thermal analysis (TGA) is important to know the temperature where the substances start to degrade. The thermogravimetric data indicate the number of period of training of thermal collapse, loss of mass of the material in each period of training, linear of temperature. The initial and final degradation temperatures of bacterial cellulose were about 2 and 6% greater than commercial cellulose.

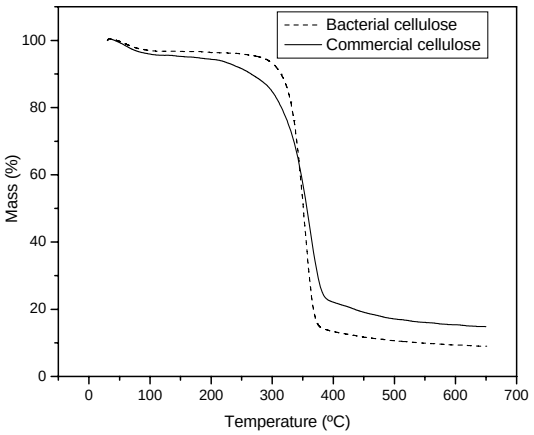

Figure 5. Termogravimetric curves of commercial and bacterial celluloses.

The TGA results showed that the initial temperature of degradation for commercial cellulose sample was 316.14°C, final of 345.89°C and a mass loss of 0.9040mg. For bacterial cellulose the initial temperature was 322.67°C and final 371.44°C and 1.6053mg of mass loss.

Conclusions

Bacterial cellulose was analyzed by thermal analysis (TGA) and by X-ray diffraction and compared with commercial wood cellulose. From the TGA curve it was possible to see the purity of the cellulose as compared with standard one.

The utilization of bacterial cellulose do not compromised swelling capacity of hydrogel membranes and we can conclude that cellulose fiber act as a physical entanglement increasing the crosslinking density of those hydrogels.

Aknowledgements

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