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NOVEL HYDROGELS MODIFIED WITH XANTHAN GUM – SYNTHESIS AND CHARACTERIZATION

INNOWACYJNE MATERIAŁY HYDROŻELOWE MODYFIKOWANE GUMĄ KSANTANOWĄ – SYNTEZA I CHARAKTERYSTYKA

Abstract

Due to their interesting features, hydrogels are attracting growing interest in the polymer materials market. Therefore, many studies are currently conducted to characterize these materials and to modify them in order to increase the range of their potential use. In the presented article, hydrogels based on acrylic acid and chitosan and modified with xanthan gum were obtained by photopolymerization. Their swelling ability and behaviour in solutions that simulate fluids in the human body were determined. The effect of incubation in various fluids on the chemical structure of the synthesized materials was characterized using spectroscopic analysis. Furthermore, the surface morphology of the attained materials was characterized with scanning electron microscopy (SEM).

Keywords: hydrogels, xanthan gum, acrylic acid, chitosan, surface morphology of hydrogels

Streszczenie

Hydrożele z uwagi na swoje interesujące właściwości należą do grupy związków cieszącej się dużym zainteresowaniem na rynku materiałów polimerowych. Dlatego też prowadzone są badania mające na celu charakterystykę tych materiałów oraz ich modyfikację w celu zwiększenia możliwości ich potencjalnego zastosowania. W artykule przedstawiono syntezę hydrożeli na bazie kwasu akrylowego i chitozanu modyfikowanych gumą ksantanową. W toku badań określono zdolności pęcznienia hydrożeli oraz ich zachowanie w symulowanych płynach ustrojowych. Ponadto określono wpływ inkubacji otrzymanych materiałów w wybranych płynach na ich strukturę chemiczną z wykorzystaniem analizy spektroskopowej. Dodatkowo scharakteryzowano morfologię powierzchni hydrożeli za pomocą skaningowego mikroskopu elektronowego (SEM).

Słowa kluczowe: hydrożele, guma ksantanowa, kwas akrylowy, chitozan, morfologia powierzchni hydrożeli

1. Introduction

In recent times a big rise in the popularity of polymers has been observed. The materials which are attracting the greatest interest are undoubtedly hydrogels (also known as superabsorbent polymers, SAPs). They consist of a three-dimensional network of polymer chains. This structure contains hydrophilic groups and, due to their presence, these materials are characterized by a strong ability to absorb liquids. This property depends on features of the hydrogels (such as crosslinking degree or type of additives introduced into the polymer matrix). However, the properties of the liquid that is absorbed (pH, type of ions in the solution) can also have an impact on the sorption capacity of the tested material [1–4].

What is more, hydrogels are also characterized by their biocompatibility, biostability and non-toxicity. Moreover, these materials are flexible and at the same strong. The hydrogel matrix can be enriched with different substances of natural origin, thus giving the hydrogel material additional properties. All these characteristics make hydrogels suitable for use in many fields such as medicine or pharmacy. Superabsorbent polymers are used in controlled drug release systems [5–6]. Hydrogels are also the raw material for the preparation of modern dressings that provide a favourable environment that is conducive to the wound healing process [7]. Another field in which these materials are applied is tissue engineering, where hydrogels find application not only in the preparation of artificial organs or implants, but also as three-dimensional scaffolds for cell growth and proliferation [8].

