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Research Article

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Polycomplexes based on *Bombyx mori* Chitosan and β -D-cyclodextrin with Hollow structure: Synthesis, Characterization, and Prolonged Drug Release

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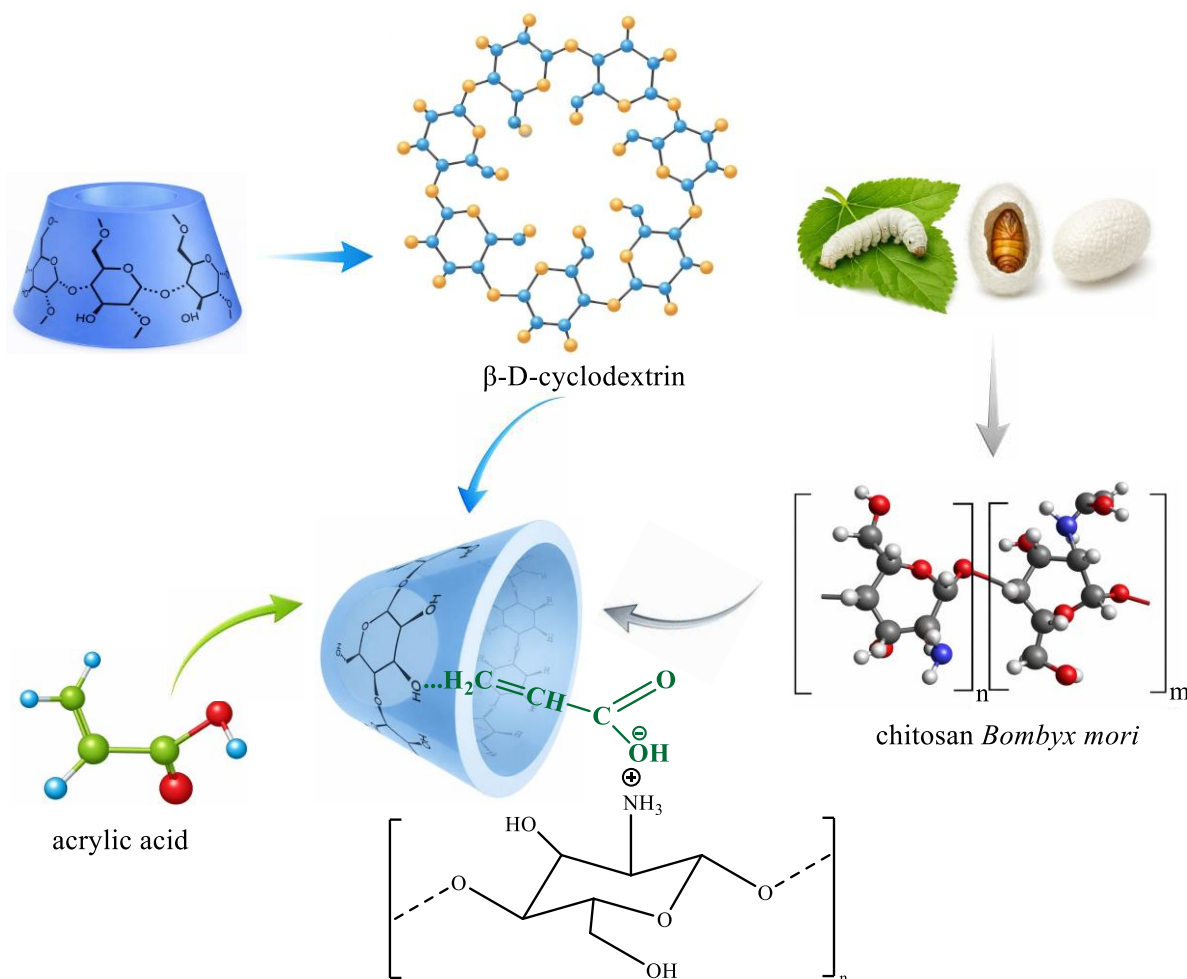
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Abstract. In this study, the synthesis method and characterization of nanocapsule structures obtained from polycomplexes consisting of *Bombyx mori* chitosan (CS) and acrylic acid-modified β -D-cyclodextrin (β -CD/AA) were investigated. The CS: β -CD/AA polycomplexes were synthesized by varying the mass ratios of the starting components, and it was found that at CS: β -CD/AA = 6:4 wt., the zeta potential value was relatively low (+1.7 mV), indicating the maximum interaction between β -CD/AA and the macromolecule. Transmission electron microscopy analysis showed that the resulting nanocapsules, which predominantly formed hollow structures through a "self-assembly" process, had sizes ranging from 70.9 to 237 nm. The nanocapsules demonstrated significant potential for biomedical applications, especially as drug delivery systems. Specifically, the encapsulation efficiency of furacilin was 51.99%, the loading capacity was 5.2%, and its sustained release reached 51.02% over 24 hours, highlighting the potential of the developed system for sustained drug delivery. These results indicate that the synthesized chitosan-based hollow nanocapsules hold significant potential for future biomedical and pharmaceutical applications.

Keywords: *Bombyx mori* chitosan, β -D-cyclodextrin, polycomplex, "self-assembly", nanocapsules, "hollow structure"

Graphical Abstract



Introduction

Chitosan is a biocompatible, biodegradable, and non-toxic natural polysaccharide widely studied for the development of drug delivery systems. These biological properties make chitosan (CS) a promising biomaterial for

use in drug delivery systems. Nanostructured chitosan and its derivatives are promising materials that have attracted the attention of researchers in recent decades. These systems provide improved biodistribution, increased specificity and sensitivity, and reduced pharmacological toxicity [1-3]. Furthermore, chitosan improves drug stability, bioavailability, and prolongation of action, as its nanoscale size can effectively overcome barriers in the nanoscopic range and increase permeability. Chitosan nanoparticles (NPs) can be used to transport both water-soluble and water-insoluble drugs [4-5]. The most common forms of chitosan NPs are nanospheres, nanocapsules, and nanotubes [6-8]. In recent years, particular attention has been paid to the development of technologies for producing nanocontainers [9, 10].

