

INVESTIGATION OF HYDROGEL SHEETS BASED ON HIGHLY ESTERIFIED PECTIN

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Abstract. The report describes the features of physical and mechanical properties and absorption capacity of hydrogels based on highly esterified pectin. Experimental data showed the correlation between these values. Also, an attempt is made to explain the obtained dependencies via the hydrogel morphology and the mechanism of its formation.

Keywords: pectin, hydrogel, gel fraction, absorption capacity, plastic deformation

1. Introduction

Hydrogels based on synthetic polymers are widely used in various fields.¹⁻⁵ At the same time, there is a growing interest in products based on natural polysaccharides, which have the necessary properties and can be used for producing hydrogel materials.⁶⁻⁸

Pectin-based hydrogels are widely used in food industry, and gelation usually takes place due to the physical forces.^{9,10} Medical materials obtained using this biopolymer are also utilized.¹¹ In particular, hydrogels based on pectin are used as drug shells, when the assimilation occurs in the large intestine.^{12,13} According to some biomedical studies, pectins have properties possessing a beneficial effect on the healing process of skin lesions.^{14,15} However, there are few reports on the pectin hydrogels for wet wound healing,¹⁶⁻¹⁹ which is a progressive therapy method to date.²⁰⁻²² Pectin hydrogels as a means of wound healing should be affordable and have several properties, in particular, the ability to absorb sufficient fluid from the wound site.^{19,23}

It is confirmed that the degree of pectin esterification significantly affects the process of pectin hydrogels formation.¹² Thus, the production of hydrogels with satisfactory characteristics is observed in the case of using low-esterified pectins.^{13,24} However, pectins with a low content of esterified groups are less available and their separation from raw materials demands a specific technology. This leads to a significant increase in the cost of the resulting product.²⁵

This work reports on the features of physical and mechanical properties and absorption capacity of hydrogels based on highly esterified pectin.

2. Experimental

2.1. Materials

Citrus pectin (CF 131B, Grindsted, Czech Republic), sodium alginate (CAS number: 9005-38-3, Sigma-Aldrich, USA), calcium chloride (E-509, Klebrig, Netherlands), calcium gluconate (E-576, Klebrig, Netherlands) polypropylene glycol (PPG, Acros organics, Germany, MW 2000), polyethylene glycol (PEG, AppiChem GmbH, Germany, MW 3500-4500).

2.2. Methods of analysis

Determination of the content of free carboxyl groups in the pectin composition was performed by direct potentiometric titration according to standard technique.^{26,27} The hydrolysis of esterified pectin groups in an alkaline medium was performed to determine the total content of carboxyl and esterified groups. The obtained mixture was titrated according to.^{26,28} The total content of free and neutralized carboxyl groups was determined according to the method.^{26,29} The hydroxyl number of PEG and PPG was determined according to methods.^{26,30} Determination of calcium content in samples of pectin hydrogels was determined according to the method.³¹

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2.3. Formation of citrus pectin-based hydrogels

The following mixture was previously prepared to obtain pectin hydrogels: 5–10 % aqueous solution of pectin; 2–5 % aqueous solution of sodium alginate; 10–15 % aqueous solution of PEG; PPG was mixed in a ratio of 2: 1: 0.1: 0.1. The resulting mixture was loaded into molds to provide a solution height of 3–8 mm, depending on the purpose of the resulting hydrogels. Then, the molds were placed in a 1–2 % solution of a structuring agent (calcium chloride or gluconate) and maintained for at least 20 hours to evenly distribute the calcium ions. The resulting hydrogel was removed from the mold, washed with water, and stored at 278 K.