Xanthan gum, which acts as a modifying agent of the polymer matrix, is a well-known polysaccharide. It is characterized by unusual rheological properties that make it applicable in many fields such as the food industry [9–13], agriculture, textiles or even in pharmacy or tissue engineering [14]. Furthermore, xanthan gum plays an important role in controlled drug delivery systems [15–16]. In many previous studies, xanthan gum was used as a substance constituting a hydrogel matrix. Tao et al. [17] described the synthesis and physicochemical properties of hydrogels based on this polysaccharide. Such materials have been proposed as polymers for the controlled release of various substances in the food industry and tissue engineering. A three-dimensional network of polymer chains of xanthan gum was obtained by the application of sodium trimetaphosphate as the crosslinking agent. Various compounds can be used as crosslinkers in the preparation of hydrogels based on this polysaccharide, and the obtained polymers are characterized by a variety of properties [18–19]. An interesting solution is the application of hydrogels based on both xanthan gum and magnetic nanoparticles for adsorption of different dyes from aqueous solutions [20–21]. It is also worth noting that hydrogels based on the described polysaccharide and methylcellulose [22] or starch [23] represent an excellent material for drug delivery. Xanthan gum is also an interesting component of nanofibers [24] or hydrogel films [25].

In the framework of the research, novel hydrogels based on acrylic acid and modified with xanthan gum and chitosan are proposed. The materials used as additives in the polymer matrix are of natural origin and therefore are designed in order to provide the attained polymers with characteristics such as biodegradability or biocompatibility. What is more, such properties are desirable in materials destined for biomedical applications.

2. Materials

The chitosan (low molecular weight), acrylic acid (anhydrous, colourless liquid, 99%, $d = 1.051$ g/ml), poly(ethylene glycol) diacrylate ($M_n = 256$ g/mol), $d = 1.12$ g/ml), phenylbis(2,4,6-trimethylbenzoyl)phosphine oxide (colourless liquid, powder, $M = 418.46$ g/mol) and xanthan gum used for preparation of the hydrogels were supplied by Sigma Aldrich; potassium hydroxide was received from Avantor Performance Materials Poland (formerly POCH SA). All compounds were characterized by analytical purity and applied without further treatment.

3. Experimental Part

The preparation of hydrogels containing xanthan gum involves several steps. In the first of them, 45 ml of acrylic acid was neutralized by the addition of 50 ml of KOH (40% solution). This reaction is exothermic, therefore the mixture had to be cooled to room temperature. The next step was to prepare the chitosan solution by dissolving 10 g of it in 200 ml of 2% acrylic acid solution. 5 ml of the prepared solution was added to the mixture of acrylic acid and KOH. Then, an appropriate amount of xanthan gum, 1 ml of initiator solution (made up by adding 2 g of phenylbis(2,4,6-trimethylbenzoyl)phosphine oxide to 10 ml of acrylic acid) and 1 ml of crosslinker (poly(ethylene glycol) diacrylate ($M_n = 256$ g/mol)) were introduced into the previously prepared mixture, which was poured onto a Petri dish and subjected to UV radiation for 1 min. An Emita VP-60 quartz lamp (120 W power, applied wavelength $\lambda = 320$ nm) was used for the process of photopolymerization. In this manner, a series of hydrogels containing different amounts of xanthan gum was obtained. The following quantities of xanthan gum were added (based on the amount of acrylic acid):

- a) 0 wt%
- b) 1 wt% (0.075 g)
- c) 3 wt% (0.224 g)
- d) 5 wt% (0.373 g)
- e) 7 wt% (0.522g).

4. Methods

4.1. Swelling measurements

A 1.0 g hydrogel sample was immersed in 100 ml of liquid (distilled water, solution of salts 0.9% NaCl, 0.9% MgCl₂; Ringer's liquid and artificial saliva solution) for 1 h. After this time the hydrogel was taken out of the solution and weighed. The swelling ratio (Q , g/g) was calculated from the following formula (1):

$$Q = \frac{w - w_0}{w_0} \quad (1)$$

where:

- w – weight of swollen sample,
- w_0 – weight of hydrogel before swelling.

4.2. Incubation studies

In order to determine the behaviour of the obtained materials in selected fluids, samples of hydrogels were introduced into 100 ml of a suitable liquid (i.e. distilled water, Ringer's liquid and a solution of artificial saliva). The incubation time in distilled water and Ringer's liquid was 63 days; in the solution of artificial saliva the hydrogels were immersed for 21 days. The pH of the solutions was measured once a week.

