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Biopolymeric materials containing brown algae polysaccharides

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ABSTRACT

This article describes the use of polymeric systems created on the basis of natural or synthetic polymers. One of the most safe and biocompatible is fucoidan. Therefore, were proposed and developed the idea of creating films based on methylcellulose with fucoidan with the addition of various plasticizers. It was found that surface films affect the healing of wounds, as it causes the ability of the cells adhesion, the adsorption of water molecules. Also was investigated the physical properties of the films with different plasticizers, for example use as a plasticizer of the lactic acid allows to increase the capacity for swelling, as well as increase the ability to retain adsorbed, which is necessary for healing of wounds after burns. The use of glycerin or propylene glycol resulted in a decrease in the degree of swelling, and the absence of the plasticizer contributes to enhancement of the adsorption of water molecules. Data from biological films possess high ability to swell, and therefore not only applicable for the treatment of wounds with lots of exudate, but due to the high holding capacity applicable for low exudative wounds.

Keywords: biocompatibility, fucoidan, methylcellulose, medical materials, wound healing, plasticizer, fimls.

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INTRODUCTION

Regeneration of damaged tissue is a priority of tissue engineering. The polymer systems created using natural and synthetic polymers are used for these purposes. Gel structures, composite hydrogel, films can be used as materials. The matrices under development must have certain properties. Firstly, it must be a bioactive system, contributing cell proliferation and differentiation for effective tissue regeneration [4]. Secondly, it must be biocompatible and has high tissue cell affinity. Thirdly it has to be physically sustainable until implantation and biodegradable after implantation. And fourthly both material and decay product must have no toxic affect on regenerated tissues [5]. Natural polymers are used in tissue engineering because they are safe and biocompatible. Fucoidan was chosen as a polymer of this type. Fucoidan is pertained to highly-sulphated polymers which secreted by brown algae. Fucoidan has antimicrobial and wound-healing value coming from its ability to interact with growth factors such as primary growth factor (bFGF) and transforming growth factor-β (TGF-β) [11,6]. However there are some researches on creating wound-healing preparations and considering biopolymer usage as an agent in tissue engineering [12,13,17,7]. That is why the aim of the research is creation and study of physical properties of films on the ground of methylcellulose with the using of Fucoidan and some several-sorted plasticizes. Thank to its molecular weight and chemical structure fucoidan and methylcellulose are satisfactory filmforming elements which can form structures stabled by hydrogen bindings.

MATERIAL AND METHODS

Materials

Fucoidan (molecular weight [MW] 80 kd from Fucus vesiculosus), lactic acid (85% wt/vol) were purchased from Sigma Chemical Co (St Louis, MO); propylene glycol and glycerol (LLC "Company ElGrupp" (Russia);methylcellulose MC-100 (162,14)n, (LLC "Chimprom Usolie" (Russia).

Preparation of films

Biofilms were obtained by solvent evaporation. Fucoidan and methylcellulose were dissolved in hot water at 65 C. Various component ratios of fucoidan , methylcellulose, glycerol, propylene glycol and lactic acid were prepared for optimizing the composition. Resulting mixtures were sonicated to remove air bubbles then transported to a Petri dish (10.0 mL), heated to 30 ° C and dried. After drying the matrices were ejected and stored in a hermetic container at room temperature for further research.

The efficiency of absorption and efficiency of retention water.

Films were weighed (Wdry), inserted in a Petri dish with the subsequent dropwise addition of phosphate-buffered saline (pH 7.4) up to a constant film weight at 25 ° C. The buffer excess was removed with filter paper and weighed (W'wet). Each experiment was performed three times. The degree of turgescing was calculated according to the formula (1).

EA = [(W'wet - Wdry) / Wdry] × 100 (1)

Biofilms were placed in phosphate-buffered saline (PBS) (pH 7.4) and kept for 1 hr., then extracted and dried with filter paper. After that, the samples were centrifuged (3500 × g, 10 min), filter paper was placed on the bottom of the tube in advance. The samples were weighed (W"wet). Measurements were performed three times. The degree of water retention was calculated according to the formula (2) [15].