2.4. Production of pressed pectin sheets with different degrees of compression

Pectin hydrogels obtained using the above-described technique were pressed to different degrees of compression using a pressing machine Sky Rack SR-42130.³² The compression degree of pectin hydrogels was calculated by the equation:

$$\text{Compression degree} = \frac{h_0 - h}{h_0} \quad (1)$$

h_0 – the thickness of the pectin hydrogel, mm; h – the thickness of the pectin hydrogel after compression, mm

2.5. Determination of the gel fraction in hydrogel samples

0.5 ÷ 1 g of pectin hydrogel samples were placed in weighed nylon bags, which were immersed in distilled water at 323K for 15 hours, with constant stirring (water

was changed every 3 hours). After washing, the samples were dried at a temperature of 333K to constant weight. The gel fraction was considered to be the ratio of the mass of the dry residue in the bag to the total mass of the hydrogel sample.

2.6. Determination of the swelling degree of pectin hydrogels

The degree of swelling of pectin hydrogels was determined by a tea-bag method.^{33,34} The calculation of the degree of swelling α was performed according to the equation

$$\alpha (\%) = \frac{m_t - m_0}{m_0} \times 100. \quad (2)$$

m_t – the mass of the swollen hydrogel sample, g; m_0 – the mass of the dry polymer, g

3. Results and discussions

Pectin due to its therapeutic properties³⁵ is attractive for the obtaining of hydrogel dressings. It has been also reported earlier that the characteristics of pectin-based hydrogels improve with the introduction of natural polysaccharide sodium alginate.³⁵

In this work, hydrogel sheets were obtained using the citrus pectin with an esterified carboxyl group content of 81 %. Since the obtained hydrogels are planned to be used as dressings for wet wound healing, it is important to study their ability to absorb fluid. Table 1 shows the values of equilibrium (maximum, MWC) swelling of pectin hydrogels in comparison with the initial water content (IWC), which is obtained during the formation of hydrogel depending on the concentration of calcium ions in the solution used to structure the hydrogel composition.

Table 1. Degrees of swelling of hydrogels obtained using pectin (4 %) and calcium gluconate of different concentrations

Calcium gluconate concentration, %	Water content, g of water/ g of pectin	
	Initial water content, IWC, g/g	Maximum water content, MWC, g/g
2.92	23.7	25.7
3.87	24	26.5
4.85	24.1	28.6

The data in Table 1 shows that the MWC of pectin hydrogels does not significantly exceed IWC. There is a very weak dependence of MWC on the concentration of

calcium ions in the solution used at the stage of hydrogel formation. It should be noted that obtained pectin hydrogel has a significantly lower swelling ability than

polyacrylamide hydrogels. Polyacrylamide-based hydrogels formed with an initial water content of $5 \div 8$ g / g can swell to a degree of $140 \div 800$, depending on the structuring conditions.³

Thus, it can be argued that such pectin hydrogels are obtained with water content close to MWC values and cannot be used for wound therapy due to their insufficient ability to absorb fluid from the wound site. Probably, it is necessary to use factors affecting the structure and morphology of the sheets to obtain pectin hydrogels with increased swelling capacity.

A study of the properties of pectin-based hydrogel sheets with varying degrees of esterification suggests that pectin hydrogels obtained by structuring with calcium ions are elastic-viscous Maxwell relaxing bodies. At the fast loading of pectin hydrogel samples, their loading curves are similar in shape to the loading curves of elastic, for example, polyacrylamide, hydrogels (Fig. 1).

However, it should be noted that the obtained pectin hydrogels do not possess elastic deformations: small but long-term loads on the sample cause plastic (irreversible) deformations in the samples (Fig. 2). The external load applied to the sample leads to the formation of corresponding stress in the hydrogel, which in time decreases almost inversely proportional to the development of plastic deformation. Thus, relaxation processes are observed in the sample, which is accompanied by a decrease in its volume and, as a consequence, the release of fluid. In other words, hydrogel collapse caused by external mechanical influences is observed.

Fig. 3 shows the relaxation curves of the samples depending on their pectin content.