4.3. Fourier Transform Infrared spectroscopy (FT-IR)

FT-IR spectroscopy was used in order to analyse the chemical structure of the obtained hydrogel polymers. The test was performed using an FT-IR spectrometer with ATR diamond/ZnSe (Perkin Elmer Spectrum 65). In order to determine the impact of immersion in suitable liquids on the chemical structure of the tested hydrogels, materials were investigated before and after the incubation period.

4.4. Scanning electron microscopy (SEM)

Surface morphology of the tested samples was determined by means of an FE-SEM Zeiss Supra 55VP scanning electron microscope. This method was used in order to characterize the surfaces of the hydrogels and to define the impact of the presence of the additive on the morphology of the materials.

5. Results and discussion

5.1. Swelling measurements

The calculated values of the swelling ratios of hydrogels containing different amounts of xanthan gum are presented in Figure 1 and Figure 2.

The attained hydrogels containing xanthan gum were characterized by a relatively low swelling capacity. The highest swelling ratios were found for the samples placed in distilled water. Under the influence of water, the hydrophilic functional groups present in the polymer chains dissociate and ions are then formed. As a result, electrostatic interactions between these ions occur. Ions with the same charge repel each other, which consequently increases the distance between the polymer chains. As a result, there is more free space for water, therefore hydrogel is able to absorb large quantities of liquid. In the case of 0.9% NaCl, lower values of Q were observed due to the presence of ions in the solution that is absorbed. An

ion interaction between the polymer matrix and the sodium cations takes place that reduces the hydrophilic nature of the carboxyl group (Na^+ joins the aforementioned group) and, as a result, the interactions between the COO^- ions that were previously observed are limited. The presence of divalent ions in 0.9% MgCl_2 contributes to the increase of the crosslinking degree of the tested hydrogel. As a result, a reduction of swelling ability is observed.

Significantly, the tested hydrogels are able to absorb salts. Thus, these materials can be used as alternative dressings which can absorb wound exudates and impurities, thereby contributing to disinfection.

Relatively high swelling ratios for the samples immersed in the solution of artificial saliva were observed. The swelling ability of the samples tested in artificial saliva was lower than those tested in distilled water but higher in comparison to the swelling ability determined in the other tested liquids. The solution of artificial saliva contains carbonate and orthophosphate ions that provide its buffering properties but do not reduce its sorption capacity. Lower values of Q were observed for samples tested in Ringer's solution. There are a lot of monovalent and divalent ions in this mixture, which reduces its absorption properties.

Based on the results of swelling in both the tested solutions, it can be also said that the presence of xanthan gum in the polymer matrix has an impact on the swelling properties of the tested materials. It is not easy to find any dependency between the amount of the introduced additive and the sorption capacity of the modified samples. However, in most cases the modified hydrogels were characterized by higher swelling capacity. This could be the result of the presence in xanthan gum of compounds that contain hydrophilic functional groups in their structure. Such groups can form additional hydrogen bonds with the molecules of the solvent and therefore the sorption capacity of samples modified with this additive can be higher. In other cases, a certain amount of this additive can cause a kind of system disorder or even accumulate between the polymer chains in a way that reduces swelling ability. Moreover, the synthesized hydrogels have a low sorption capacity that is important from the point of view of the potential applications of these materials.

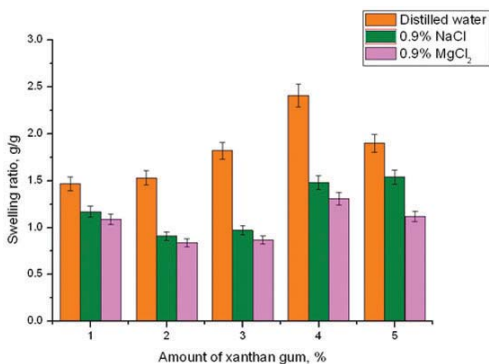


Fig. 1. Results of swelling in distilled water, 0.9% NaCl and 0.9% MgCl_2 (number of repetitions $n = 3$)