Nanocontainers have important advantages in the delivery systems of biologically active substances, as they protect drugs from chemical and enzymatic degradation and increase the effective solubility and bioavailability of poorly water-soluble compounds in physiological environments. Nanocontainers ensure the release of the active substance at the target site, increasing its effective concentration there and, as a result, enhancing the therapeutic effect. At the same time, they reduce overall systemic toxicity by limiting the distribution of the drug to non-target healthy tissues [11]. Since the effectiveness of drug delivery systems is directly dependent on the physicochemical properties of the active substance and the biological properties of the target tissue, the selection of carrier materials based on scientific evidence is of great importance [12].

The ability of nanodrugs to be delivered to the target is determined by the size, surface charge, surface modification, hydrophobicity, and high surface-to-volume ratio of particles. These properties are also important for reducing drug dosage and frequency of administration, thereby reducing the toxicity and side effects of chemotherapy drugs [13-14].

In the biomedical field, β -D-cyclodextrin, along with chitosan, is an important natural oligosaccharide that is widely used in drug delivery systems [15]. The β -D-cyclodextrin molecule has a cyclic structure with a hydrophilic outer part and a relatively hydrophobic inner part [16]. This cavity structure allows β -CD to accommodate hydrophobic bioactive compounds in its inner part and form an inclusion complex [15]. Inclusion complexes are formed mainly due to hydrophobic and van der Waals interactions, ensuring the effective distribution of the drug or bioactive substance in an aqueous environment within the inclusion complex without changing the true thermodynamic solubility of the substance. Consequently, the effective solubility and stability of the substance in a hydrophilic environment increase [17].

Simultaneously, the absence of ionizable functional groups in β -CD limits its direct electrostatic interaction with cationic polymers such as chitosan. Therefore, the chemical modification of β -CD, particularly its transformation into anionic derivatives by introducing carboxyl functional groups, is a promising approach for the formation of polycomplexes with chitosan.

Accordingly, nanocontainers based on citric acid-modified β -cyclodextrin and chitosan were obtained using the ionotropic gelation method and proved to be effective as a controlled drug delivery system for curcumin [18]. In another study, self-assembled oligochitosan/carboxymethyl- β -cyclodextrin macromolecular micelles exhibited high drug loading efficiency and prolonged release of paclitaxel, indicating that they are promising nanocarriers for anticancer drug delivery systems [19]. *Self-assembly* occurs through the association of molecules or through hydrogen bonds, van der Waals forces, ionic and hydrophobic interactions. The formation of polyelectrolyte complexes between chitosan and anionic modified β -cyclodextrin is an effective method for synthesizing nanocomplexes [20].

To expand the functional capabilities of β -CD, it has been proposed to chemically modify it, including by introducing carboxyl functional groups into the molecule [21]. The resulting anionic β -cyclodextrin derivatives enter into electrostatic interactions with the positively charged amine groups of chitosan, allowing the formation of polycomplexes CS/ β -CD: AA.

Thus, the objective of this research is to synthesize and characterize chitosan/ β -D-cyclodextrin:AA polycomplexes aimed at enhancing drug encapsulation efficiency and providing sustained release profile.

Materials and methods

Chitin was isolated in the laboratory from silkworm pupae readily available sources in Uzbekistan. Subsequently, chitosan was obtained from chitin using a previously established methodology. Briefly, pupae oil was first extracted, followed by demineralization, deproteinization, and bleaching steps to obtain purified chitosan. In this study, chitosan *Bombyx mori* with a molecular mass (MM) of 82 kDa and a DD of 86% was synthesized.

β -D-cyclodextrin (CAS No. 7585-39-9, Macklin, China) was used as a complexing component. Chemically pure acrylic acid (AA, Himedia Laboratories, India) was used as a source of anionic functional groups. Potassium dihydrogen phosphate (KH_2PO_4 , GOST 4198-75) catalyzed the reaction, and polyethyleneglycol (PEG-3000, LobaFeinchemie, Austria) served as an additive to stabilize the modification process. Furacilin (nitrofur, 20 mg/tablet, JSC "Irbis Chemical-Pharmaceutical Plant", Russia) was used as a model drug. Standard acetate buffer solution (pH 4.75) and phosphate-buffered saline (PBS, pH 7.4) were used in drug loading and release experiments.

Acetone (Chemical Invest, Russia), ethyl alcohol ($\text{C}_2\text{H}_5\text{OH}$, GOST-18300, Uzbekistan), and deionized water were used as precipitating and dissolving media. All chemical reagents were AR grade. Dialysis membrane MEMBRANE-CEL MD44 (14x100 CLR) was purchased from Sigma-Aldrich (St. Louis, MO, USA).

Dynamic light scattering (DLS) and Zeta potential

DLS was used to study the particle size distribution in solutions using a Photocor Compact-Z analyzer (Photocor, Russia) equipped with temperature-stabilized semiconductor laser: 635 nm, 25 mW and a viewing angle of 90°. The test solutions were stored at a thermostated temperature of 25±0.1 °C. The sample concentration was maintained at 0.05 mg/mL to minimize the aggregation effect. Each measurement was performed for 3 minutes and repeated at least three times.

Zeta potential of solutions were studied at 25 °C on the nanoparticle size and Zeta potential analyzer GD-919SZ (China). Prior to measurement, the instrument was calibrated according to the manufacturer's guidelines. 0.8 mL sample solution was carefully loaded into a clean, disposable cuvette, ensuring the absence of bubbles or contaminants. The measurement was conducted at a scattering angle of 90°, with the refractive index set to 1.590. Three independent measurements were performed for each sample, and the results were averaged to ensure reliability and reproducibility.

IR Fourier (FTIR) spectroscopy

FTIR spectroscopy in the range of 400-4000 cm⁻¹ using a Bruker INVENIO-S spectrophotometer (Bruker Optik GmbH, Ettlingen, Germany) was used to study the chemical structure of the samples and interactions. The samples were prepared in the form of tablets containing potassium bromide (150-250 mg KBr), and the solid sample (1.5-2 mg) was finely ground using a porcelain mortar, then placed in a mold and kept under vacuum conditions. After that, it was pressed under a pressure of 7×10⁸ Pa for 2 minutes.

Ultraviolet-visible (UV) spectroscopy

UV spectroscopic measurements were conducted for study of prolongation effect by using a SPECORD 210 (Germany) spectrophotometer, with a scanning range of 190 nm to 1000 nm, a slit width of 1 nm, and a scanning speed of nm per second. The accuracy of UV photometry was determined using potassium dichromate, in accordance with Ph. Eur. guidelines, with an accuracy of ± 0.01.

