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ORIGINAL ARTICLE

Synthesis and study on the properties of polysaccharides modified via the Steglich reaction

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Abstract
Modified polysaccharides are widely used as polymers for medical and biomedical purposes. Among them, the products with a crosslinked macromolecular structure occupy an important place. For instance, using N-derivatives of glutamic acid as a crosslinking agent makes it is possible to obtain non-toxic biodegradable polymer materials. In the current work, the results of study on the polysaccharides modification with N-derivatives of glutamic acid via the Steglich esterification reaction are provided. The factors influencing the efficiency of developed synthetic approach were considered, and the structure and composition of the obtained reaction products were investigated. The reaction was shown to proceed successfully by exploiting both carboxyl groups of N-derivatives of glutamic acid, resulting in the cross-linking of polysaccharide molecules under mild conditions. The study of the properties of the obtained products and the NMR study of their structure and composition showed that two fundamentally different polymers were formed—macromolecules with branched and crosslinked structures. The latter, under certain reaction conditions, formed microhydrogels while dispersing in water. Meanwhile, the branched macromolecules were capable of forming self-stabilized aqueous dispersions. Thus, the article presents the research results on the creation of novel polymers for potential medical applications.

Keywords Modification · Polysaccharide · Structuring · Medical polymers · STEGLICH reaction

Introduction

Taking into account their beneficial properties, polysaccharides have undeniable potential when used in biological, medical, and pharmaceutical fields (Barclay et al. 2019, Alhaique et al. 2015, Ankur et al. 2021, Lima et al. 2020, Nastyshyn et al. 2022). This is due to their prevalence, belonging to renewable raw materials, non-toxicity, chemical resistance, hydrophilicity, and tolerance to the human body, as well as the presence of a glycosidic bond that unites mono- and disaccharide fragments in the polysaccharide molecule (Metaxa et al. 2021, Huang et al. 2015,

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Mavromoustakos et al. 2021, Nayak and Hasnain 2020, Barclay et al. 2018). The structure of most natural polysaccharides contains hydrophilic groups (hydroxyl, carboxyl, and amino groups), which can form non-covalent bonds with biological tissues, such as epithelium and mucous membrane, forming bioadhesives (Sood et al. 2021, Lee et al. 2000, Candace et al. 2018, Prasher et al. 2021).

The presence of functional groups in the molecular chains of polysaccharides allows their easy chemical modification (Otache et al. 2021, Yang et al. 2011). According to the structural characteristics of polysaccharides, their modification can occur mainly by the following four mechanisms: covalent crosslinking, ionic crosslinking (Duru et al. 2021, Zhuang et al. 2017, Stetsyshyn et al. 2020), polyelectrolyte complexation (Sasaki et al. 2020, Raczkowska et al. 2014), and self-crosslinking of hydrophobically modified polysaccharides (Luo and Wang 2014).

Covalent crosslinking of polysaccharides using functionalized amino acids under mild conditions ensures the preservation of their natural characteristics. Such a transformation can be carried out via the Steglich esterification reaction. The production of linear and crosslinked polyesters, based on polydiols of the polyoxyethylene and polyoxypropylene series and N-derivatives of glutamic acid, as well as the possibility of obtaining polyesters under mild conditions according to the Steglich reaction is described in our previous papers (Nagornyak et al. 2016; Chekh et al. 2017; Varvarenko et al. 2013; Yakoviv et al. 2020; Stasiuk et al. 2022a, b).

A series of model studies on the interaction of hydroxyl groups of sucrose with acetic, stearic, and glutaric acids according to the Steglich reaction were carried out (Nagornyak et al. 2015). It was established that both primary and secondary hydroxyl groups of saccharides can be involved in the reaction.

This work is aimed to create aqueous dispersions of dextrin structured by N-derivatives of L-glutamic acid via the Steglich reaction for medical and biomedical applications. Moreover, this work assesses the effect of reaction conditions on the properties of the obtained product, in particular, the structure of the main chain, the ability to swell in water, the ability to absorb/adsorb proteins (albumin).

Experimental part

Reagents and solvents

N,N-Dimethylformamide (DMF) was purified to remove water and other impurities according to the method described by Fihurka et al. (2018).

N,N-dicyclohexylcarbodiimide (DCC, Aldrich) was used without additional purification (the main substance content is not less than 99.0%). Dextrin (Aldrich), a mixture of products of partial decomposition of starch homopolysaccharides, consisting of glucosidic residues connected by α-1,4 bonds, was used with a molecular weight of 6200 ÷ 7000 Da. 4-(N,N-dimethylamino)pyridine (DMAP), 99%, known as Steglich catalyst, was bought from Alfa Aesar.

N-stearoyl-L-glutamic acid (GluSt) was synthesized using the procedure described by Stasiuk et al. (2022a, b).

Synthesis of polyesters based on polysaccharides and dicarboxylic acids

N-steroyl glutamic acid (GluSt) and solvent (DMF) were loaded in a double-necked reaction vessel, with connected reflux condenser with a calcium chloride tube, and equipped with a reflux funnel and a magnetic stirrer. The concentration of acid in the reaction mixture was in the range of 12 ÷ 18%. An appropriate amount 5 ÷ 8% polysaccharide solution was added to the mixture and the reactor was placed in a water bath at a temperature of 278–280 K and stirring was turned on. A 2–5% solution of 4-dimethylaminopyridine and 18–20% solution of N,N-dicyclohexylcarbodiimide were added using dropping funnel. The starting molar ratios of the reactants were as follows: R-COOH:DCC = 1:1.1; DCC:DMAP = 1:0.125. N,N-dimethylformamide was used as a solvent for all starting compounds.