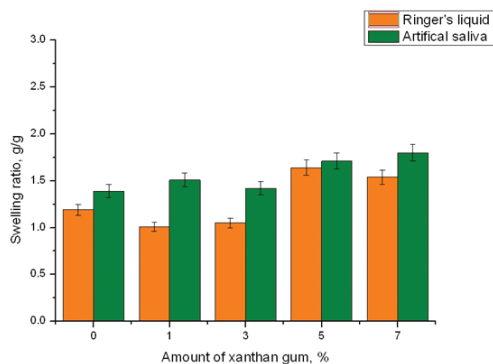


Fig. 2. Results of swelling in Ringer's liquid and in artificial saliva solution (number of repetitions $n = 3$)

5.2. Incubation studies

The pH values of the solutions determined once a week during the 63 days of incubation are shown below in Figures 3–5.

Sudden changes in pH values were not observed during the incubation; this indicates the compatibility of the tested material with a given environment. Therefore, these hydrogel matrices can be considered as materials that are useful in medicine. The pH value fluctuates around neutral, which promotes the process of wound healing. Importantly, the form and shape of the hydrogel did not change during incubation in distilled water, i.e. absorption of water did not cause the disintegration of the material.

The highest values of pH were observed in the case of the liquid containing hydrogel without xanthan gum. When immersed in Ringer's liquid, the pH of the hydrogels modified with this polysaccharide show a slight tendency to change. This could attest not only to their in vitro stability, but also to the compatibility of these materials with bodily fluids (Ringer's liquid is isotonic with human blood). These features allow the use of these hydrogels as biomedical materials.

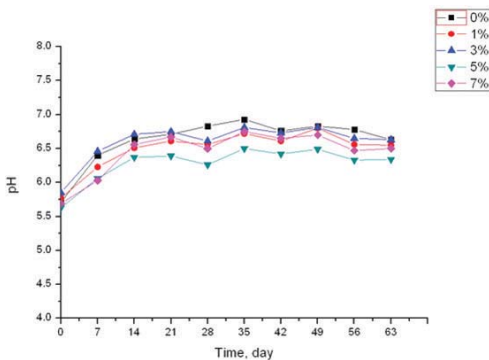


Fig. 3. Changes in pH of distilled water containing hydrogels

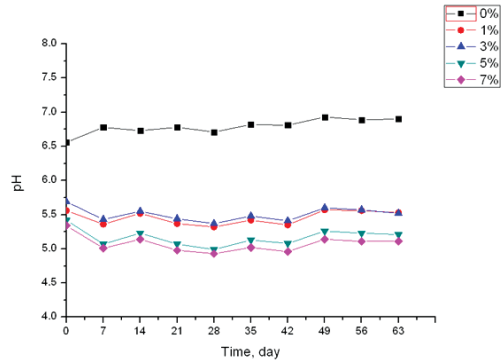


Fig. 4. Changes in pH of Ringer's liquid containing hydrogels

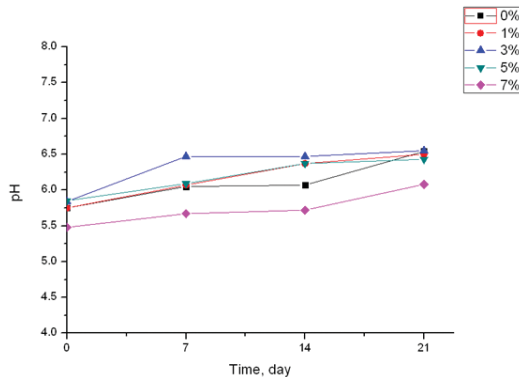


Fig. 5. Changes in the pH of the solution of artificial saliva containing hydrogels

The results do not show any relationship between the pH of the liquid in which the sample is immersed and the amount of the xanthan gum in the hydrogel matrix. The lowest pH values were observed for a sample containing the biggest amount of the aforementioned polysaccharide. Additionally, in each case a gradual increase in pH during incubation was observed that is a result of the ion exchange that takes place between the polymer matrix and the ions in the Ringer's liquid. It should also be mentioned that the carbonate and orthophosphate anions in the saliva which are responsible for its buffering properties cause the whole system to tend to reach a pH of approx. 6.8–7.