X-ray diffraction (XRD)

X-ray diffraction was performed using an XRD-6100 radiograph (Shimadzu, Japan). Diffraction patterns were recorded and monitored using a computer. The XRD-6100 device used CuKα radiation with a Ni filter, operating in the current and voltage mode on a lamp of 30 mA and 30 kV, respectively. The constant rotation speed of the detector was set at 4°/min with a range of scanning angles from 4 to 80° in 0.02° increments (w/2θ ratio). The synthesized samples were dried to constant mass and ground to a fine powder by grinding in an agate mortar. Then the resulting powder was placed evenly in the sample holder of the XRD, and the surface was carefully smoothed with a spatula. The prepared sample was mounted on the diffractometer, and XRD analysis was performed.

Scanning Electron Microscopy (SEM)

The SEM method was used to study the morphology of filmson a JSM-IT 210 scanning electron microscope (JEOL, Japan). During the measurements, an accelerating voltage (EHT-Extra High Tension) was used in the range from 5 to 15 kV, and the working distance was 10.8 mm. Images at various scales were obtained using the *In Touch Scope* software. Energy dispersion spectroscopy (EDS) was used to determine the elemental composition of the synthesized samples, which was used to obtain both spectra and images for mapping the elements. Before SEM analysis, the surface of the samples was coated with a carbon layer approximately 5-10nm thick using a ZD 400 high vacuumed evaporation system (Vigor Technologies Suzhou Co., Ltd., China) to increase ether electrical conductivity. For SEM analysis, 50 μL of 0.05% solutions of the samples were taken and diluted with 200 μL of distilled water. 3-4 μL of the resulting solution was dropped onto the surface of a glass substrate and dried using ultraviolet light to form a thin film. The dried film was then coated with a thin layer of graphite coating before analysis.

Transmission Electron Microscopy (TEM)

High-resolution TEM on a Talos F-200i transmission electron microscope from Thermo Fisher Scientific (USA) was used to determine the size of nanoparticles and the morphology of the film. The voltage range was 20-200 kV. TEM images were obtained using samples deposited on standard carbon-coated copper grids. EDS measurements were performed using a Bruker XFlash 6|30 energy-dispersive detector (Bruker, Germany). For TEM analysis, the sample suspensions were dispersed using ultrasonication for 3 minutes. Then, the suspensions were dropped onto a special carbon-coated copper grid and dried in a vacuum desiccator. After complete drying, the samples were placed in a transmission electron microscope and analyzed.

Determination of the total amount of carboxyl groups

The percentage of carboxyl groups in the β -D-cyclodextrin-acrylic acid derivative was determined [22-23]. 20 mL of a 0.5% solution of β -D-cyclodextrin-acrylic acid derivative was prepared and titrated with 20 mL of 0.1 N HCl solution in the presence of 1% phenolphthalein solution. The calculations were performed as follows (1):

$$N_2 = \frac{N_1 \cdot V_1}{V_2} = \frac{0.1 \cdot 20}{19} = 0.105 \quad (1)$$

- where N_1 and V_1 are the normal concentration and volume of the HCl solution;

V_2 is the volume of the NaOH solution.

N_2 is the normal concentration of the NaOH solution.

The volume of alkali (NaOH) consumed was determined when a 20 mL sample of a 0.5% solution of β -D-cyclodextrin-acrylic acid derivative was titrated.

$$X = \frac{(V_2 - V_1) \cdot N_2 \cdot 0.0045 \cdot 100}{g} = \frac{(0.3 - 0.02) \cdot 0.105 \cdot 0.45}{0.1} = 0.1323 \quad (2)$$

The amount of carboxyl groups (K.K) formed during the modification of β -D-cyclodextrin with acrylic acid was determined based on the following formula (3):

$$K.K = \frac{(V_2 - V_1) \cdot N_2 \cdot 4}{g} = \frac{(0.3 - 0.02) \cdot 0.105 \cdot 4}{0.1} = 1.176 \quad (3)$$

The β -D-cyclodextrin-acrylic acid=1:2 molar ratio derivative contains 13.23% (1.176 mol) carboxyl groups.

Determination of encapsulation efficiency (EE) and loading content (LC)

To load furacilin into the polycomplex, 40 mg of the CS: β -CD/AA=3:7 mass polycomplex was dissolved in 20 mL of acetate buffer solution with pH4.75 [24-25]. Then, 4 mg of furacilin was added to the total solution. The initial concentration of furacilin was 0.2 mg/mL (200 μ g/mL). The resulting mixture was intensively stirred at 500 rpm for 24 hours at 22°C. After that, the furacilin-loaded polycomplex was separated from the suspension by centrifugation at 11,000 rpm for 30 minutes at 4°C. Based on the amount of free furacilin in the resulting supernatant, the encapsulation efficiency (EE) and drug loading capacity (LC) were calculated using the following formulas [26-27]:

$$EE\% = \frac{\text{total amounts of drugs} - \text{amounts of free drugs}}{\text{total amounts of drugs}} \times 100 \quad (4)$$

$$LC\% = \frac{\text{total amounts of drugs} - \text{amounts of free drugs}}{\text{NPs weight}} \times 100 \quad (5)$$

Drug release studies

The release of furacilin from the polycomplex structure was studied by dialysis using a dialysis membrane [18, 28-29]. For this, 2.08 mg of furacilin-loaded CS: β -SD/AA=3:7 mass polycomplex was obtained and suspended in 5 mL of phosphate buffer (PBS, pH7.4). The resulting suspension was placed on a dialysis membrane, dropped into 50 mL of phosphate buffer solution and stirred at 37 °C at 100 rpm for 24 hours. Then, at intervals of 2; 4; 6; 12 and 24 hours, 2 mL aliquots were taken from the buffer solution in the beaker, and the same volume of PBS was added to the beaker instead. Then, the amount of furacilin released was determined using the UV-spectroscopy method and compared with the total amount of furacilin. The furacilin concentration was calculated based on a standard calibration curve constructed for PBS, pH7.4.

