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STUDY OF PHYSICO-CHEMICAL PROPERTIES OF BIOMATERIAL OBTAINED BASED ON STRUCTURED COLLAGEN

Abstract. The article presents the results of the study of the physicochemical properties of the biomaterial in the form of a film based on collagen, obtained from the hide of cattle with the preservation of the original fibrous structure, and Na-CMC. The temperature dependence of the rheological properties of collagen, as well as compositions based on collagen and Na-CMC in different mass ratios, was studied. To study the preservation of the three-helix structure of collagen, the dynamic viscosity of aqueous solutions of collagen and compositions based on it were studied at temperatures of 30 and 35°C. It has been established that at a temperature of 35 °C the native structure of collagen is destroyed. The optimum temperature for the formation of a biomaterial based on an aqueous composition of collagen/polysaccharide in the form of a film is less than 30 °C. It is shown that the presence of a plasticizer significantly affects the mechanical properties of the films. When glycerol was added as a plasticizer to aqueous solutions of the collagen/Na-CMC composition, the formation of a highly elastic film was observed. When studying the dissolution time of the experimental samples, it was shown that with an increase in the mass fraction of Na-CMC in the composition of the film, a decrease in swelling and an increase in the dissolution time are observed due to the formation of intermolecular hydrogen bonds between the functional groups of collagen molecules and Na-CMC.

Keywords: collagen; polysaccharide; composition; viscosity; film.

1. Introduction

Collagen contains a significant number of diverse functional groups that can interact with medicinal substances and form complexes, the stability of which is determined by many factors. In addition, collagen macromolecules with an asymmetric three-helical conformation are prone to aggregation with the formation of supramolecular fibrous structures built from fibrillar elements, which can also cause the inclusion of various drugs in their composition [1].

Collagen has long been used in wound care as a wound dressing in various forms such as powder, amorphous gels/pastes, gel-impregnated dressings, and pads. In the skin, collagen peptides act as false collagen degradation peptides that send a false signal to fibroblast cells to synthesize new collagen fibers. In addition, collagen peptides have chemotactic properties; they can promote cell migration and proliferation, which is an important process in wound healing [2–4]. Abroad in 1959, for the first time, films reconstructed from a collagen solution were used for the treatment of wounds [5]. Subsequently, several researchers in the experiment and clinic showed the effectiveness of the use of film materials from bovine skin collagen in the treatment of wounds of the skin and internal organs [6–7]. Employees of the 1st Moscow Medical Institute. THEM. Sechenov and the Institute of Light Industry 1963 began research on the study of plastic materials based on type I collagen obtained from the dermis of cattle [8–10].

Biocompatibility and biodegradability are two important parameters to consider when choosing a polymer for its biomedical use. Biocompatibility refers to the specific properties of a material that does not have toxic or harmful effects on biological systems. Biocompatible polymers for pharmaceutical dosage forms have allowed the advancement of pharmaceuticals, providing better therapy [11–13].

When developing new medical biomaterials and improving formulations and technology, pharmaceutical factors are of particular importance. One of them is the biomaterial shapers, which can provide the structural and mechanical properties of the dosage form, create an optimal microenvironment for wound regeneration, have an absorption capacity for excess wound exudate, contribute to the creation of optimal wound surface moisture, and have sufficient mechanical performance [14].

To obtain a medical biomaterial in the form of a film based on collagen, it is necessary to choose a biocompatible polysaccharide with collagen. Since due to the high viscosity of the water mass of collagen, it is impossible to obtain a biomaterial in the form of a film, we have conducted studies on the preparation of an aqueous composition of collagen and Na-CMC.

The purpose of this work is to study the physicochemical properties of a biomaterial obtained based on structured collagen.

2. Materials and Methods

2.1. Material

The object of the study was the aqueous mass of neutral collagen isolated from the skin of cattle (dry residue 4.5–5%) [15] and sodium salt of carboxymethylcellulose (Na-CMC) with a degree of polymerization of 530 and a degree of substitution for $-CH_2COONa$ groups of 85%.

2.2. Obtain a composition

Distilled water was added to the neutral aqueous mass of collagen (dry residue 4.5–5%), up to a collagen concentration of 1.5%, and Na-CMC powder was added by stirring in different mass ratios. Mixing was carried out at room temperature for 30 min until the components were completely dissolved and a homogeneous dough-like mass was formed.

2.3. Obtain the film

Glycerol was added to the aqueous composition as a plasticizer and mixed on a magnetic stirrer at room temperature for 10 min, the mixture was poured into a mold and dried at room temperature not exceeding 30 °C.

2.4. Studies of the rheological properties

Studies of the rheological properties of aqueous solutions of collagen and the composition were carried out on a rotational viscometer HAAKE Viscotester 2 plus, at temperatures of 20, 25, 30, and 35 °C.

2.5. Determination elasticity of film

The elasticity of the film was determined visually by the absence of adhesion or brittle fracture when the film was folded 180° ("++" – elastic; "+" – elastic, but elastic when folded; "–" – no elasticity).