5.3. Results of FT-IR spectroscopy

The IR spectra of attained hydrogels before and after immersion in particular fluids are presented in Figures 6–9. This study makes it possible to determine the effect of incubation on the chemical structure of polymer samples.

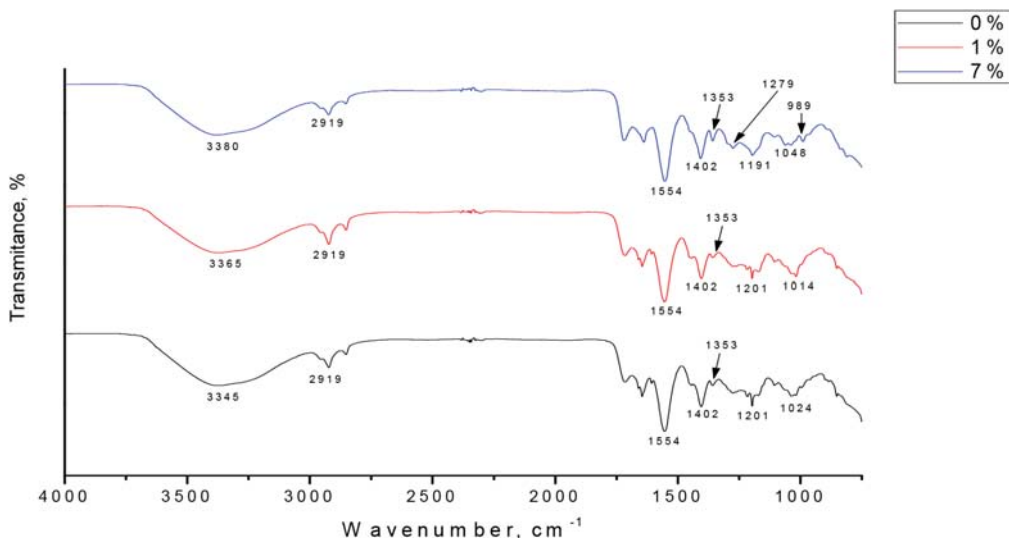


Fig. 6. IR spectra of samples modified with xanthan gum before immersion

The IR spectra of the hydrogel samples prior to the incubation show considerable similarity. Table 1 shows a compilation of the vibrations occurring in the studied materials.

On the basis of the attained IR spectra, it can be stated that long-term immersion of the obtained materials in simulated body fluids has an impact on their chemical structure. According to the results of the spectroscopy, a decrease or total disappearance of the peaks characteristic of particular bonds is observed (e.g. the peaks at 1548 cm^{-1} or 1406 cm^{-1}).

Incubation in Ringer's liquid and in distilled water lasted 63 days; immersion of samples in a solution of artificial saliva lasted 21 days. Nevertheless, the greatest change was observed in case of incubation in the artificial saliva.

Table 1. Compilation of characteristic vibrations occurring in the tested samples

Region of vibration [cm^{-1}]	Assigned to
3300–3500	N-H
2850–2970	C-H
1550–1650	N-H
600–1500	C-C
1180–1360	C-N
1050–1300	C-O
675–995	C-H

