SYNTHESIS OF NEW HYDROGELS BASED ON XANTHAN AND CELLULOSE ALLOMORPHS

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The large availability of cellulose in nature and the low cost of cellulose derivatives make the cellulose-based hydrogels particularly attractive. New hydrogels were obtained by chemical cross-linking of different cellulose allomorphs (cellulose I, cellulose II and cellulose III) and xanthan with epichlorohydrin. The preparation conditions of the transparent cellulose hydrogels were established. The equilibrium swelling capacity of the obtained hydrogels was investigated at 37 °C, in distilled water. The obtained values were discussed in correlation with the dehydration heats estimated from the differential scanning calorimetry (DSC) curves. The morphology of hydrogels was studied by optical microscopy. The present work provides a simple and fast method for preparing eco-friendly hydrogels.

Keywords: hydrogels, cellulose allomorphs, xanthan, dissolution, swelling, DSC, optical microscopy

INTRODUCTION

Hydrogels are defined as three-dimensional networks of hydrophilic polymers, which can absorb and retain a significant amount of water. To obtain such networks, chemical cross-linking,[1] physical entanglement,[2] ionic bonds[3] and hydrogen bonds[4] are used. The hydrogel properties mainly depend on the cross-linking degree, on the chemical composition of the polymeric chains, and on the interaction between the network and the surrounding liquids.[5] Due to their high water content, the hydrogels possess a flexibility degree very similar to that of the natural tissues. The physical properties of hydrogels make them attractive for a variety of biomedical and pharmaceutical applications. Thus, they can be used in ophthalmology, tissue engineering, urology, plastic surgery, orthopedics, as well as in pharmaceutical and biotechnological fields.[6-9] Hydrogels have been of great interest for controlled drug release, due to their excellent biocompatibility, hydrophilicity and flexibility in tailoring the physico-chemical properties.

The hydrogels from natural polymers are promising for applications in the biomaterial field, due to their unique advantages, such as abundance, non-toxicity, biocompatibility, biodegradability and biological functions.[10]

Cellulose is the world's most abundant natural, renewable and biodegradable polymer. The crystalline structure of cellulose is highly ordered, with extensive hydrogen bondings among the molecular chains. Cellulose can crystallize into several different polymorphs.[11] Native cellulose (cellulose I) is the form found in nature, occurring[12] in two allomorphs, Iα and Iβ. Cellulose II is the crystalline form that emerges after regeneration from different media or after mercerization with an aqueous sodium hydroxide.[13] Celluloses III_I and III_{II} are obtained from cellulose I and II, respectively, by liquid ammonia or organic amine treatments.[14] The advantages and limitations of the structural architectures of the cellulose allomorphs are reflected on their accessibility. Starting from cellulose substrates with different supramolecular structures and, implicitly, different reactivities, a large and interesting area of utilization has been opened.[15]

Similarly to other polysaccharides, cellulose has a long history in medical applications, essentially due to its lack of toxicity (the monomer residues are part of the metabolites found in the human body), water solubility or high swelling ability, and stability to temperature and pH variations.[16] The excellent biocompatibility of cellulose

and of its derivatives has prompted the large use of cellulose-based devices in biomedical applications.[17] With regard to *in vivo* applications, it is worth reminding that cellulose is a biodurable material.[18]

Xanthan is a high molecular mass extracellular hetero-polysaccharide with cellulose as a backbone. This is one of the most extensively investigated polysaccharides with respect to biocompatibility, stability and safety.[19] Xanthan has been used in combination with other polymers, like gellan – for immobilizing cells, alginate – for encapsulation of urease enzyme, and chitosan – for immobilizing xylanase.[20]

This work is focused on developing new cross-linked cellulose–xanthan hydrogels by using epichlorohydrin (ECH) as a cross-linker. As a cellulosic material, the three polymorphic forms of cellulose (cellulose I, II and III) were used. The swelling behavior in water was investigated by measuring the swelling degree at equilibrium, while the dehydrating heat of the hydrogels conditioned at 65% was estimated by DSC. The morphology of hydrogels, studied by optical microscopy, was compared with that of pure cellulose and xanthan.