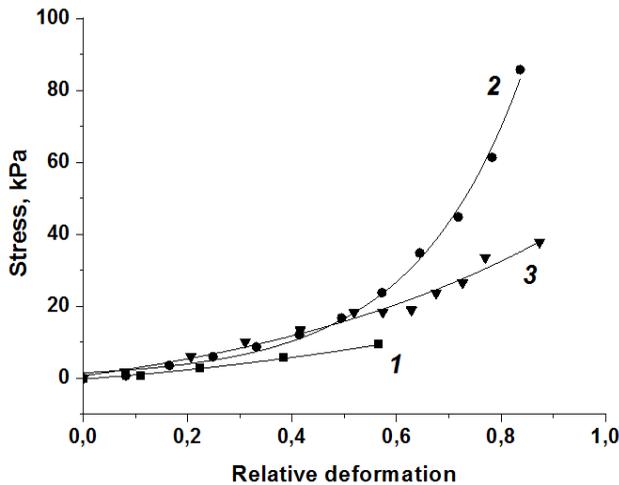


Fig. 1. Fast loading curves for pectin hydrogels samples with various pectin content. Pectin concentrations: 1–3 %, 2–4 %, 3–6 %

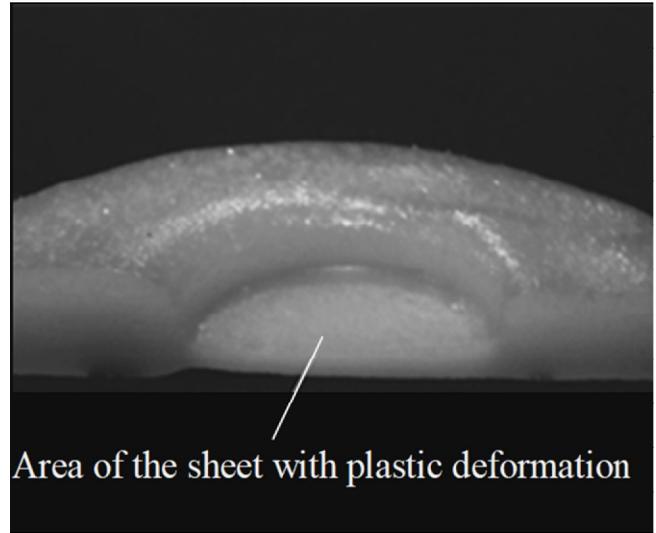


Fig. 2. Image of pressed pectin hydrogel, which illustrates the nature of plastic deformation at a compression degree of 0.65

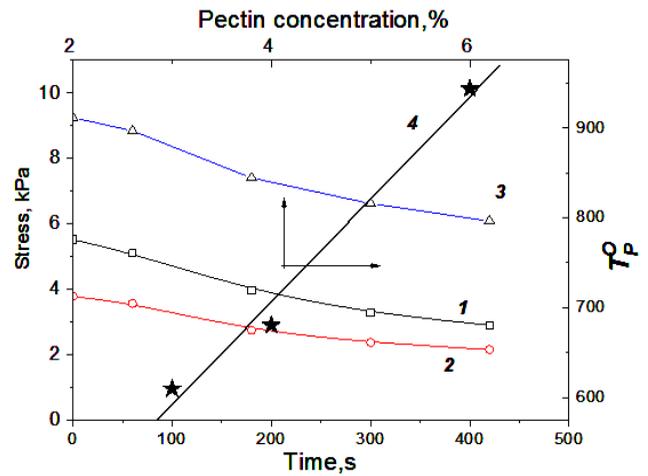


Fig. 3. Relaxation curves (1–3) for pectin hydrogels obtained at different concentrations of pectin (1–3 %, 2–4 %, 3–6 %) and the dependence of the relaxation period T_p^0 on the pectin concentration (curve 4)

The dependence of relaxation stresses in Maxwell's bodies on the relaxation time can be described by an exponential law:

$$t = t_0 \times e^{-\frac{G}{h} \times t} \quad (3)$$

$h/G = T_p$ – relaxation period, h – the coefficient of viscosity of a medium, G – Young's modulus, t – relaxation time.