After adding DMAP and DCC, the mixture was heated to 283–287 K and the reaction was carried out under stirring for 4 ÷ 4.5 h. At the end of the reaction, the formed precipitate of N,N-dicyclohexylurea (DCU) was separated from the reaction mixture by filtration. The reaction mixture was evaporated using the vacuum of a water jet pump to remove ¾ of its original volume. After this stage, the product was isolated from the concentrated reaction mixture and simultaneously divided into two fractions: the first one is a crosslinked polysaccharide and the second one is a fraction with a branched structure. To obtain the first fraction, methanol was added to the reaction mixture to obtain a precipitate, which was separated by centrifugation (rotation rate is 3000 ÷ 4000 rpm). The precipitate after centrifugation was washed three times with water and dried to a constant weight. To obtain the second fraction, fugat (a mixture of methanol and DMF) was evaporated in the vacuum of a water jet pump.

Analysis methods

^{1}H NMR spectroscopy

^{1}H NMR spectra of monomer and crosslinked polyester samples were obtained in the appropriate deuterated solvents (deuterobenzene, deuterochloroform, deuterated water) using JEOL's ECA Series Nuclear Magnetic Resonance

(NMR) Spectrometer at a frequency of 400 MHz in automatic scanning mode. Signal assignment was performed using database (Yamazaki et al. 2004).

Determination of particle size of dispersions of crosslinked polymers by light scattering

Determination of the particle size of crosslinked polymer dispersions by the light scattering method was carried out on a spectrophotometer "UNIKO-1201" at 5 different wavelengths (λ = 430; 535; 590; 610; 680) using a 24 × 40 mm cuvette with an optical path length of 10 mm. It was compared with distilled water. To determine the particle size of crosslinked polymer dispersions, 10 g of 0.1% polymer dispersion in water was prepared.

After that, a graph lgD vs lgλ was constructed and the index n was determined by the angle of inclination of the obtained curve. According to this index, the Z indicator was selected, according to the table data, and the size of the particles of the dispersed phase was determined according to the Heller equation (Melik and Fogler 1983).

Determination of the particle size of of polyester dispersions by dynamic light scattering (DLS)

The effective hydrodynamic radius of particles of aqueous polyester dispersions in the concentration range of 0.01–0.5% was determined on a Particle Sizing Systems Nicomp 380 ZLS device (California, USA), with a resolution of 3 nm. Particle charge was measured on a Zeta Sizer Nano-ZS90 device (Malvern Instruments Ltd, England).

Surface tension

Surface tension of aqueous dispersions of modified dextrin was measured at 20 ℃ using Du Noüy ring method (Varvarenko et al. 2013). The critical micelle concentration (CMC) was determined using surface tension isotherms of corresponding polymers.

Swelling degree

The degree of swelling was determined at 20 ℃ in distilled water. A hydrogel sample (~ 0.5 g) was placed in Dogadkin's apparatus filled with water. The amount of water absorbed during a certain period of time was determined as the difference in the water level before and after swelling. The degree of swelling α was calculated according to the equation:

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

where m_t is the weight of the swollen hydrogel sample at time t, m_0 is the weight of dry polymer.

Results and discussion

The general mechanism of the Steglich reaction involving dextrin and N-substituted glutamic acid is shown in Fig. 1. The features of the interaction of N-substituted glutamic acid with the hydroxyl groups of dextrin were previously described in (Nagornyak et al. 2016, 2015). NMR studies revealed that there is no significant difference in the reactivity of primary and secondary hydroxyl groups of dextrin, so the scheme does not focus on the substitution position.

According to the scheme (Fig. 1), a polysaccharide with an introduced carboxyl group in its structure is formed via the Steglich reaction, the essence of which is the activation of the carboxyl group of N-substituted glutamic acid (reaction I, Fig. 1) by interaction with dicyclohexylcarbodiimide with the formation of a reactive intermediate compound (compound A, Fig. 1), and its subsequent rapid interaction with the hydroxyl groups of the polysaccharide in the presence of the nucleophilic additive 4-(N,N-dimethylamino) pyridine (reaction II, Fig. 1). During the further transformation of the obtained product (compound B, Fig. 1), free carboxyl groups react with hydroxyl groups of polysaccharide macromolecules forming cross-links. In this case, the dibasic acid acts as a crosslinking agent (reaction IV, Fig. 1). Analysis of the NMR spectra of the obtained products shows that not all GluSt residues are involved in the further transformation according to the reaction IV (NMR spectra (figures SI.1 and SI.3) and their descriptions are presented in the Supplementary Information). This is due to the passivation of the activated form of the carboxyl group (compound C, Fig. 1), and thus, crosslinking cannot occur (reaction V, Fig. 1).

The ratio between reactions II, IV, and V (Fig. 1) leads to the formation of polymeric materials of different structures. The dominance of reactions II and IV leads to the formation of a crosslinked polysaccharide (CD in Fig. 2), and reactions II and IV with a significant proportion of reaction V, lead to the formation of a product with a branched structure (BD in Fig. 2). It should be noted that the ratio between reactions can be regulated by the synthesis conditions and the reagents nature, but in any case, both products are formed in the reaction mixture.