ER = [(W'wet - W" wet) / W"wet] × 100 (2)

The degradation and biodegradation of films

To determine the films stability the systems with pH 1.5, pH 7.4, pH 9.1 and the amylase solution (1000 U / L) were used. To determine the degradation of films 1g were placed in Petri dishes, added 20 ml of the buffer and kept at 25 ° C. To understand a biodegradation mechanism the samples of composite film matrices with the mass 1g were added to the amylase solution (1000 U / L) in 1M phosphate-buffered saline with the pH 7.4 (1 × PBS) and incubated for 1 to 24 hours at 37 ° C. After completion of the incubation period, the samples were washed with deionized water to remove ions and dried with filter paper. Then they were placed in a drying cabinet (50 ° C) and weighed (Wt). The measurement was made three times. Biodegradation degree was calculated according to the formula [16]:

(Bio)degradation = (W0 - Wt) / W0 × 100 (3)

Statistical analysis.

To confirm the validity of the results a statistical data processing was carried out using one-way analysis of variance (ANOVA) and Microsoft Excel, Statistica 6.0 programs. The level of statistical significance of differences was accepted at a value of P <0,05.

RESULT AND DISCUSSION

Film surface influences wound-healing process as it influences cell ability to adhesion, water molecule adsorption. Film physical properties especially its porosity, adhesion by surface depends on properties of used polymers particularly on grid density, degree of branching, flexibility. Functional groups of used elements can also influence film adhesive properties. Bioflims degrees of swelling and water retention were determined in the course of the research, presented in Fig. 2. Usage of nothing but lactic acid (A2) as a plasticizer allows to raise swelling ability (73,58%) as well as to raise water retention ability. Absorption and retention ability was estimated to define if it possible to use obtained films as healing wound covering. It was defined that evolution process of cells from different biomimetical materials has a great impact on chemical (presence of certain atomic groups) and physical features of surface [1, 2]. Films that are used for wound and burn healing must absorb exudates effectively and at the same time they must keep the moisture on wound surface. Films must be strong and able to prevent growth of pathogenic flora. So the water adsorption ability is an important feature of woundhealing films because effective exudates absorption leads to wound moisture level reduction and its faster healing [16]. Usage of nothing but lactic acid as a plasticizer (A2) leads to increase of water adsorption ability which corresponds to obtained results [14]. Propylene glycol (A4) and glycerin (A3) injections help to reduce swelling degree because of slower salvation of hydrophilic groups of plasticizer on film surface in comparison with the lactic acid. And also because of increase of surface tension that leads to reduce of wetness degree. Absence of plasticizer (A1) also help to increase water molecule adsorption (69,3%) but with the reduce of ER at the same time. That is caused by presence of uncombined water molecules. Usage of propylene glycol also leads to water retention ability reduction which is possible because of water clusters formation on film surface. That leads to increase of matrix volume and its permeability [4, 9].