The values of the relaxation period determined using this law (according to the curves in Fig. 3) are shown in the same figure (curve 4). Almost directly proportional dependence of these values on the concentration of pectin suggests that the elastic properties of the hydrogel naturally increase with the concentration of pectin. However, the physical and mechanical properties of hydrogels (for example, the instantaneous Young's modulus according to the curves of Fig. 1) do not show such a dependence. Since the physical and mechanical properties of elastic-plastic bodies should not be characterized only by Young's modulus, the volumetric compression modulus for samples of hydrogels with different pectin concentrations was determined by the equation:

$$-P \times \frac{V_0}{DV_0} = E\phi - \frac{E_v}{2} \times \frac{DV_0}{V_0} \quad (4)$$

P – the pressure, DV_0/V_0 – the relative loss of volume observed at a given pressure, E_v i E'_v – volumetric compression module and supposed compression module.

The straightening of the loading curves in the coordinates of equation (4) and the dependence of the volumetric compression modulus on the pectin content in the sample is shown in Fig. 4.

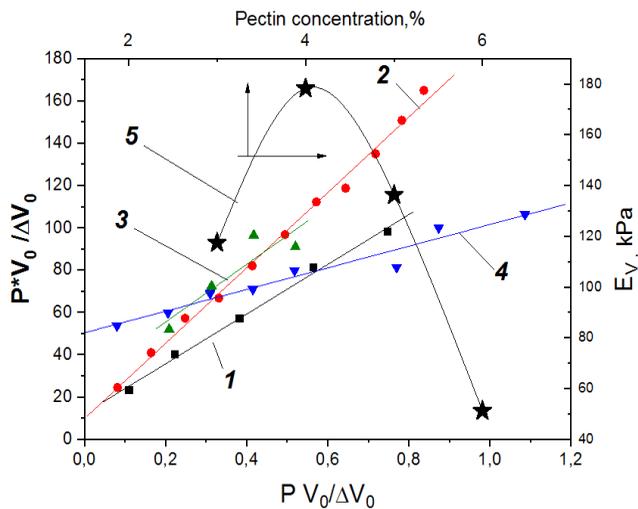


Fig. 4. Processing of pectin hydrogel load curves obtained at different pectin concentrations (1–3 %, 2–4 %, 3–5 %, 4–6 %) and the dependence of the volume compression modulus of these hydrogels on the pectin concentration (curve 5)

Curve 5 in Fig. 4 allows concluding it has an optimal character with a maximum modulus value in the range of pectin concentrations close to 4 %. The instantaneous Young's modulus confirms this conclusion.

Studies show that the compression of hydrogel sheets to significant degrees of plastic deformation is one of the factors that significantly affect the ability of pectin hydrogels to swell. The effect of the compression degree of pectin hydrogels on their absorbent capacity is shown in Fig. 5.

Bars 1 in Fig. 5 show the IWC of hydrogel sheets at different degrees of compression. The first bar of this group shows hydrogel IWC after molding (without pressing), which makes about 17 grams of water per 1 gram of pectin. Naturally, the IWC value decreases with increasing compression degree during pressing, for example, at a compression degree of 0.7 (the last bar of this group) the IWC makes 5 g/g. Bars of group 2 show MWC of hydrogel samples. One can observe that the difference between the initial and equilibrium (maximum) swelling of the unpressed sample is insignificant and corresponds to the data in Table 1. This difference increases significantly for the compressed samples. If only the process reversed to collapse was observed, the degree of swelling of the sample would be the same as for the unpressed sample (horizontal line in the figure).