EXPERIMENTAL

Materials

Microcrystalline cellulose, Avicel PH-101, was purchased from Fluka, as well as xanthan, Mw > 1,000,000. Epichlorohydrin (ECH), of analytical grade, was used without further purification. Three cellulose allomorphs were prepared, namely: *cellulose I* – microcrystalline cellulose; *cellulose II* – mercerized cellulose with the crystalline form of cellulose II was prepared from cellulose I, by soaking it in 17.5% NaOH for 24 h, at 15 ºC, followed by thorough washing with distilled water and drying in air; *cellulose III* – samples with the crystalline form of cellulose III were prepared from cellulose I, by soaking in organic amine (100% ethylenediamine) for 24 h, at room temperature. The cellulose amine complex was washed with anhydrous methanol and finally the cellulose III samples were air-dried.

Dissolution of cellulose samples in aqueous NaOH solutions

Dissolution of microcrystalline cellulose was realized in a 8.5% NaOH solution, at -30 °C, for 24 h. The frozen solid was then thawed at room temperature.[21]

Preparation of hydrogels

Cellulose I hydrogel (CI) – 0.5 g microcrystalline cellulose was dissolved in an alkaline solution by freezing at low temperature (-30 °C). After thawing, 2.5 g epichlorohydrin were added to the clear cellulose solution, under continuous stirring. The obtained composition was transferred onto two glass plates and the cross-linking reaction was allowed to proceed for 6 h at 80 °C. The hydrogels were washed with water and acetone, after which the samples were dried in vacuum, at room temperature.

Cellulose II hydrogel (CII) and *cellulose III hydrogel* (CIII) were prepared under the same conditions as the CI hydrogel, cellulose II and cellulose III being used in their composition.

Xanthan hydrogel (X) – 0.5 g xanthan was swollen in an alkaline solution and epichlorohydrin was added under continuous stirring. The obtained composition was cross-linked at 80 °C, for 6 h. The hydrogels were washed with water and acetone, then dried in vacuum, at room temperature.

Cellulose I – xanthan hydrogel (CIX) – microcrystalline cellulose was dissolved at low temperature (-30 °C) in a NaOH solution. Xanthan was added to the obtained cellulose solution, in different ratios (Table 1). The cross-linking reaction was allowed to proceed in the presence of 2.5 g ECH at 80 °C, for 6 h. The hydrogels were washed with water and acetone and dried in vacuum, at room temperature.

Cellulose II – xanthan hydrogel (CIIX) and *cellulose III – xanthan hydrogel* (CIIIX) – were prepared under the same conditions as the CXI hydrogel, only cellulose II and cellulose III, respectively, being used in their composition.

Methods

The ***swelling degree of the hydrogels*** in distilled water was gravimetrically determined with the following relation:

$$Qmax = [(m - m_0)/m_0] \cdot 100\ (\%) \quad (1)$$

where: m_0 – dried hydrogel mass; m – swollen hydrogel mass.

Optical microscopy

The hydrogels have been examined with an IOR MC1 optical microscope in ordinary light, at room temperature and 4x and 10x magnification.

Differential scanning calorimetry (DSC)

The DSC curves of celluloses were recorded on a Mettler DSC 12E, at a heating rate of 5 °C/min. The samples were conditioned in desiccators at constant relative humidity of 65% and a temperature of 25 °C, until constant mass.

RESULTS AND DISCUSSION

In this study, new transparent hydrogels based on different cellulose allomorphs, such as cellulose I, II and III and xanthan, were prepared. Two-step synthesis consists of cellulose dissolution in a NaOH aqueous

solution and its mixing with xanthan, followed by cross-linking with ECH as a cross-linker. ECH, a convenient base-catalyzed cross-linking agent, has been widely used for the cross-linking of carbohydrates in polysaccharide chemistry.[22-26]

Swelling properties of hydrogels

The first research direction was to establish the influence of hydrogel composition on their swelling properties. Thus, different ratios of cellulose and xanthan were mixed and the quantity of cellulose was increased from hydrogel CX1 to CX5, as presented in Table 2. One may observe that the swelling capacity decreases with the increase in the cellulose amount in the hydrogels, the maximum swelling degree being achieved for the CX1 hydrogels prepared from 25% cellulose and 75% xanthan. An explanation for the decrease of the swelling degree with the increase in the cellulose content could be the physical cross-linking of cellulose in an alkaline solution.[27] Consequently, entanglements of the cellulose chains through hydrogen bonds could occur easily in solutions of high cellulose concentration; at a high temperature, irreversible gelation of the cellulose-NaOH solution occurs, leading to the decrease of swelling.