The amount of cumulative release was determined based on the following formula:

$$M_t = C_t \cdot V_0 + V \sum_{i=1}^{t-1} C_i \quad (6)$$

Here: M_t -cumulative amount of released furacilin;

C_t -furacilin concentration at time t ;

V_0 -volume of PBS (50 mL);

V -volume of the withdrawn sample (2 mL);

C_i -furacilin concentrations at previous time points.

The release percentage was calculated using the following equation:

$$\text{Release (\%)} = \frac{M_t}{M_{\text{loaded}}} \times 100 \quad (7)$$

where M_t is the cumulative amount of furacilin released at time t , and M_{loaded} is the total amount of furacilin loaded into the polycomplex.

Experimental part

Synthesis of β -D-cyclodextrin modified with acrylic acid (β -CD:AA)

For hydrophilization, 1.134 g of β -D-cyclodextrin was weighed and completely dissolved in 40 mL of bidistilled water at 50°C for 30 min, while continuously stirring in a water bath. Then, 1.71×10^{-2} g of PEG-3000 and

3.4×10^{-3} g of KH_2PO_4 as catalyst were added to the resulting homogenic solution. The reaction mixture was stirred for 15 minutes at 50 °C. Then, acrylic acid and β -CD were added in a 1:2 molar ratio, respectively. The reaction mixture was then stirred at 50°C for 1 hour at 500 rpm. PEG-3000 was used as an auxiliary stabilizer to ensure the homogeneity and uniform distribution of molecules in the reaction medium, as well as to reduce the risk of aggregation. Acrylic acid was used as a modifying agent to introduce -COOH groups into the β -CD molecule, thereby enhancing its hydrophilic properties, i.e., promoting its hydrophilization. The modified β -CD was isolated by precipitation in 80 mL of acetone and washed twice with 96% ethanol to completely remove unreacted reagents and auxiliary additives. It was then dried in a freeze-drier (Christ Alpha 1-4 LD plus) for 8 hours at -50°C until it reached a constant weight (Fig. 1).

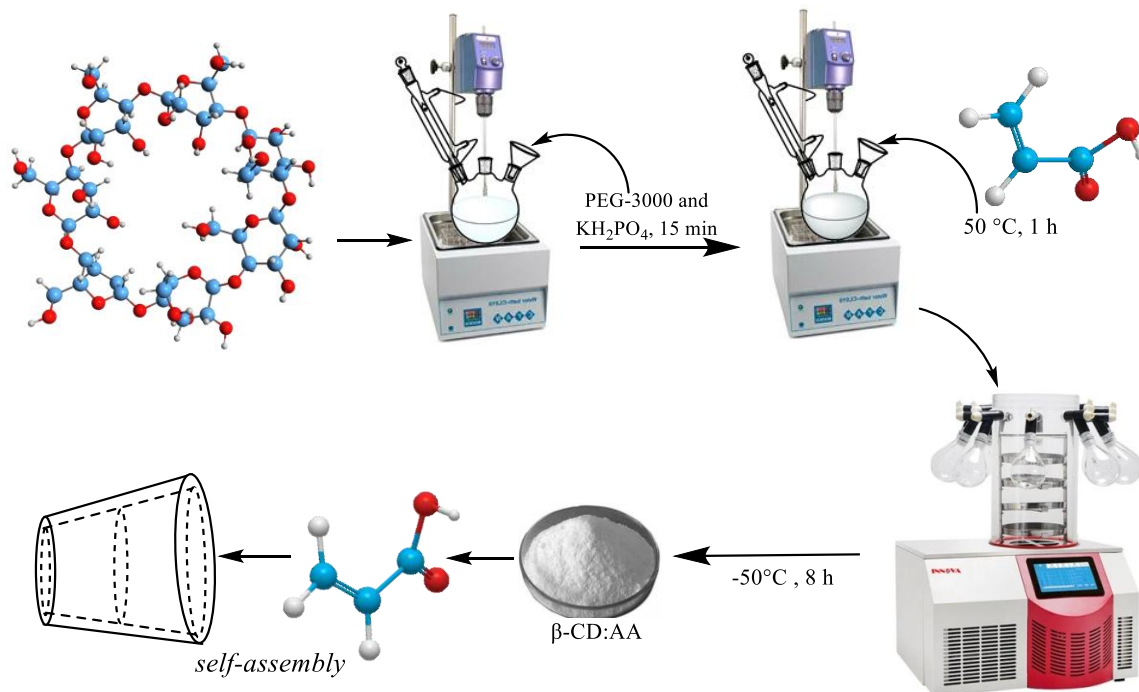


Fig. 1 Sequence of the β -D-cyclodextrin modification process with acrylic acid

Synthesis of CS: β -CD/AA polycomplexes

Polycomplexes based on CS and β -CD/AA were obtained by varying the mass ratio of the initial components from 1:9 to 9:1, respectively. For the synthesis of polycomplexes, a 0.5% solution of chitosan and a 0.5% solution of β -CD:AA in a buffer medium with a pH of 4.75 were prepared. The pH of the reaction system ensured the protonation of the chitosan amino groups, while the carboxyl groups of β -CD:AA were ionized, creating favorable conditions for the electrostatic interaction of the components. The β -CD:AA solution was added to the chitosan solution in mass ratios of 1:9 and 9:1, respectively. The resulting reaction mixture was vigorously stirred on a magnetic stirrer for 30 minutes at 500 rpm. Finally, the polycomplexes were isolated from the reaction mixture by precipitation with acetone, and then repeatedly washed with ethanol to remove unreacted components. The purified samples were lyophilized at -50°C for 8 hours (Table 2).

Results and discussion

According to the presented Fig. 1, by varying the molar ratios of the initial components, modifications of β -D-cyclodextrin with acrylic acid were obtained (Table 1).

Table 1 The influence of the ratio of the starting components on the synthesis of β -CD:AA. $t=50$ °C; $\tau=1$ h; $v=500$ rpm

No.	β -CD: $\text{CH}_2=\text{CHCOOH}$, mol	PEG-3000, mol	KH_2PO_4 , mol	-COOH, %	Reaction yield*, %
1	1:0.25	0.25	0.05	2.1	84 $\pm$ 0.85
2	1:0.5	0.25	0.05	4.7	79 $\pm$ 1.52
3	1:1	0.25	0.05	6.4	85 $\pm$ 1.64
4	1:1.5	0.25	0.05	9.4	86 $\pm$ 1.25
5	1:2	0.25	0.05	13.2	89 $\pm$ 1.32
6	1:4	0.25	0.05	12.0	87 $\pm$ 1.15

* the reaction yield was determined by the gravimetric method

The results in Table 1 show that at a molar ratio of β -CD:AA=1:2, respectively, the maximum content of carboxyl groups up to 13% in β -CD and the reaction yield up to 89% were achieved. However, further increasing

the amount of acrylic acid to β -CD:AA=1:4 resulted in a decrease in the modification efficiency and reaction yield; therefore, the molar ratio of β -CD:AA=1:2 was selected as the optimal ratio and used for further syntheses.