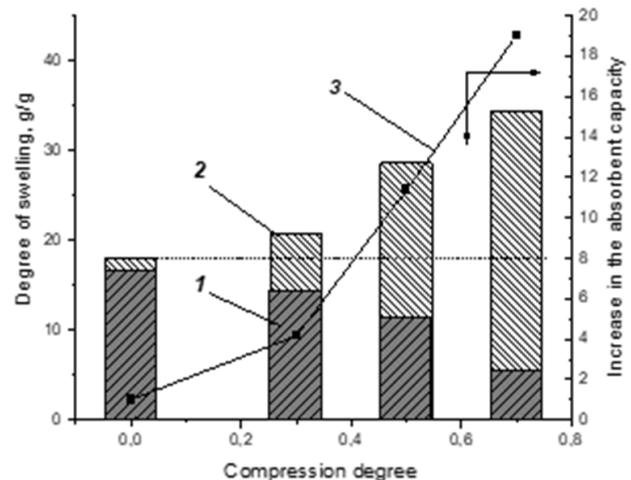


Fig. 5. Dependence of the degree of swelling for pectin hydrogel sheets on the degree of their compression.

However, there is a significant excess of this level. This means that there are significant changes in hydrogel structure during its mechanical collapse. Therefore, the unit of mass of pectin, structured by calcium ions, becomes capable of absorbing more water. Thus, 1 gram of pectin in an unpressed gel sample absorbs about 18 ÷ 19 grams of water at equilibrium swelling, and after pressing to a compression degree 0.7 one gram of pectin absorbs up to 33 ÷ 35 g of water. Curve 3 in Fig. 5 shows the increase in the absorbent capacity of the pressed sheet (to the compression degree 0.7) 19 times

compared to the unpressed sheet. Hydrogel materials with such water absorption capacity are able for a satisfactory fluid drain from the wound site, and this allows using them for wet wound therapy.

The importance of hydrogel morphology transformations during mechanical collapse caused by an external force is shown in Fig. 6. In particular, curve 1 of this figure shows an increase in the proportion of gel fraction: at the compression degree 0.7, its content reaches 75–80 %. With the increasing content of the gel fraction, there is an increase in the density of the spatially structured polymer.

The processes that occur in the sample of pectin hydrogel during compression, can be assessed using the data of the material balance of the released liquid composition (Table 2). One can observe that the liquid contains a low molecular weight part of the sol fraction, i.e. weakly structured macromolecules or those molecules that did not participate in the structuring process. It is confirmed by the low content of calcium ions in the liquid released from the gel. Thus, according to experimental data, the content of calcium ions in the liquid is about 0.0064 %, while the content of calcium ions in the sample of the original hydrogel is 0.34 %, and in the sample of pressed

hydrogel – 0.56 %. In addition, the whole amount of gel fraction remains in the hydrogel sample and there is no significant increase in its amount due to the increase in the concentration of calcium ions. Probably, the increase in the content of the gel fraction occurs only due to the loss of the total weight of the hydrogel sample.

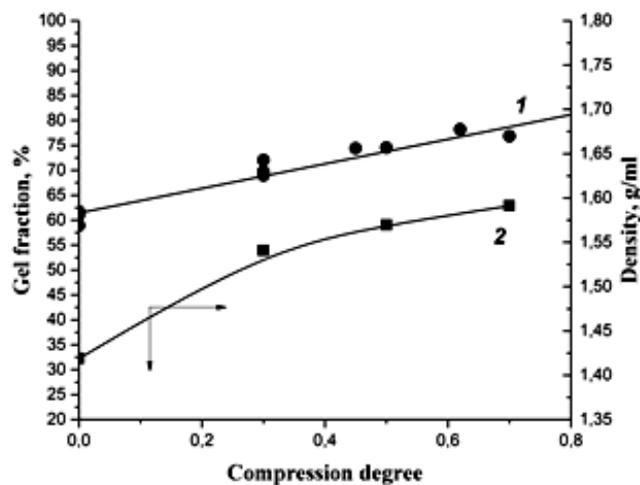


Fig. 6. Dependence of gel fraction (curve 1) and density (curve 2) of gel-forming polymer on the compression degree of pectin hydrogel sheets

Table 2. The composition of the liquid released during the pressing of pectin hydrogels