Figure 3 shows the dependence of the BD and CD products yield on the ratio of the starting reagents. According to the given diagram: i) at any ratio of starting reagents, both products are present in the reaction mixture; ii) at a starting ratio of functional groups OH:COOH (mol/mol) below 15:1 the total yield of products is close to 100%; iii) above the 15:1 ratio, the initial unreacted polysaccharide is found

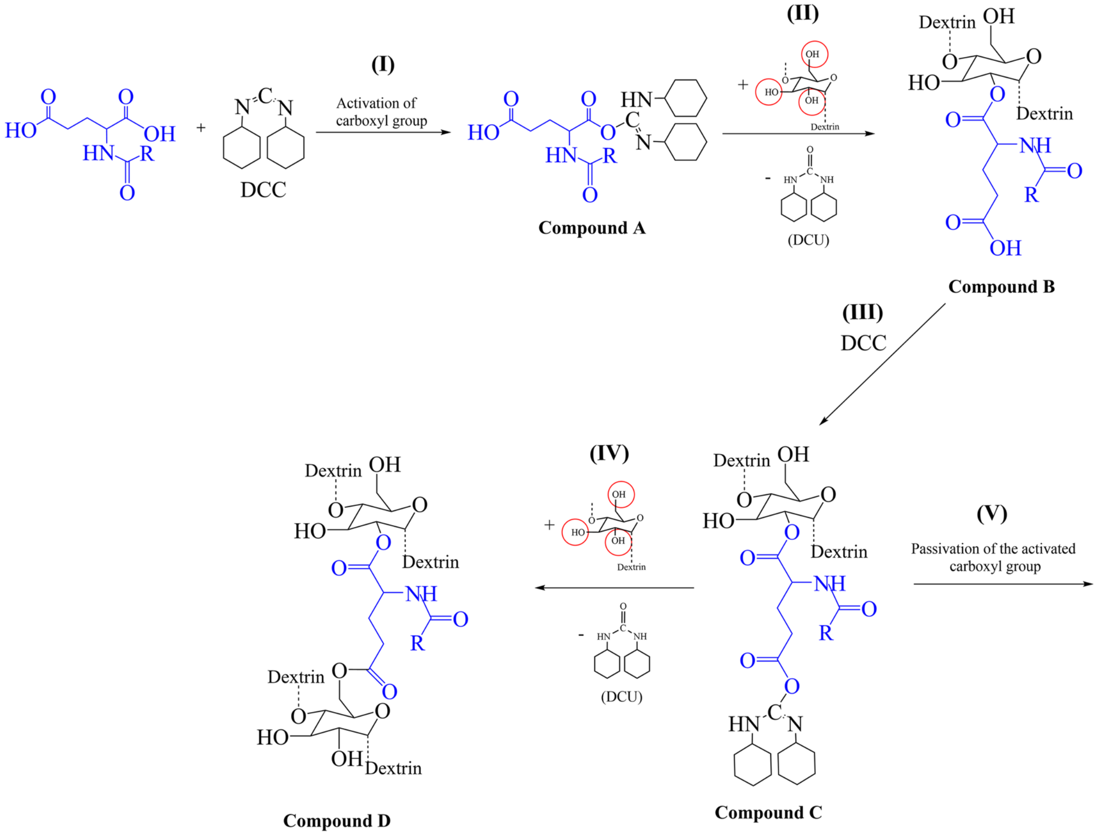

Fig. 1 Interaction of N-substituted glutamic acid with polysaccharide according to the Steglich reaction: (I) activation of the carboxyl group of N-substituted glutamic acid with dicyclohexylcarbodiimide; (II) reaction of the activated carboxyl group with the hydroxyl group of dextrin according to the Steglich reaction; (III) activation of the free carboxyl group of N-substituted glutamic acid grafted to the dextrin macromolecule; (IV) interaction of the activated carboxyl group of N-substituted glutamic acid grafted to the dextrin macromolecule with the hydroxyl group of dextran by the Steglich reaction; (V) formation of a passive form of a carboxyl group incapable of further participation in the Steglich reaction

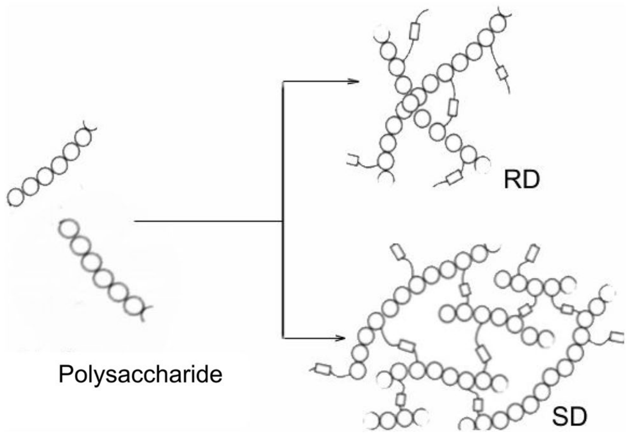

Fig. 2 Scheme of modification of polysaccharide molecules by the Steglich reaction with the formation of branched (BD) and crosslinked (CD) derivatives

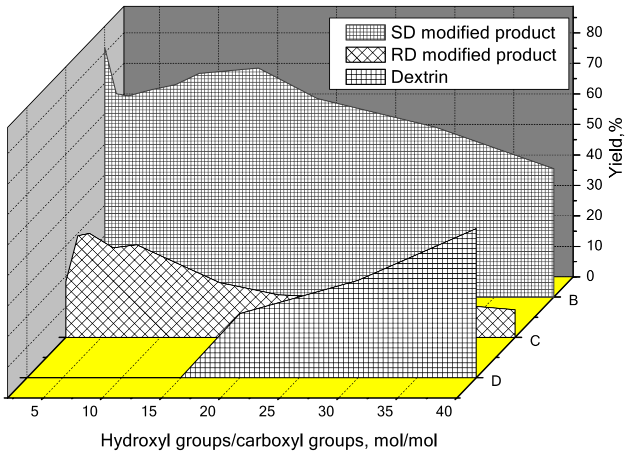

Fig. 3 Dependencies of the reaction products yield on the molar ratio of starting reagents functional groups

in the reaction mixture (or degree of its modification is so insignificant that it does not change the properties).