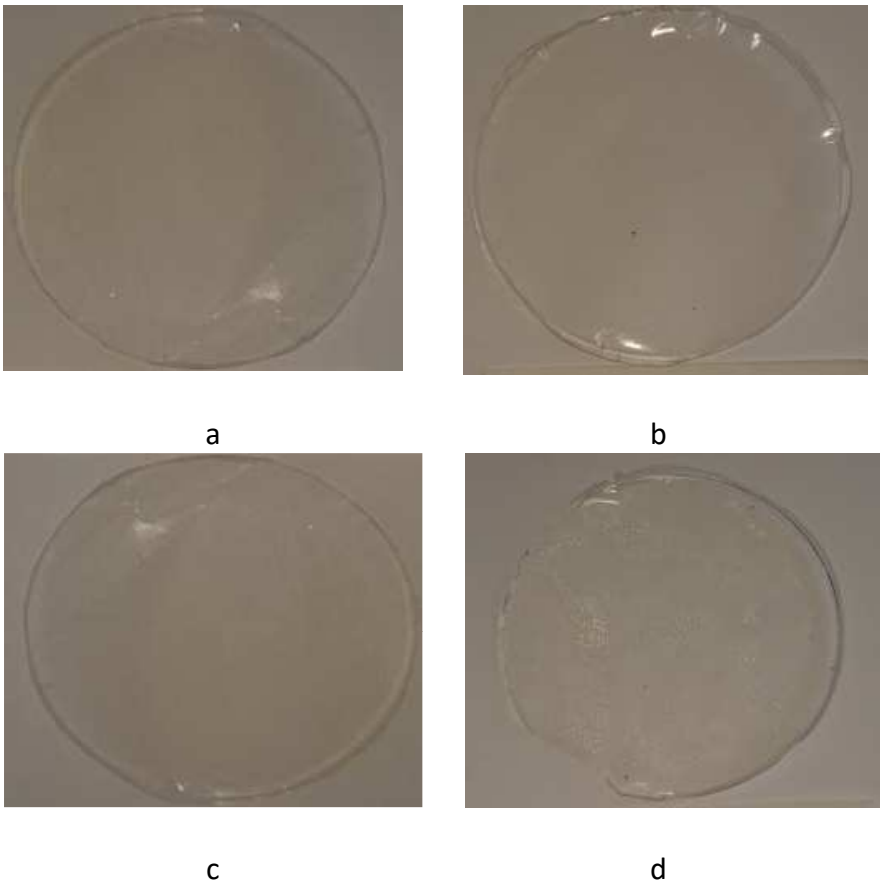

Fig. 1. Film photography. *a-A1, b-A2, c-A3, d-A4*

Table 1: Composition and code of biofilms

Code combination	Methylcellulose, Conc (%)	Fucoidan, Conc (%)	Types of plasticizer		
			Glycerol, Conc (%)	Lactic acid, Conc (%)	Propylene glycol, Conc (%)
A1	1,1	0,5			
A2	1,1	0,5		0,1	
A3	1,1	0,5	0,4	0,1	
A4	1,1	0,5		0,1	0,4

** Conc - final concentration, P <0.05.*

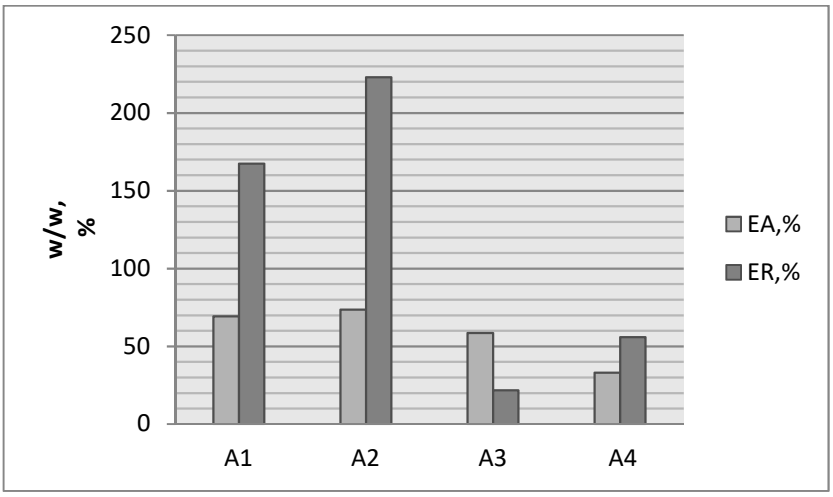

Fig. 2. Swelling degree (EA) and water retention degree (ER)