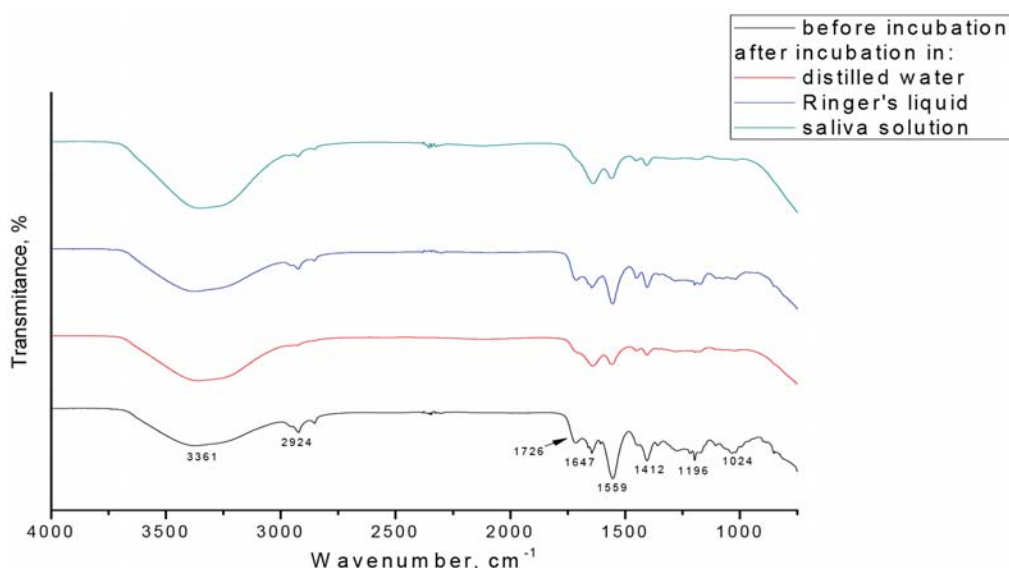


Fig. 7. IR spectra of samples without xanthan gum (0 wt%)

In Figure 6, the impact of the introduction of xanthan gum into the hydrogel matrix on the structure of the tested material can be observed. A lower intensity of the peak at a wavelength of approx. 1720 cm^{-1} derived from carboxylic acid can be seen that is due to the fact that with the increasing amount of the additive the total amount of the acid in relation to the amount of the rest of the reagents becomes lower. Furthermore, the disappearance of the band at 989 cm^{-1} (observed in the case of samples with 1%wt. and 7%wt. of xanthan gum) that is derived from the C-H group can be seen. This group becomes less visible as a result of the increasing amount of the additive.

Based on the obtained spectra, it was found that incubation in Ringer's liquid caused only a slight change in the structure of the material. Regardless of the incubation, vibrations characteristic of structures such as N-H ($3300\text{--}3500 \text{ cm}^{-1}$), N-H ($1550\text{--}1650 \text{ cm}^{-1}$), C-C ($600\text{--}1500 \text{ cm}^{-1}$) or the band corresponding to the carboxylate ion ($1554\text{--}1552 \text{ cm}^{-1}$) were observed. Also, the band characteristic of the -CH_2 group ($2925\text{--}2923 \text{ cm}^{-1}$) can be seen on

all obtained spectra of samples incubated in Ringer's liquid. Thus, it can be concluded that during incubation in Ringer's liquid, unreacted reactants were not completely eluted from the inside of the tested hydrogel. This may have been caused by the presence of divalent calcium ions in the Ringer's liquid, which contributes to the increase of the crosslinking density of the material hindering the elution of unreacted substances.