The yield data and characteristics of each of the products, depending on the synthesis conditions, are given in Table 1. The reaction products described above have significantly different properties, which allows them to be separated from the reaction mixture. The fundamental difference between the obtained products is their solubility, in particular, in methanol. A branched product with an insignificant crosslinking degree dissolves in methanol with the formation of solutions close to the true ones, and the crosslinked product is precipitated with methanol from the reaction mixture. The behavior of these two products in the aqueous medium is also different. Branched (BD) derivative obtained using wide range of the reagent ratios, swells in water and forms self-stabilized colloidal solutions. The behavior of the crosslinked (CD) product in aqueous medium is ambiguous. Samples of the gel fraction (CD product) obtained at the hydroxyl:carboxyl groups ratio lower than 8:1 (Table 1, Fig. 3) are highly hydrophobic powders. Sample 6 (Table 1), obtained at the OH:COOH ratio of 10:1, has intermediate properties, i.e. it is water-wettable, but its swelling in water is very limited. Samples 6 and 7 (Table 1) both swell, however their maximum swelling degree is different and they are not water dispersible. The maximum degree of swelling for sample 8 (Table 1) is 700 percent, however, this sample does not form a stable aqueous dispersion. A further increase of hydroxyl groups content in the reaction mixture leads to the formation of gel fractions of the derivatives (see samples no 9 and 10 in Table 1), which quickly swell in water and easily form dispersions. At the same time, unlike samples of a branched product, their swelling in water remains limited. Thus, it might be suggested that these samples in an aqueous medium form microhydrogels.

The analysis of NMR spectra of BD samples and some CD samples allowed to evaluate the effectiveness of GluSt residues grafting (description and analysis are presented in the Supplementary Information (Table SI.1)). The grafting effectiveness is considered as a ratio of number of grafted acid residues to the number of loaded ones per mole of dextrin link. For the gel fraction, this ratio in the entire range is close to 1. Thus, CD polymer is always formed with a high total grafting efficiency. However, this conclusion cannot be referred to the grafting efficiency involving two groups. At low ratio of reagents, efficiency of grafting exploiting two groups and the gel fraction formation decreases sharply. Although, it cannot be claimed that at the (OH:COOH) ratio below 3:1 the number of branches is insufficient for the formation of a crosslinked polymer. One molecule of dextrin contains 4–5 interactions with two carboxyl groups,

Table 1 Synthesis conditions and yield values for DCU, gel (CD) fractions, sol (BD) fractions, and individual characteristics of the products obtained at 288 K temperature, at total concentration of reagents of 12% and 22 ÷ 24 h reaction duration

Sample	OH:COOH group ratio	Amount of reagents per 1 mol of dextrin chains, moles		Yield, %			Characteristics	
				Range of DCU yield	Proportion of gel and sol fraction in the resulting product		Gel fraction (CD)	Sol fraction (BD)
		Acid*	DCC		Gel fraction	Sol fraction	Maximum swelling degree in water**, %	Surface tension, mN/m
1***	2:1	0.75	1.65	96 ÷ 104	82 ± 3	18 ± 3	nw	–
2	3:1	0.5	1.1	93 ÷ 97	67 ± 4	33 ± 4	nw	–
3	4:1	0.375	0.825	99 ÷ 111	66 ± 4	34 ± 4	nw	43.2
4	6:1	0.25	0.55	89 ÷ 92	71 ± 4	29 ± 4	nw	–
5	8:1	0.188	0.413	94 ÷ 100	70 ± 4	30 ± 4	nw	43.0
6	10:1	0.15	0.33	78 ÷ 83	74 ± 4	23 ± 4	< 5	–
7	15:1	0.1	0.22	84 ÷ 88	82 ± 4	18 ± 4	382	43.2
8	20:1	0.075	0.165	75 ÷ 81	65 ± 4	14 ± 2	~ 700/d	–
9	30:1	0.05	0.11	99 ÷ 120	56 ± 4	12 ± 3	d	–
10	40:1	0.038	0.083	98 ÷ 110	42 ± 5	9 ± 3	d	43.1
11****	6:1	0.25	0.275	93 ÷ 96	28 ± 2	72 ± 2	d	–

*Moles of dibasic acid (2 –COOH)

***nw* hydrophobic, not water-wettable, *d* dispersible in water

***The experiment was carried out under conditions where general gelation of the reaction mixture is possible

****The experiment was carried out under the conditions of a half-deficiency of DCC, i.e. COOH:DCC = 1:1.1 mol/mol of dextrin

*****DCU yield for some samples exceeds 100% due to the formation of a stable complex with reaction products. In this case, a more accurate determination of the yield is difficult which is quite sufficient for the formation of the crosslinked network as there are 24–30 interactions with one carboxyl groups to the addition to these interactions effective for two groups (reaction V, Fig. 1). Such an interaction does not lead to branching of the macrochain, but introduces a strongly hydrophobic fragment into the macromolecule structure. This explains the fact that the gel fraction obtained under these conditions is a polymer of hydrophobic character incapable of swelling in water (Table 1).