Table 2 The influence of the ratios of the initial components in the synthesis of polycomplexes; $t=30$ min, $v=500$ rpm, $pH=4.75$

No.	Samples	Mass ratio		Reaction yield, %
		Chitosan	β -CD:AA	
1	CS: β -CD/AA	1	9	60 $\pm$ 2.1
2	CS: β -CD/AA	2	8	68 $\pm$ 1.6
3	CS: β -CD/AA	3	7	90 $\pm$ 0.2
4	CS: β -CD/AA	4	6	75 $\pm$ 3.2
5	CS: β -CD/AA	5	5	79 $\pm$ 1.2
6	CS: β -CD/AA	6	4	82 $\pm$ 1.8
7	CS: β -CD/AA	7	3	96 $\pm$ 0.6
8	CS: β -CD/AA	8	2	84 $\pm$ 1.1
9	CS: β -CD/AA	9	1	86 $\pm$ 2.4

The results indicated that the polycomplexes CS: β -CD/AA=7:3 mass and CS: β -CD/AA=3:7 mass ratios resulted in a superior reaction yield from 96 $\pm$ 0.6% to 90 $\pm$ 0.2%, respectively compared to alternative ratios ranging.

DLS analysis results

a) Acrylic acid-modified β -D-cyclodextrin (β -CD:AA)

Solutions of β -CD:AA samples obtained at various molar ratios of β -CD and AA were studied using the DLS method (Table 3). It was found that solutions of all studied samples were polydisperse systems containing both micro- and nanosized particles. Significant changes in the particle size distribution were observed with increasing degree of modification. In particular, in the derivative prepared at a β -CD:AA=1:2 molar ratio, the proportion of nanoparticles was 63.2%, indicating a comparatively predominant formation of nanoparticles in this sample. In other ratios, an increase in the percentage of micron-sized aggregates was observed, due to particle agglomeration. It was revealed that the degree of modification of β -CD with acrylic acid affects its size and distribution.

Table 3 The influence of molar ratios of the initial components on the hydrodynamic diameter of particles of their modifications

No.	β -CD:CH ₂ =CHCOOH, molar ratio	$d_h, \mu m$	standard deviation, μm	d_h, nm	standard deviation, nm
1	β -D-cyclodextrin	19 (87.3 %)	8	88 (12.7 %)	7.4
2	1:0.25	74 (42 %)	22	72 (58 %)	19
3	1:0.5	14 (78 %)	4.6	204 (22 %)	42
4	1:1	22 (70.3 %)	9	112 (29.7 %)	19
5	1:1.5	20 (68.5 %)	5.8	114 (31.5 %)	24
6	1:2	22 (36.8 %)	3.6	188 (63.2 %)	22
7	1:4	28 (74.7 %)	19	210 (25.3 %)	25

Cyclodextrins are cyclic oligosaccharides composed of glucopyranose units arranged to form a hydrophilic outer surface and a hydrophobic internal cavity. The acrylic acid molecule contains a hydrophobic double bond (CH₂=CH-) and a polar carboxyl group (-COOH). It is assumed that the interaction between β -D-cyclodextrin and acrylic acid occurs through hydrogen bonds between the carboxyl groups of acrylic acid and the hydroxyl groups of β -CD or between water molecules by increasing the hydrophilicity of the β -CD molecule. The formation of nanoparticles in this system is related to the peculiarities of self-assembly processes during interactions between molecules. Specifically, the hydrophobic interactions between the functional groups of acrylic acid with the hydrophobic internal surfaces of β -D-cyclodextrin contribute to the aggregation of individual complexes, ultimately leading to the formation of nanoparticles by self-organization.

The standard deviation values indicate that, unlike native β -CD, solutions of its complexes with acrylic acid exhibit a moderately polydisperse particle size distribution, irrespective of the acid content.

b) Polycomplexes based on CS: β -CD/AA

The hydrodynamic diameters of the initial components and polycomplexes based on them were studied. The data demonstrate that the solutions of the starting components, both CS and β -CD/AA, are polydisperse systems and confirm the existence of micro- and nanoparticles in the solutions (Table 4).

Table 4 The influence of mass ratios of the initial components on the hydrodynamic diameter of particles their polycomplexes

No.	Samples	$d_h, \mu m$	standard deviation, μm	d_h, nm	standard deviation, nm
1	Chitosan	2.6 (11 %); 42 (41 %)	0.82 24	552 (48 %)	194
2	β -CD:AA=1:2 molratio	22 (37 %)	3.6	188 (63 %)	22

3	CS:β-CD/AA=7:3 mass ratio	15 (24%)	5.2	110 (21%) 346 (55 %)	22 130
4	CS:β-CD/AA=3:7 mass ratio	14 (32 %)	4.04	212 (68 %)	56

The results demonstrated that the ratio of the initial components significantly affects both the average particle size and their distribution. It was found that in solutions of polycomplexes with a mass ratio of CS:β-CD/AA=7:3 and CS:β-CD/AA=3:7, respectively, nanoparticles were formed with a relatively high percentage and uniform distribution. In the remaining ratios studied, there was an increase in the proportion of micron-sized aggregates, as well as an increase in the polydispersity of particle size. Therefore, from the standpoint of the formation of stable nanoparticles, mass ratios of 7:3 and 3:7 were chosen as suitable for further studies. These ratios correspond to systems in which either the cationic component (CS:β-CD/AA=7:3, mass.) or the anionic component (CS:β-CD/AA=3:7, mass.) prevails. This allowed a comparative assessment of the effect of the ratio of components on the morphological and physicochemical properties of the obtained polycomplexes.