Another direction of investigation followed the influence of the allomorphic forms of cellulose on the swelling degree of hydrogels. Thus, for each composition, three kinds of allomorphic forms of cellulose were used for hydrogel preparation, namely cellulose I, II and III. The obtained values show that the utilization of cellulose II led to the highest value of the swelling degree, of 2.146%, which demonstrates the superabsorbant character of this hydrogel type. The swelling capacity of hydrogels decreases when using cellulose III (1.639%), followed by the one into which cellulose I has been incorporated (1.743%). These differences are determined by the structural modification suffered during the chemical treatments performed for obtaining cellulose allomorphs.

The experimental results proved that all CX hydrogels (CXI, CXII and CXIII) exhibited high values of the swelling degree, which could be modified by changing the components ratio, which is especially important for different applications in the biomedical field.

Table 1
Compositions and yield of hydrogels

Sample	Xanthan, wt%	Cellulose, wt%	η^a, wt%		
			Cellulose I	Cellulose II	Cellulose III
X	100	0		89.95	
CX1	75	25	87.50	84.52	91.08
CX2	66	33	90.26	89.76	94.62
CX3	50	50	95.26	94.88	97.78
CX4	33	66	97.98	97.26	97.98
CX5	25	75	99.88	99.46	99.86
C	0	100	99.90	99.52	99.90

[a] yield of hydrogel

Table 2
Maximum swelling degree of hydrogels

Sample	Qmax, %		
	Cellulose I	Cellulose II	Cellulose III
X		2278	
CX1	1639	2146	1743
CX2	1543	1708	1592
CX3	1284	1538	1370
CX4	1273	1519	1341
CX5	1254	1378	1309
C	1097	1236	1145

DSC study

For a further evaluation of cellulose-xanthan hydrogels, differential scanning calorimetry (DSC) was used. To establish the dependence of the dehydration heat on the composition of hydrogels, the samples were first conditioned for 72 h in desiccators, at a relative humidity of 65% and 25 °C, then the DSC curves were recorded over the 50-150 °C range. The structural peculiaritiers of hydrogels were evidenced by the presence of endothermic peaks with characteristic shapes for each composition.

Figure 1 plots the DSC curves for the hydrogels obtained from cellulose allomorphs (CI, CII and CIII). In the case of hydrogels based on different allomorphic forms of cellulose, a shift of the maximum temperature of the dehydration process (Tmax) to higher values was recorded, from CI hydrogel to CII and CIII, respectively (Table 3).

Table 3
Main parameters determined on the basis of DSC curves

Sample	Tmax,[a] °C	ΔH,[b] J/g
CI	57	372
CII	61	441
CIII	64	395
X	86	909
CIX1	70	584
CIX5	59	459
CIIX1	70	786
CIIX5	60	473

[a] maximum temperature of dehydration process;
[b] heat of dehydration

Thus, in the case of the CI hydrogel, the maximum temperature of the peak appears at 57 °C, followed by hydrogel CII at 61 °C, and by CIII at 64 °C. This behavior is influenced by the structural peculiarities of each cellulose allomorph present in hydrogels. Moreover, an increase of the endothermic peak from CI to CII was noticed, explained by the sorption of a higher amount of water. The dehydration heat was determined by measuring the areas of the endothermic peak corresponding to each sample. The DSC data of the samples conditioned at 65% humidity agree with the maximum sorption capacity, Qmax, determined by swelling in water at equilibrium, as shown in Figure 2; this indicates that differential scanning calorimetry (DSC) could be applied to evaluate the swelling properties of hydrogels.

In the case of the hydrogel obtained only from xanthan, the maximum temperature of the dehydration process appears at 86 °C, when the dehydration heat takes the highest value (909 J/g), as expected from the swelling data (Fig. 3).

For cellulose- and xanthan-based hydrogels, Tmax is situated between the values obtained for the CI and X hydrogels, which demonstrates the good compatibility of the components (Fig. 3). The dependence of the Tmax of the samples on hydrogel composition was also recorded. Thus, the temperature of the endothermic peak of CIX1 (75% X - 25% CI) is of 70 °C while, for the CIX5 hydrogel (75% CI - 25% X), it is of only 59 °C, as correlated with the xanthan content from the hydrogels, which is more stable to dehydration than cellulose. Also, the presence of xanthan in CX hydrogels leads to an increase in the dehydration heat, the values becoming higher by raising the xanthan amount in the composition of hydrogels.

Figure 4 evidences an increase of the area under the endothermic peak characteristic of CIIX hydrogels in comparison with CIX hydrogels, which also indicates an increase in the dehydration heat (Table 3). This observation demonstrates that the area of the endothermic peak due to the loss of absorbed water is directly related to the supramolecular structure of the cellulosic samples from hydrogel composition.