When analyzing the sol fraction formation, the ratio of grafted acid residues to the loaded acid residues per one mol of dextrin (Fig. 4b) has a completely different character. With an increasing amount of Glu(St) molecules per one dextrin molecule, a constant decrease is observed for both total grafting efficiency and grafting efficiency by two carboxyl groups. At the same time, at (OH:COOH) ratio higher than 6:1, i.e., less than 11 Glu(St) molecules per one dextrin molecule, a significant excess of grafted Glu(St) residues over the loaded Glu(St) is observed. This applies to those grafted by one and two groups. Thus, one dextrin molecule contains 4–11 crosslinks and macromolecules of the sol fraction are highly branched. At the same time, the number of formed cycles is insignificant, since the polymer does not lose its ability to swell both in water and organic solvents.

At OH:COOH ratio below 3:1, i.e., 22 or more Glu(St) molecules per one dextrin molecule, the ratio of grafted to loaded molecules decreases sharply to 0.4 ÷ 0.5 for total grafting efficiency and lower than 0.1 for grafting by two groups. It leads to the formation of a completely different sol-fraction polymers. If at OH:COOH ratio higher 10:1 sol-fraction polymer are highly branched with a high degree of crosslinking but with a small number of inefficiently grafted Glu(St) residues, at the ratio below 3:1 sol-fractions consist of 2 ÷ 3 cross-linked dextrin molecules with a significant amount of grafted acid residues.

As mentioned above, DCU is insoluble in the reaction mixture, so the increasing turbidity of the reaction mixture corresponds to the rate of the Steglich reaction in the studied system. Figure 5 shows the dependence of the optical density (which is proportional to the DCU amount) of the reaction mixtures on the time. The given curves have an s-shaped character and three separate sections could be distinguished.

There is no change in the optical density in the first (initial) section of the curves. This is caused by the accumulation of the activated form of acid in the mixture according to the reaction I (Fig. 1). Increasing turbidity is observed in the second section as rapid precipitation of DCU occurs. The rate of DCU release at this stage significantly depends on the initial concentration of carboxyl groups which is confirmed by the data presented in Fig. 5. Figure 5a shows curves obtained at the same initial concentration of hydroxyl groups and different initial concentrations of carboxyl groups. The clear dependence of the reaction rate is observed while the same dependence is not visible in the Fig. 5b. This fact can be explained by the formation of the internal anhydride of N-derived glutamic acid (the reaction scheme is given in the Supplementary Information, fig. SI.4), and the concentration of hydroxyl groups does not affect this process. It should also be noted that the last stage is the longest. If the fast second stage is completed in 8 ÷ 12 min at a temperature of

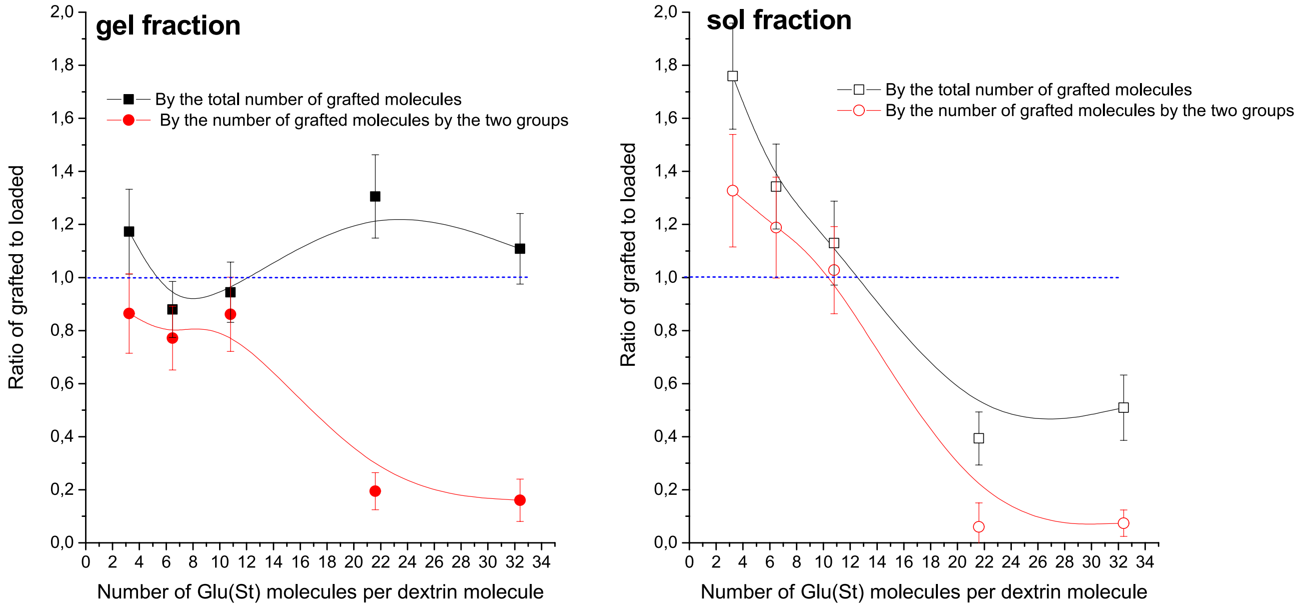

Fig. 4 Dependence of Glu(St) substitution efficiency by one or two carboxyl groups when forming sol (BD) and gel (CD) fractions of the product