Notably, the part of nanoparticles in the CS:β-CD/AA polycomplexes was predominantly nanoscale. In particular, the fraction of nanoparticles in the complex - CS:β-CD/AA=7:3 mass. was 76%, while in the complex - CS:β-CD/AA=3:7 mass., it was 68%. Compared to the initial components, the formation of the polycomplex contributed to a shift in the particle size distribution toward the nanoscale. Perhaps, this occurs during the formation of polycomplexes as a result of electrostatic interactions that lead to the compaction of macromolecules.

The obtained data show that in a CS solution, the forming micro- and nanoparticles are polydisperse, with standard deviations of 24 microns and 194 nm, respectively. However, in solutions of complexes CS:β-CD/AA, the particle distribution is moderately bimodal. The obtaining of polycomplexes, the standard deviation is low, perhaps, demonstrating the system's efforts to achieve stability.

Zeta potential results

To determine the zeta potential, 5×10^{-3} % solutions of chitosan and β-CD/AA were prepared in $\text{CH}_3\text{COOH} + \text{CH}_3\text{COONa}$ buffer (pH4.75). Each solution was stirred vigorously and filtered through a filter with a $d=0.45 \mu\text{m}$. Then, β-CD/AA and chitosan solutions were taken in mass ratios from 1:9 to 9:1, and the reaction mixture was magnetically stirred in a mixer at a speed of 500 rpm for 30 minutes. Then, 0.8 mL of each resulting polycomplex was taken, placed in a cuvette, and the zeta potential values of the solutions were determined. The zeta potential values of CS, β-CD/AA, and their based polycomplexes were recorded in the range from $\xi=+20$ mV to $\xi=+1.7$ mV (Table 5).

Table 5 Effect of CS:β-CD/AA mass ratios on zeta potential and mobility values

No.	Chitosan:β-CD/AA mass ratio	Zeta potential, mV	Mobility, $\mu\text{m} \cdot \text{cm}/\text{V} \cdot \text{s}$
1	Chitosan	+20	1.548
2	β-CD/AA	+4.1	0.324
3	1:9	+11.1	0.864
4	2:8	+8.4	0.667
5	3:7	+8.3	0.640
6	4:6	+8.6	0.677
7	5:5	+7.8	0.607
8	6:4	+1.7	0.136
9	7:3	+13.6	1.051
10	8:2	+8.5	0.648
11	9:1	+8.6	0.665

When titrating the CS solution with β-CD/AA, a gradual decrease in the positive values of the solution zeta potential and mobility indices was observed. In the complex obtained with a mass ratio of chitosan:β-CD/AA=6:4, the solution zeta potential value reached a minimum of +1.7 mV, which is probably due to the maximum interaction of the protonated amino groups in the chitosan macromolecule with the negatively charged $-\text{COO}^-$ groups in the β-CD/AA molecule.

Subsequent titrations showed an 8-fold increase in the Zeta potential value (from $\xi=+1.7$ mV to $\xi=+13.6$ mV) and a 7.73-fold increase in the Mobility value (from $0.136 \mu\text{m} \cdot \text{cm}/\text{V} \cdot \text{s}$ to $1.051 \mu\text{m} \cdot \text{cm}/\text{V} \cdot \text{s}$) of the solutions at a mass ratio of chitosan:β-CD/AA=7:3. In subsequent measurements, the average electrokinetic potential value was $\xi=+8.5 \pm 0.5$ mV, and the Mobility value was $0.6565 \pm 0.0085 \mu\text{m} \cdot \text{cm}/\text{V} \cdot \text{s}$.

This phenomenon most likely indicates the presence of additional β-CD/AA molecules not bound to CS in the system, which are formed when the positively charged amino groups of CS are blocked by the negatively charged carboxyl groups of β-CD/AA. Thus, the electrokinetic parameters of the studied solutions obtained at different mass ratios indicate that at the mass ratio of Chitosan:β-CD/AA=6:4, CS and β-CD/AA are maximally combined with each other through ionic interactions. These results are in good agreement with the literature data [29].

FTIR spectroscopy

IR spectroscopic analysis showed significant changes in the spectral characteristics of β -CD modified with acrylic acid, confirming the presence of hydrogen bonds between them, leading to an increase in the hydrophilicity of β -CD molecules (Fig. 2).

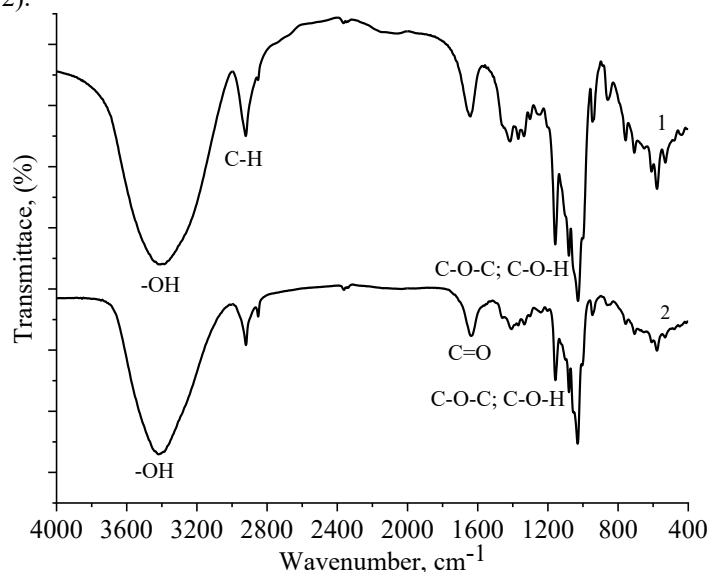


Fig. 2 FTIR spectra of the samples: 1) β -CD; 2) β -CD:AA=1:2 molar ratio

The results of the IR spectral analysis are presented in Table 6. Also, according to the literature, characteristic absorption bands of PEG-3000 used as a stabilizer in the synthesis process were observed. In particular, the peak in the region of 3440-3450 cm^{-1} corresponds to the stretching vibrations of hydroxyl (-OH) groups, the peaks at 2880-2950 cm^{-1} correspond to the symmetric and asymmetric C-H stretching vibrations of methylene (-CH₂-) groups in the PEG-3000 chain, and the peaks at 1150, 1105 and 1060 cm^{-1} correspond to the stretching vibrations of ether (C-O-C) bonds in the chain [30-31].