A good compatibility among components was also noticed for cellulose II-xanthan hydrogels, established on the basis of the maximum temperature values of the

dehydration process (Tmax). In terms of dehydration heat, it is observed that the hydrogels made with cellulose II have higher values than those made with cellulose I.

Optical microscopy analysis

The optical micrographs of cellulose I (CI) and xanthan (X) hydrogels are presented in Figure 5. Cellulose hydrogels have a uniform aspect, with small folds (which appear clearer at a magnification of 10x), being probably due to a weak incorporation of ECH into the cellulose solution. The hydrogel obtained from xanthan evidences a more uniform aspect. In this case, small particles of swollen xanthan, incorporated in the polymeric matrix, may be observed. The hydrogels obtained from cellulose and xanthan are white, transparent, evidencing a uniform surface. The optical image of the hydrogel obtained from 50% cellulose I and 50% xanthan (CX3) reveals a homogeneous aspect, explained by a good incorporation of cellulose into the xanthan matrix.

The presence of small voids in the structure of hydrogels can explain the high values of the swelling degree.

The optical micrographs of hydrogels based on cellulose II and cellulose II-xanthan are presented in Figure 6.

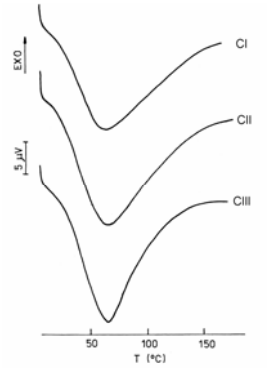


Figure 1: DSC curves recorded for hydrogels obtained from cellulose allomorphs (CI, CII and CIII)

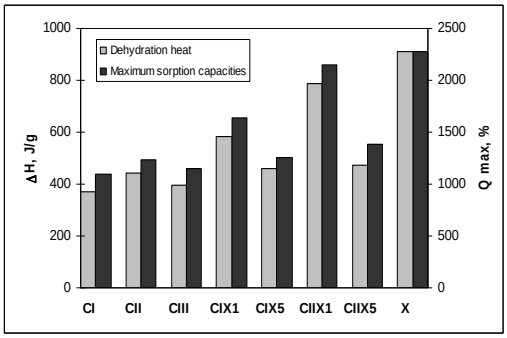


Figure 2: Comparative graphical representation of maximum sorption capacities, Qmax, and dehydration heat values, ΔH, estimated by DSC

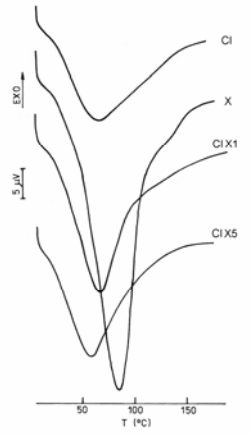


Figure 3: DSC curves of hydrogels obtained from xanthan (X), cellulose I (CI) and from xanthan-cellulose I (CIX)

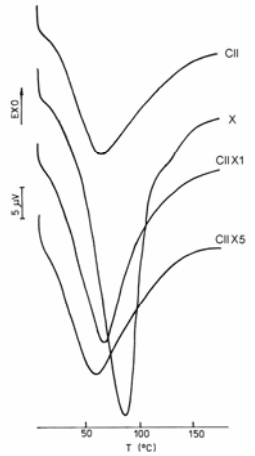
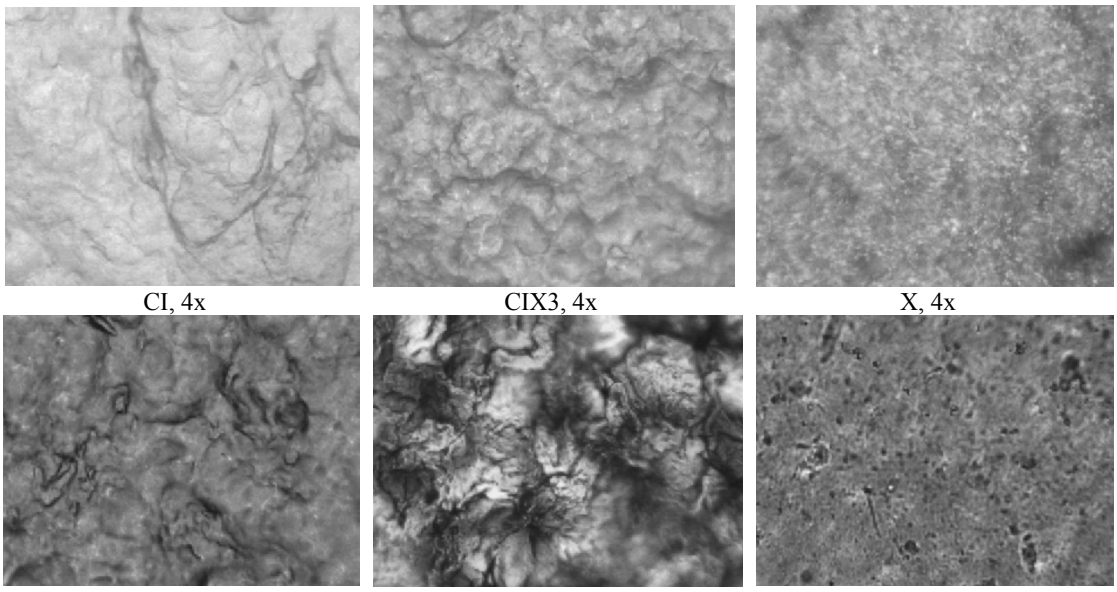