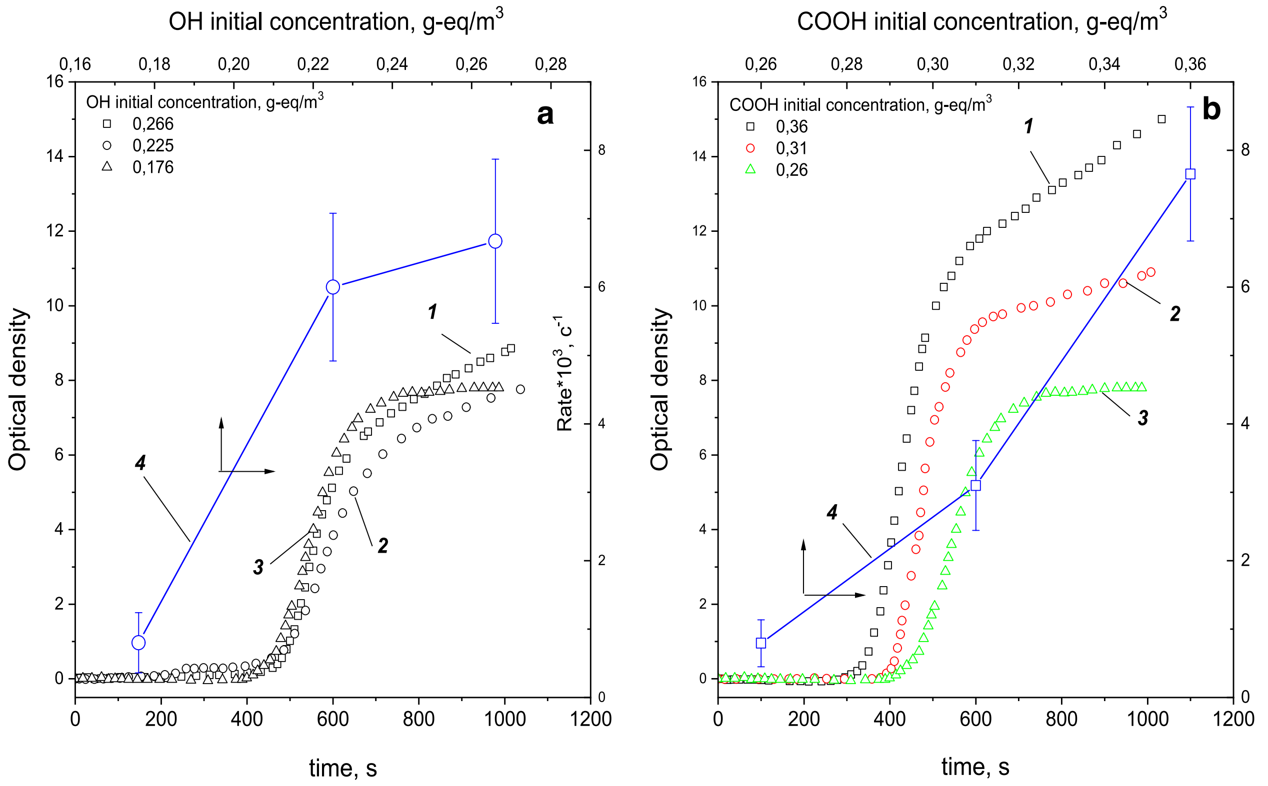

Fig. 5 The dependences of the optical density on the time for the reaction mixtures obtained at **a** the initial concentration of carboxyl groups of 0.260 g-eq/m^3 and different initial concentrations of hydroxyl groups, and **b** the initial concentration of hydroxyl groups of 0.176 g-eq/m.^{3} and different initial concentrations of carboxyl groups. Curve 4 in both figures corresponds to the rate of change in optical density in the ending sections of curves 1–3

288 K, the final stage lasts for 20 ÷ 22 h to reach the 95% of DCU. Meanwhile, the reaction rate at the final stage at this temperature does not significantly depends on the concentration of reactants.

Summarizing the obtained experimental data, it can be stated that BD polymers are dextrin molecules modified only via reactions I-III and possibly passivated by the reaction V (Fig. 1). As a result, these samples mainly contain modified dextrin molecules with a branched structure. For CD products the modification process occurs further including reaction IV in Fig. 1. In this case, macromolecules with a crosslinked structure are formed. Therefore, the properties of the above-described the products and their yields are determined by the crosslinking degree of polysaccharide macromolecules.

It was established that CD products do not possess surface-active properties but at a certain degree of crosslinking are capable of swelling in water and forming aggregate-resistant dispersions of microhydrogels. Samples of BD derivatives with a branched structure possess surface-active properties and are capable of forming self-stabilized aqueous dispersions.

Further, the properties of prepared aqueous dispersions were evaluated. As stated above, the properties of the resulting dispersions, their stability, and the morphology of the dispersed phase significantly depend on the synthesis conditions and the structure of the reaction products. The samples of the sol fraction possess significant surface-active properties. The surface tension of water dispersions decreases to 43–46 mN/m (Table 1) at the concentration of the dispersed phase above 0.1% with the formation of self-stabilized particles dispersion.

Figure 6 shows histograms of the particle size distribution of the dispersed phase for the sol fraction samples obtained at different reagent ratios. Figure 6a presents histograms obtained at a concentration of the dispersed phase of 0.03%. It should be noted that not all sol fraction samples form dispersed phase particles at this concentration range but only polymers obtained at OH:COOH ratio lower than 6:1. The hydrodynamic radius of such particles is 70 ÷ 170 nm. At the same time, the particle size depends on the nature of the sample. Particles formed by polymers which are obtained at a higher ratio, have a smaller radius.