2.6. Determination of geometric dimensions

The geometric dimensions of the film were measured with a micrometer with a division value of 0.01 mm according to OST 64–072–89.

2.7. Film dissolution time determination

The dissolution time of the film was evaluated by dissolving with continuous stirring film samples of 2 × 2 cm in 20 ml of purified water (temperature 37 ± ± 2 °C) and measured the time of complete dissolution in minutes.

2.8. Determination of the pH of the aqueous extract

The pH of the aqueous extract of the film was determined potentiometrically at a temperature of 20 °C.

3. Results

It is known from the literature data that an increase in temperature to 30 °C leads to a conformational change in the structure of collagen helices, accompanied by a "helix-coil" transition, as a result of which the supramolecular structure of collagen solutions also changes, and the viscosity and structural parameters drop sharply [16]. When aqueous solutions of collagen are heated to 30 °C and higher, a sharp irreversible decrease in viscosity is observed, a change in the structure of the collagen molecule, and denaturation also occurs. Therefore, during our research, the temperature dependence of the rheological properties of collagen, as well as compositions based on collagen and Na-CMC in different mass ratios, were studied (Figures 1, 2).

4. Discussion

As can be seen from figure 1, at temperatures of 20 and 25 °C, there are no sharp changes in the dynamic viscosity of collagen and compositions based on collagen/Na-CMC.

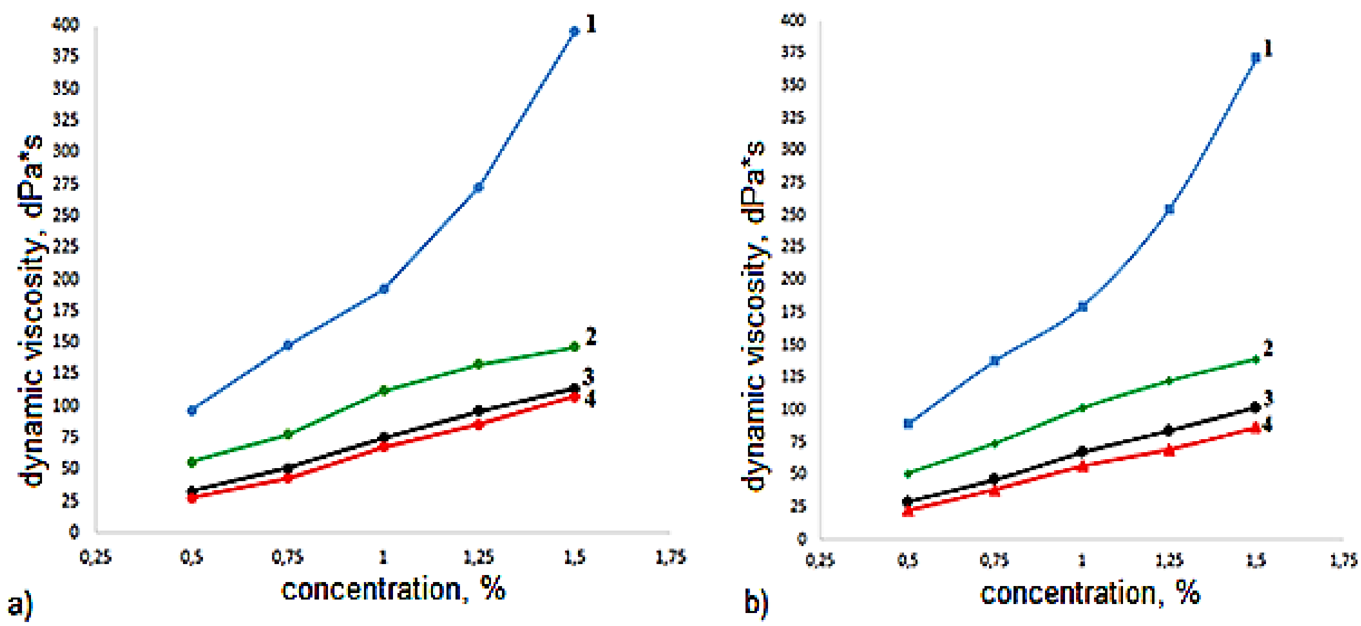

Figure 1. Dynamic viscosity at temperatures, a) 20ºC; b) 25ºC: 1 – collagen, 2 – collagen/ Na-CMC80/20, 3 – collagen/Na-CMC75/25, 4 – collagen/Na-CMC70/30

To study the preservation of the three-helix structure of collagen, we studied the dynamic viscosity of aqueous solutions of collagen and compositions based on it at temperatures of 30 and 35 °C (Figure 2).