Characteristics of the sample	Hydrogel sample before compression	The hydrogel sample after compression to a compression degree 0.5	Liquid released after compression
Sample weight, g	100	49÷52	48÷51
Non-volatile substances, g	5.9÷6.2	4.2 ÷4.6	1.3÷1.5
Gel-forming polymer (pectin, sodium alginate, calcium ions)	5.6 ÷5.8	4.0÷4.3	1.2 ÷1.4
The amount of calcium in the terms of the weight of free metal, g	0.31÷0.36	0.26÷0.29	0.0028÷0.0035
Gel fraction, g	3.6÷3.9	3.1 ÷ 3.4	0
Sol fraction, g	2.2 ÷ 2.4	1.0 ÷ 1.25	1.2 ÷1.4

Notes: averaged results on studies of the released liquid composition for several samples are given; values are given in terms of 100 g of the initial weight of hydrogels

The obtained experimental data do not allow to unambiguously state the reason why hydrogel after mechanical collapse increases its absorption capacity. However, one can assume unequivocally that the formation of hydrogel sheets via the described method occurs under conditions of pronounced microhysteresis. According to this mechanism, the formation of a cross-structured network in a solution of gel-forming polymers occurs not

uniformly throughout the volume, but in separate nucleation centers.³⁶ After the formation of nucleation centers, their growth occurs frontally with auto-acceleration and capture of calcium ions, macromolecules that did not participate in crosslinking, and poorly structured macromolecules (sol fractions). Thus, a heterogeneous system with an inhomogeneous distribution of reagents is formed. This structure allows us to explain the plastic properties of the

formed hydrogel sheets and the low content of calcium ions in the liquid after the collapse. According to this mechanism, the gel fraction is formed as separate inclusions with a significant gradient of the degree of structuring from their core to the periphery. It should be noted that this structure does not have an increased ability to swell. Therefore, the formation of a gel-forming network in the terms of syneresis can explain the formation of pectin hydrogel in a state close to the equilibrium degree of swelling.

There is a partial separation of the continuous phase, as well as a significant convergence of the microhydrogel particles during the pressing of such a heterogeneous system. As a result, physical forces of interaction between these particles appear leading to increased elastic properties of the sample. The increase in absorption properties can be explained by the equalization of the distribution of calcium ions in volume, which can be caused by diffusion processes when the particles of microhydrogel combine or macromolecules move relative to each other under the action of external forces.

4. Conclusions

The results described in the report suggest that hydrogels formed from highly esterified pectin or composition of highly esterified pectin and sodium alginate can be described by the model of Maxwell's elastic-viscous relaxing body. It is shown that these properties, as well as absorption ones (in particular the ability to absorb water), are determined by the method of hydrogel formation. The microhysteresis occurs during hydrogel formation, with a pronounced heterogeneous system with significant redistribution of components in volume and explains these properties. Then, during the collapse caused by external forces, there is a decrease in the volume of the hydrogel samples with the release of a liquid containing only a sol fraction of the polymer. Thus, it can be concluded that there are significant morphological changes in the pectin hydrogel sheet after its mechanical pressing. As a result, the elastic properties of the pressed samples significantly as well as their absorption capacity increase.

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ДОСЛІДЖЕННЯ ВЛАСТИВОСТЕЙ ГІДРОГЕЛЕВИХ ПЛАСТИН НА ОСНОВІ ВИСОКОЕСТЕРИФІКОВАНОГО ПЕКТИНУ

Анотація. У повідомленні описані особливості фізико-механічних властивостей і абсорбційної спроможності гідрогелів, отриманих на основі вискоестерифікованого пектину. На основі ряду експериментальних даних продемонстровано взаємозв'язок між цими величинами та зроблена спроба пояснити отримані залежності через морфологію гідрогелю та механізм його формування.

Ключові слова: пектин, гідрогель, гель-фракція, абсорбуюча здатність, пластична деформація.