Table 6 Absorption bands identified in the IR spectrum of β -D-cyclodextrin and their interpretation

No.	ν , cm^{-1}	Functional groups	Type of vibration	Note
1	3390	-OH	stretching vibration	numerous hydroxyl groups with strong H-bonding
2	2925	C-H	stretching vibration	attributed to the C-H bonds of the glucopyranose ring
3	1645	C-O-H	bending vibration	water molecules located in the cavity and bound within the structure
4	1163	C-O-H	stretching vibration	characteristic of secondary alcohol groups
5	1035	C-O-C	stretching vibration	ether bonds characteristic of β -cyclodextrin
6	857	cyclic ring	bending vibration	internal vibration of the β -cyclic structure

The FTIR spectrum of β -CD agrees with and confirms the data reported in the literature [32-35]. The FTIR spectrum of acrylic acid was obtained from the SDBS (Spectral Database for Organic Compounds, National Institute of Advanced Industrial Science and Technology, Japan) database (CAS No. 79-10-7), with characteristic absorption bands shown in Table 7.

Table 7 Absorption bands identified in the IR spectrum of acrylic acid and their interpretation

No.	ν , cm^{-1}	Functional groups	Type of vibration	Note
1	3300-2500	-OH	stretching vibration	characteristic of the carboxyl group, broadened due to strong H-bonding
2	2950-2850	C-H	stretching vibration	C-H bonds of the chain
3	1720-1700	-C=O	stretching vibration	The main characteristic peak of carboxylic acids
4	1635-1610	C=C	stretching vibration	confirms the presence of a double bond in acrylic acid
5	1290-1210	-C-O	stretching vibration	C-O bond of the carboxyl group

Table 8 Absorption bands identified in the IR spectrum of β -D-cyclodextrin modified with acrylic acid and their interpretation

No.	ν , cm^{-1}	Functional groups	Type of vibration	Note
1	3418	-OH	stretching vibration	hydrogen bonding between the -OH groups of β -cyclodextrin and the -COOH group of acrylic acid
2	2920	C-H	stretching vibration	aliphatic C-H bonds of the β -cyclodextrin ring
3	1637	C-O-H; -C=O	stretching vibration	The vinyl fragment and -COOH group of acrylic acid are preserved
4	1157	C-O-C	stretching vibration	glycosidic bonds of β -cyclodextrin
5	1099-1079	C-O	stretching vibration	cyclodextrin ring and structure

A comparative analysis of the IR spectra revealed that the interaction between β -cyclodextrin and acrylic acid occurred through the formation of strong hydrogen bonds and ion-dipole interactions. A bathochromic shift of the C=O stretching peak, characteristic of the carboxyl group, in the IR spectrum of the modified dextrin confirms the hydrogen-hydrogen bond between acrylic acid and the hydroxyl groups of β -cyclodextrin. The absence of new spectral peaks characteristic of ester bond formation indicates that the esterification reaction did not occur. The preservation of vibrations corresponding to the vinyl fragment of acrylic acid indicates that the double bond structure was not broken during the interaction. Furthermore, the modification promotes an increase in the degree of hydrophilization of dextrin molecules.

X-ray diffractograms

The X-ray diffraction pattern of β -CD shown in Fig.3 was found to be consistent with the data in the JCPDS database, No. 00-066-1370 and No. 00-032-1626 (Fig.3) [17, 36].

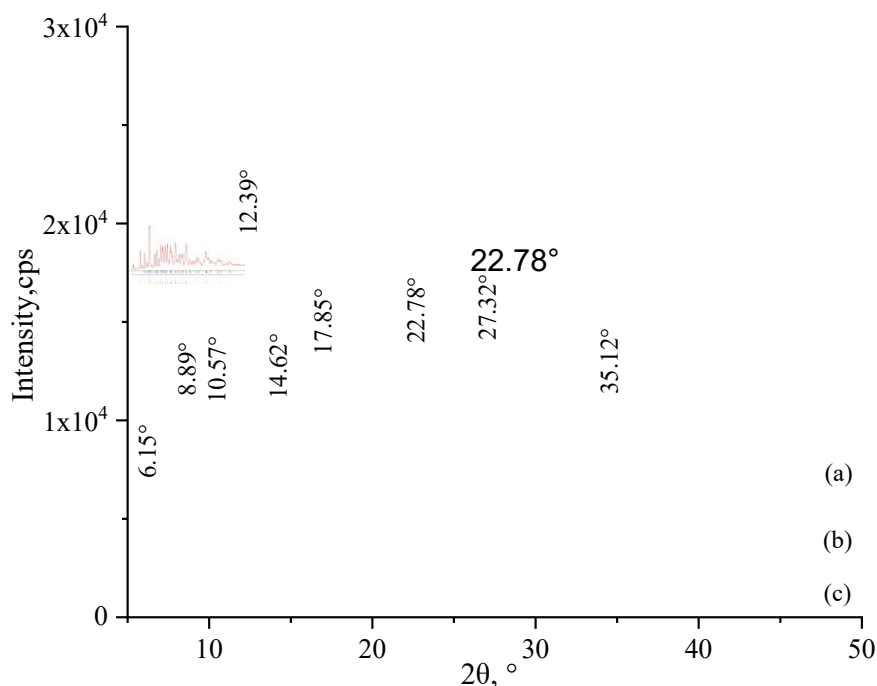


Fig. 3 X-ray diffraction pattern of the β -D-cyclodextrin sample: (a) diffractogram of the β -D-cyclodextrin sample; (b) reference diffraction peaks No.00-066-1370 and No.00-032-1626 from the JCPDS database; (c) intensities of the reference diffraction peaks

β -D-cyclodextrin has a crystalline macrocyclic molecular structure, which is confirmed by the diffraction peaks observed at $2\theta = 6.15^\circ, 8.89^\circ, 10.57^\circ, 12.39^\circ, 14.62^\circ, 17.85^\circ, 22.78^\circ, 27.32^\circ$, and 35.12° . Similar diffraction peaks have also been reported in the literature for β -CD, indicating that it has a semi-crystalline nature and belongs to the monoclinic crystal system [37-38].