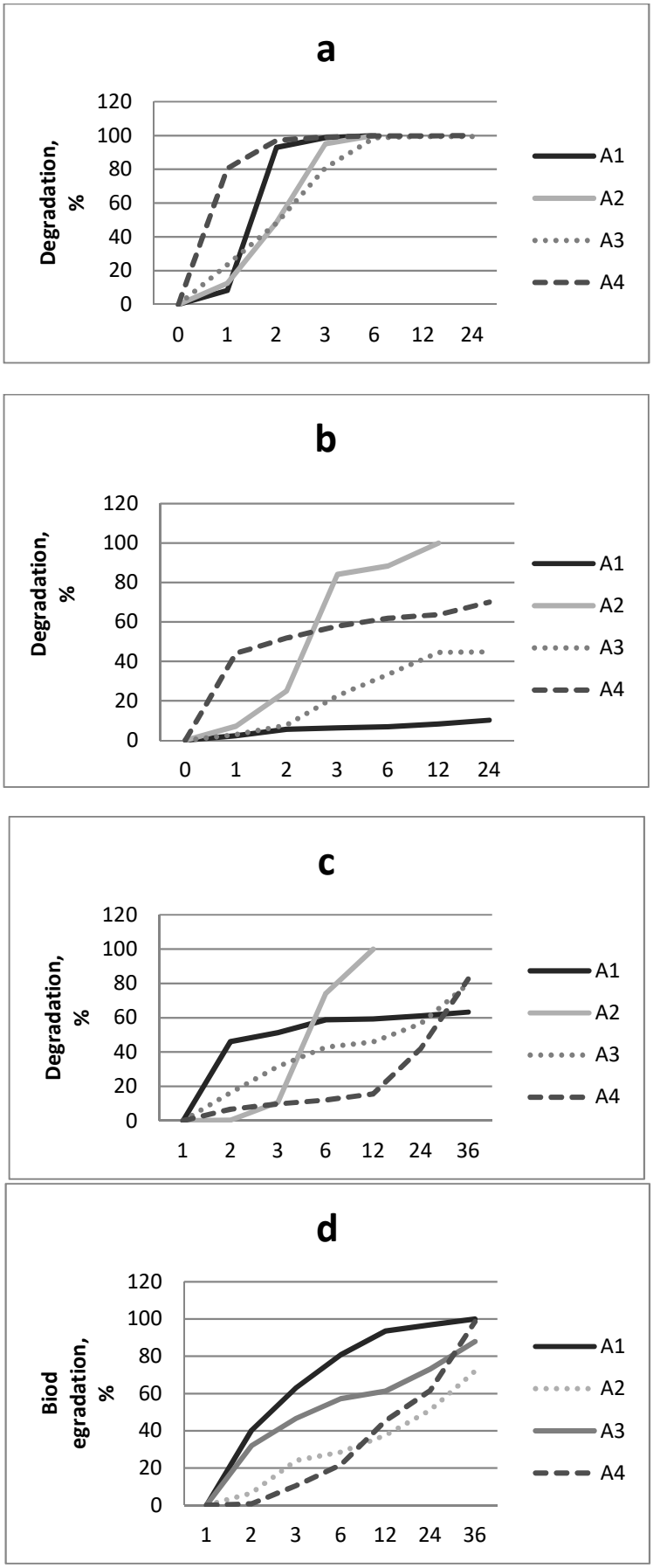

Fig 3. Stability of biomimetical films at pH 1.5 (a), pH 7,4 (b), pH 9,1 (c), wish lysozyme (d).

Hydrophility of film surface A1 and A2 leads to increase wound adhesion what shows the ability of usage as I-phase wound cover. However current films applicable for poorly exudative wounds because of low effectiveness of water adsorption but high holding capacity at the same time. Determination of biometric film degradation allows not only to state the ability of its usage as wound covers but also to optimize conditions of its usage and storage and ability of its removal as well. According to achieved data films formed with plasticizers (A3, A4) are more stable in alkaline, weakly alkaline and enzyme-containing environment. Plasticizer injection leads to reduction of wetness degree which resulted in reduction of degradation degree.

All film systems had low stability in strongly acidic environment (Fig. 3a). Complete destruction occurred after 3 hours in buffer with pH 1,5. In alkaline, weakly alkaline environment (Fig. 3 b, c) films that contained glycerin (A3) and propylene glycol (A4) as a plasticizer had the highest stability. In condition of degradation the system with the lactic acid as a plasticizer appeared as stable.

CONCLUSION

Achieved biomimetical films have low swelling capability and can't be highly applied in healing of vast wound which produce lots of exudate. However films can be used for healing II-phase or low-exudated wounds due to high retention ability and stability in enzymological environment. Films contained glycerin (A3) and propylene glycol (A4) have stronger surface tension than others (A1 and A2). That would reduce material adhesion with the wound and would simplify its removal.

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Authors contribution
SVE and DAA designed the experiments and performed the experimental works. DEV and SVE contributed ideas, gave critical feedback during data analysis and manuscript write-ups. All authors read and approved the final manuscript.

Conflict of Interest
The authors declare that they have no competing interest.

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