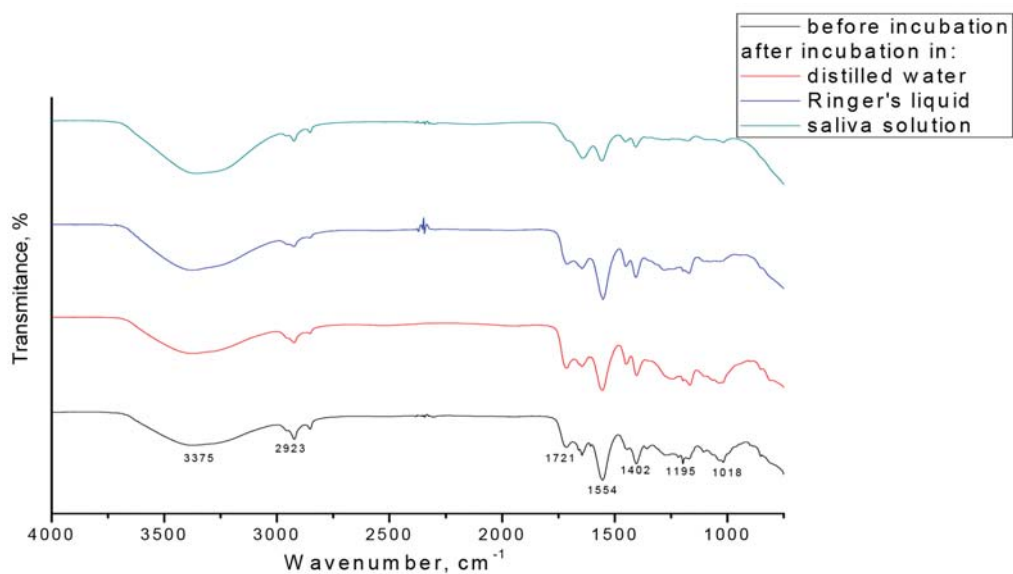


Fig. 8. IR spectra of samples with 1%wt. of xanthan gum

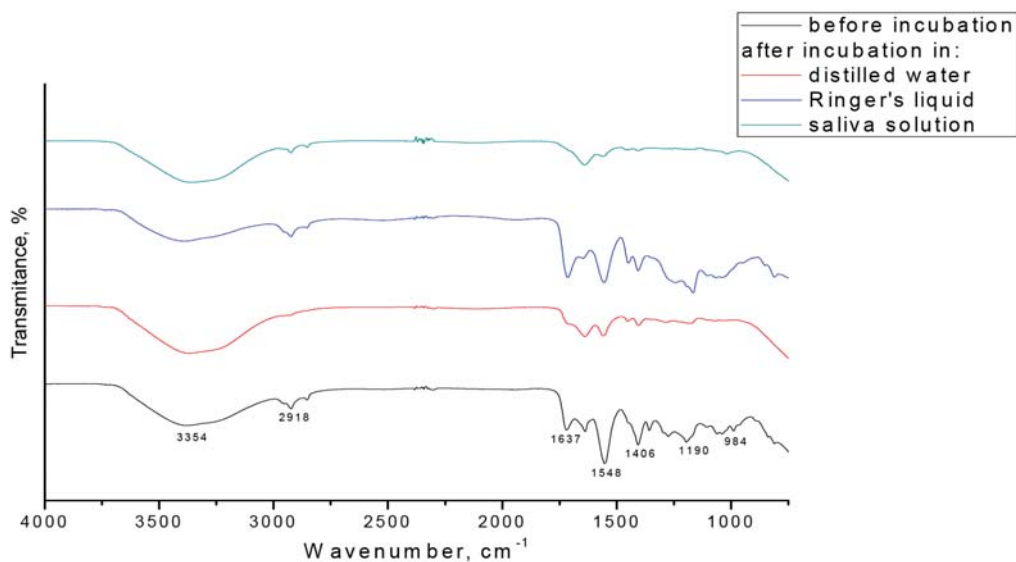


Fig. 9. IR spectra of samples with 7%wt. of xanthan gum

In the case of the spectra of the hydrogels after incubation in both distilled water and the solution of artificial saliva, there were no bands characteristic of the vibration of groups C-C and C-O. Vibrations of these bands were observed in the structure of hydrogels before incubation. Samples that were incubated in distilled water and in the solution of artificial saliva degraded to a greater extent. This applies both to the sample without additive, and of that containing 7%wt. xanthan gum in its matrix.

The sample containing 1%wt. of xanthan gum degraded the least. This is probably because this system is crosslinked most intensively, i.e. a suitable amount of xanthan gum improves the crosslinking of the tested structure, but this does not disrupt the system, as might happen in the case of 7%wt. of this additive.

5.4. Scanning electron microscopy (SEM)

In Figure 10, the surface of the synthesized hydrogels modified with xanthan gum is shown.