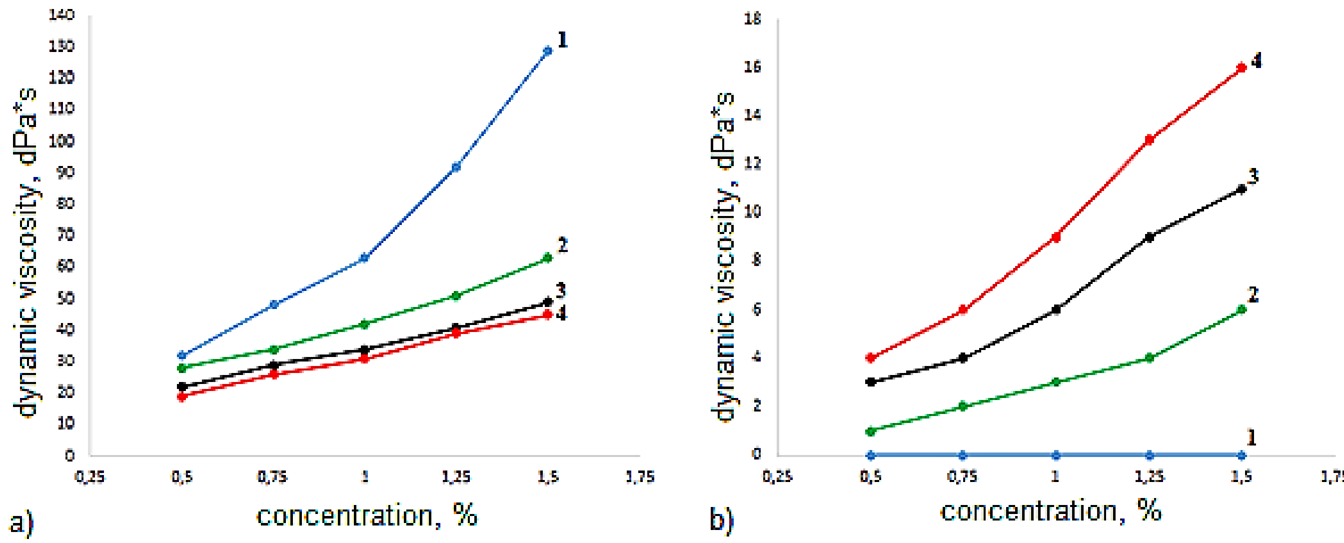

Figure 2. Dynamic viscosity at temperatures, a) 30ºC; b) 35ºC: 1 – collagen, 2 – collagen/Na-CMC80/20, 3 – collagen/Na-CMC75/25, 4 – collagen/Na-CMC70/30

As can be seen from figure 2, at temperatures of 30 and 35 °C there is a sharp change in the dynamic viscosity of collagen and compositions based on collagen/Na-CMC. However, at a temperature of 35 ºC, the dynamic viscosity of an aqueous solution of collagen in all studied concentrations is equal to zero. These data indicate that at a temperature of 35 °C, the native structure of collagen is destroyed. The data obtained indicate that for the formation of biomaterial-based on collagen/polysaccharide in the form of a film, the optimum temperature is less than 30 °C.

To obtain a biomaterial based on collagen in the form of a film, glycerol was added to a homogeneous aqueous solution of the collagen/Na-CMC composition with stirring and poured into a mold at room temperature. The presence of a plasticizer significantly affects the mechanical properties of the films. It turned out that when glycerol is added as a plasticizer to aqueous solutions of the collagen/Na-CMC composition, the elasticity of the film increases. Glycerin conformed to the compositions and showed good plasticizing properties, which led to the formation of a highly elastic film.

Elasticity is one of the indicators of physiology, the ability of the coating to take the shape of the human body and come into contact with the entire surface of the tissues. All obtained film samples have good elasticity. The physical and mechanical properties of the studied films are shown in (Table 1).

Table 1. – Physical and mechanical properties of biopolymer films

№	Film composition	Elasticity	Thickness, mm	Density, g/cm³	Dissolution time, min	the pH of water extract
1	Collagen-Na-CMC80/20	+ +	0,11	1,09	180	6.8
2	Collagen-Na-CMC75/25	+ +	0,11	1,07	over 200	6.9
3	Collagen-Na-CMC70/30	+ +	0,11	1,04	over 200	6.9

The study of the dissolution time of experimental film samples showed that with an increase in the mass fraction of Na-CMC in the composition of the film, a decrease in swelling and an increase in the dissolution time are observed, respectively. This is explained by the fact that intermolecular hydrogen bonds are formed between the functional groups of the collagen molecule and Na-CMC, and films based on them become stronger.

5. Conclusions

The temperature dependence of the rheological properties of collagen, as well as compositions based on collagen and Na-CMC in different mass ratios, was studied. It has been established that at a temperature of 35 °C the native structure of collagen is destroyed. The optimum temperature for forming a biomaterial based on collagen/polysaccharide in the form of a film is less than 30 °C. It is shown that the presence of a plasticizer significantly affects the mechanical properties of the films. When glycerol was added as a plasticizer to aqueous solutions of the collagen/Na-CMC composition, the formation of a highly elastic film was observed. It has been established that with an increase in the mass fraction of Na-CMC in the film composition, a decrease in swelling and an increase in the dissolution time are observed due to the formation of intermolecular hydrogen bonds between the functional groups of collagen molecules and Na-CMC.

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