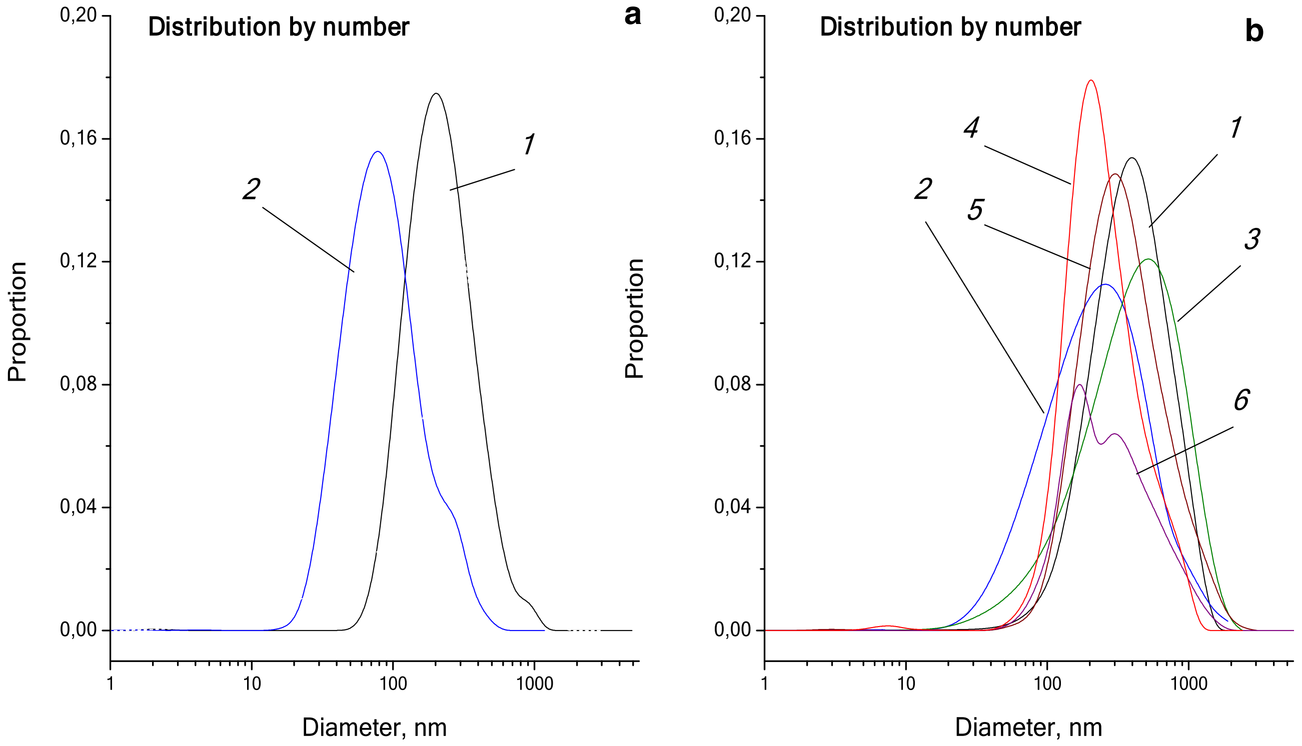

Fig. 6 Particle size distribution of the dispersed phase of the sol fraction. **a** particles at concentrations below CMC values; **b** particles at concentrations above the CMC values (1: sol fraction of sample 2; 2: sol fraction of sample 3; 3: sol fraction of sample 4; 4: sol fraction of sample 5; 5: sol fraction of sample 6; 6: sol fraction of sample 8). The characteristics of the samples are given in Table 1

Figure 6b shows histograms of particle size distribution at a concentration of 0.1%. At this concentration range all sol fraction samples form self-stabilized particles with a diameter of 160–500 nm. The size of dispersed phase particles does not depend on synthesis conditions, however the particle size at higher concentrations is significantly higher than the size of the same sample at lower concentrations.

An important feature of polymer materials tailored for medical and bio-medical purposes is their ability to interact with blood plasma proteins, in particular albumin. If such an interaction occurs, the polymers can be classified as promising materials for the drug delivery systems, in particular for the delivery of protein drugs and vaccines. To determine the possibility of this interaction, electrophoretic studies of the albumin mobility in a polyacrylamide gel in the presence of the dispersed phase particles of both gel and sol fractions were conducted. An albumin solution (control) and solution containing albumin and different amounts of the corresponding dispersion of modified polysaccharide were applied at the start of the electrophoregram. The particles of the dispersed phase did not have electrophoretic mobility and remained at the start of the plate. As a result, the sorption of the albumin on the particles also decrease of its mobility and its movement can be observed only after desorption from the particle surface and, accordingly, its path is significantly shorter than that of the albumin control solution. Thus, it can be concluded that the dispersed phase particles are capable of equilibrium sorption of albumin.

To estimate the amount of sorbed albumin, equal volumes of aqueous dispersions of modified polysaccharide particles of different concentration were titrated with conductometric sensor using 1% aqueous solution of albumin. The dependence of conductivity on the albumin amount in the solution in the presence of the particles of sample 7 gel and sol fractions (Table 1) is shown in Fig. 7a and b, respectively.

Figure 7a shows the dependence of the slope of the curve on the number of particles of the modified polysaccharide in the solution. The decrease in the slope of the curves shown in the figure compared to the titration curve of the blank sample is due to the binding of albumin by the dispersed phase particles. Sorption of protein on the particles surface reduces its concentration in the solution, leading to a decrease in the electrical conductivity of the solution compared to a solution with free albumin.