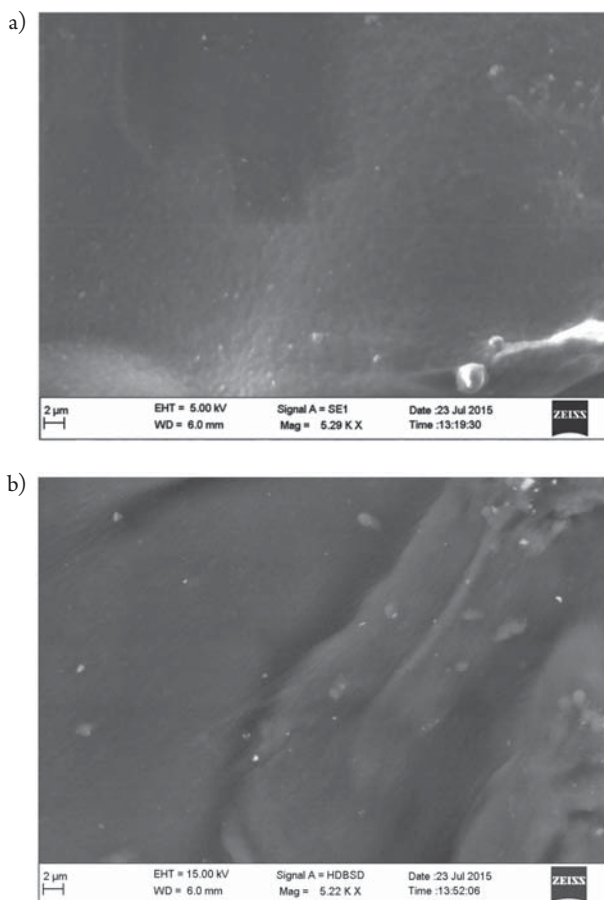


Fig. 10. SEM microphotographs of hydrogel: unmodified (a) and containing xanthan gum (b)

On the basis of the presented microphotographs, it can be seen that the surface of the unmodified hydrogel material is smooth and homogeneous. The surface morphology of the sample containing xanthan gum is similar to the surface morphology of the sample without xanthan gum. However, on the surface of the hydrogels modified with xanthan gum, some irregularities can be observed. The sample with the additive in its matrix is characterized by a slight porosity that could be the cause of the higher swelling ability of the modified hydrogels. These irregularities could cause a specific surface area of the tested materials and therefore this material has higher sorption capacity.

6. Conclusion

A series of hydrogels based on acrylic acid and chitosan and containing different amounts of xanthan gum in the polymer matrix was synthesized using UV radiation. The obtained materials were characterized by a very low swelling ability. Many factors have an impact on this property, including the composition of the absorbed liquid as well as the structure of the tested material. The highest swelling ratios were calculated in the case of distilled water due to the absence of ions, which cause the crosslinking density to increase and thereby reduce the swelling capacity. Also, it can be noticed that in some cases the presence of xanthan gum in the polymer matrix resulted in higher sorption capacity of the modified hydrogels. This could be the result of the slightly more heterogeneous and porous structure of the hydrogels containing xanthan gum, as can be observed in the SEM microphotographs. This causes a specific surface area and therefore the sample has higher sorption capacity.

The samples of the tested hydrogels did not cause any sudden pH changes of the selected fluid solutions in which they were submerged. This indicates the *in vitro* stability and biocompatibility of the synthesized hydrogels with body fluids. Also, on the basis of FT-IR spectroscopy, it was found that immersion of the hydrogel samples in distilled water and in artificial saliva solution causes the disappearance of bands specific to certain functional groups. Moreover, it was observed that the samples incubated in these solutions degraded to a greater extent.

To sum up, the features of the synthesized hydrogels modified with xanthan gum allow them to be considered for biomedical purposes. Therefore, in the near future more advanced research including further modifications or studies on cell lines are planned.

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