Figure 4: DSC curves of hydrogels obtained from xanthan (X), cellulose II (CII) and from xanthan-cellulose II (CIIX)

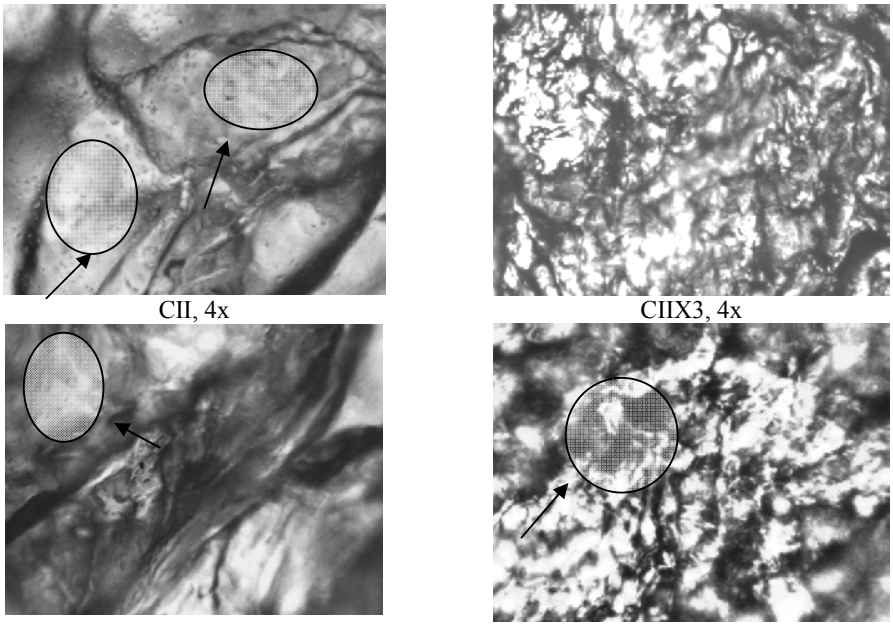


CI, 4x CIX3, 4x X, 4x

CI, 10x CIX3, 10x X, 10x

(a) (b) (c)

Figure 5: Optical micrographs for cellulose I hydrogel (a), cellulose I-xanthan hydrogels (b) and xanthan hydrogels (c), at a magnification of 4x and 10x



CII, 4x CIIX3, 4x

CII, 10x CIIX3, 10x

(a) (b)

Figure 6: Optical micrographs for cellulose II hydrogel (a) and cellulose II-xanthan hydrogels (b), at a magnification of 4x and 10x

While the CII hydrogels evidence large areas with homogeneous structure and small pores, in the case of CIIX3 hydrogels, the size of pores increases, leading to a more open and looser structure and, moreover, to a higher swelling ratio.

CONCLUSIONS

The treatment of cellulose in an alkaline solution, at a low temperature, permits to obtain clear, transparent hydrogels with increased swelling degrees. New polymeric systems have been obtained from different allomorphic forms of cellulose and xanthan. The swelling degree of cellulose-xanthan hydrogels depends on their composition. When introducing different allomorphic forms of cellulose (cellulose I, II and III) in the composition of hydrogels, cellulose II leads to superabsorbant hydrogels. The effect

of the structural features of the studied hydrogels on their dehydration heat was evaluated by differential scanning calorimetry (DSC). The optical micrographs reveal the morphological aspects of the obtained hydrogels.

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