Since each of the titration curves has a constant slope within the entire range of albumin concentrations studied, it can be argued that equilibrium sorption of albumin occurs, i.e., the amount of bound protein is proportional to the amount of free albumin in the solution and within the limits of concentrations created during titration, the maximum

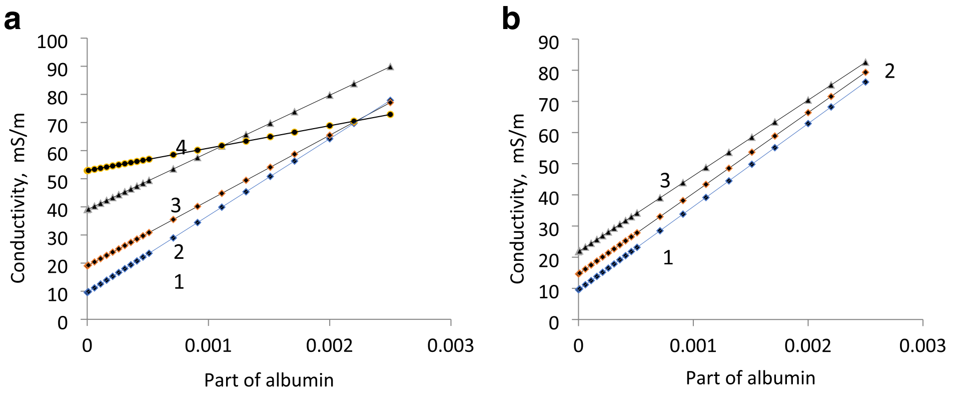

Fig. 7 Conductivity of the albumin solution in the presence of particles of sample 7 gel (**a**) and sol fractions (**b**): a—curve 1 corresponds to a blank sample (sample without particle dispersion), curves 2, 3, and 4: to titration curves of aqueous dispersions with concentrations of 0.25%, 0.75%, and 1.25%, respectively. b—curve 3 corresponds to a blank sample (a sample without particle dispersion), curves 1 and 2: to the titration curve of aqueous dispersions with concentrations of 0.51% and 0.17%, respectively

Table 2 Values of the interpolation coefficients of the curves shown in Fig. 7

Sample	Concentration of dispersed phase, %	Amount of polyester in the solution, g	Coefficients of linear interpolation	
			Tangent of the slope, b_i	Free coefficient, a_i
Gel fraction of sample 7 (Table 1)	0	–	10 ± 1	10 ± 1
	0.25	0.0375	19 ± 2	19 ± 2
	0.75	0.1125	39 ± 4	39 ± 4
	1.25	0.1875	53 ± 5	53 ± 5
Sol fraction of sample 7 (Table 1)	0	–	10 ± 1	10 ± 1
	0.17	0.0255	15 ± 1.5	15 ± 1.5
	0.51	0.0764	23 ± 2	23 ± 2

possible sorption by albumin particles was not achieved. Table 2 shows the established coefficients of linear interpolation. It was determined that linear interpolation coefficients are directly proportional to the amount of dispersed phase in the sample. This allows us to assert that the conductivity in the system is determined by the amount of dispersed phase and the concentration of dissolved albumin. The contribution to the conductivity of the dispersed phase is constant if its amount in the sample is not changed.

The afore mentioned facts allow using an additive scheme to derive the equation for the redistribution coefficient K for albumin between the dispersed phase and the dispersion medium:

$K = \left(1 - \frac{b_0}{b_i}\right) \cdot \frac{V}{m}$,where: V is the volume of the tested solution, ml; m is the weight of polyester particles in the investigated solution, g; b_0 and b_i are the tangents of the slope of the interpolation line for the curve of albumin without dispersion and the curve obtained by weighing the dispersion m_i. The adequacy of this equation is confirmed by the directly proportional dependence of the value $(1-b_0/b_i)$ on the concentration of the dispersed phase.

The tangent of the slope determines the numerical value of the redistribution coefficient. For the gel fraction sample, this value was 58 ± 7, which means a significant excess of the albumin amount in the dispersed phase particles over the amount of dissolved albumin in the dispersion medium.

The curves shown in Fig. 7b demonstrate that the sorption of albumin by the dispersed phase particles of the sol fraction is significantly lower than the sorption by the gel fraction. In this case, the value of the distribution coefficient estimated by the equation was 21 ± 2. Such a value allows us to conclude that the sol fraction of sample 7 (Table 1) exhibits almost three times less albumin sorption ability than its gel fraction. Considering that the surface area of the dispersed phase particles of the sol fraction is much larger (their size is smaller), and the sorption ability is smaller, it can therefore be concluded that the major part of albumin is absorbed by the gel fraction particles. It also can be assumed that for sol fraction albumin adsorption is predominant.

The data presented indicates that the obtained novel modified polysaccharides can be further exploited to create protein drug delivery systems.

Conclusions

In summary, optimized Steglich reaction was confirmed to be convenient synthetic approach for polysaccharide modification. Dextrin modification resulted in the formation of two different products with different properties. The peculiarities of the modification reaction, in particular, the range of synthesis conditions and the dependence of the yield and composition of the products on the initial reagents ratio, have been established.

It has been shown that the products with a branched structure obtained via polysaccharide modification with N-stearoylglutamic acid possess surface-active properties and are capable of forming self-stabilized aqueous dispersions, while products with a crosslinked structure do not have surface-active properties, but, at a certain degree of crosslinking, they are swellable in water and form aggregate-stable microhydrogel dispersions. Moreover, aqueous dispersions of the obtained polyesters are capable of sorption of water-soluble plasma proteins, and in the case of bovine serum albumin, the established redistribution coefficient ranges from 60 to 20, depending on the macromolecule structure.

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Data availability The authors declare that all of the data that support the findings of this study are available within the article and its Supplementary Information files or from the corresponding author upon reasonable request.

Declarations

Conflict of interest The authors have no competing interests to declare that are relevant to the content of this article.

Ethical approval This article does not contain any studies with human participants or animals performed by any of the authors.

Informed consent In this article, no patient care was involved.

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