The X-ray diffraction pattern of CS *Bombyx mori* shows two distinct reflections at $2\theta = 10.63^\circ$ and $2\theta = 19.92^\circ$, which correspond to the crystallographic patterns (020) and (110), respectively (Fig. 4). The main diffraction peak at $2\theta = 19.92^\circ$ corresponds to ordered regions stabilized by intermolecular hydrogen bonds and indicates the predominance of -NH₂ groups, while the comparatively weak reflection at $2\theta = 10.63^\circ$ is associated with residual acetamide groups remaining after deacetylation. The coexistence of these reflections with a broad amorphous background indicates that the obtained CS has a semicrystalline structure [35, 39].

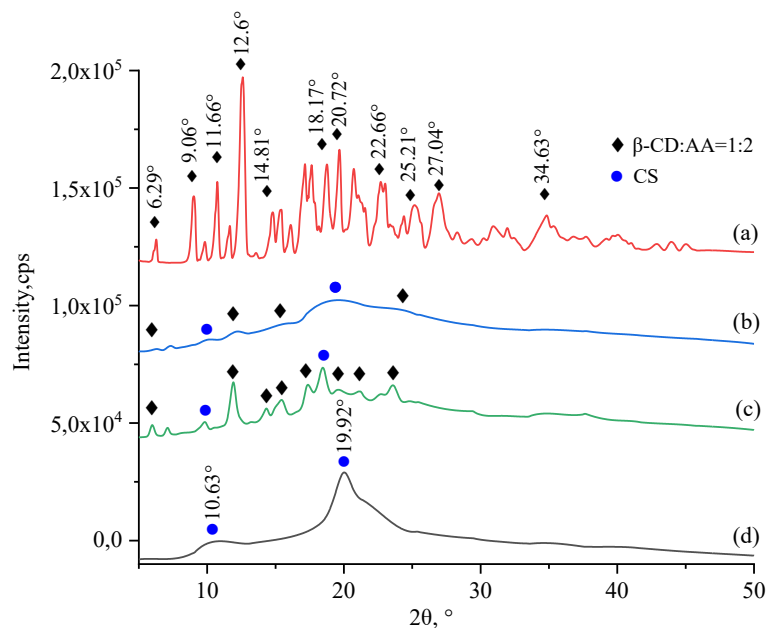


Fig. 4 X-ray diffractograms of samples: (a) β -CD:AA=1:2, mol; (b) CS: β -CD/AA=7:3, mass.; (c) CS: β -CD/AA=3:7, mass.; (d) CS

XRD results for the β -CD:AA=1:2 mol modification indicate its crystalline nature. The observation of intense diffraction peaks at $2\theta=6.297^\circ$, 11.659° , 12.605° , and 18.172° in the diffraction pattern indicates that the elements of crystalline order characteristic of β -CD were preserved even after the modification process. Crystallite sizes, calculated based on the width of the diffraction peaks, were found to be in the range of 7.2-56.9 nm.

The results of X-ray diffraction analysis (Table 9) showed that the polycomplex with a mass ratio of CS: β -CD/AA=7:3 has a semicrystalline structure. Pronounced peaks in the diffraction pattern were observed at angles of $2\theta=12.16^\circ$, 18.69° , 20.07° , 36.04° , and 38.3° . The peak at a small angle of 12.16° ($d=7.27$ Å, crystallite size 145 Å) represents ordered segments of the substance. For comparison, the peaks at 18.69° and 20.07° ($d=4.744$ - 4.420 Å, crystallite sizes 21-59 Å) represent a semi-amorphous phase formed as a result of the interaction of chitosan and β -D-cyclodextrin-acrylic acid. Perhaps, the broad peaks at high angles of 36.04° and 38.3° ($d=2.490$ - 2.348 Å, size 17-30 Å) confirm the presence of short-range ordered crystallite domains in the nanoscale.

Table 9 Crystal pattern parameters of initial components and their polycomplexes

No.	$2\theta, ^\circ$	$d, \text{\AA}$	(FWHM) $\beta, ^\circ$	(Size) L, nm
Chitosan				
1	10.63	8.32	3.30	2.52
2	19.92	4.45	2.96	2.84
β -CD:AA=1:2, mol				
3	6.297	14.03	0.252	32.9
4	11.66	7.58	0.183	45.5
5	12.60	7.02	0.43	19.27
6	18.17	4.88	0.15	56.9
7	34.63	2.59	1.09	7.9
8	39.91	2.26	1.22	7.2
9	40.97	2.20	0.21	43.0
CS: β -CD/AA=7:3, mass				
10	12.16	7.27	0.57	14.5
11	18.69	4.74	1.42	5.9
12	20.07	4.42	3.9	2.14
13	36.04	2.49	2.9	3.0
14	38.30	2.35	4.9	1.8
CS: β -CD/AA=3:7, mass				
15	5.98	14.8	0.36	233
16	7.07	12.5	0.31	265

17	11.9	7.43	0.51	163
18	21.2	4.20	0.37	230
19	36.1	2.49	0.87	101

Overall, the sample with a CS:β-CD/AA=7:3 mass contained both amorphous and crystalline phases. This confirms the formation of a nanoscale semicrystalline structure during modification. The X-ray diffraction spectrum of the complex with a mass ratio of 3:7 showed distinct diffraction peaks at $2\theta=5.98^\circ$, 7.07° , 11.90° , 21.15° , and 36.09° . The peaks at 5.98° and 7.07° are characteristic of the β-CD crystal lattice, indicating the preservation of the β-CD crystal structure in the complex. The peak observed at 11.90° suggests the formation of hydrogen bonds between CS and β-CD. The peak at 21.15° belongs to AA, indicating the presence of partially ordered AA structures in the complex. The peak at 36.09° indicates partial ordering in the amorphous part of the chitosan base.

To summarize, X-ray diffraction results show a broadening and decrease in intensity of characteristic crystalline peaks for the CS and β-CD/AA systems, which is atypical for a mechanical mixture. The observed changes indicate intermolecular hydrogen interactions.

SEM

The morphology of the synthesized samples was examined to measure particle size and shape, which are important for evaluating the effectiveness of the modifications. The results of the SEM study show that the surface morphology of the chitosan's film has a dense and relatively homogeneous structure (Fig. 5 (a)). In addition, the morphology of β-cyclodextrin modified with acrylic acid was investigated (Fig. 5 (b-d)). Micrographs obtained at different scales show that the modification process leads to significant morphological changes in the surface.

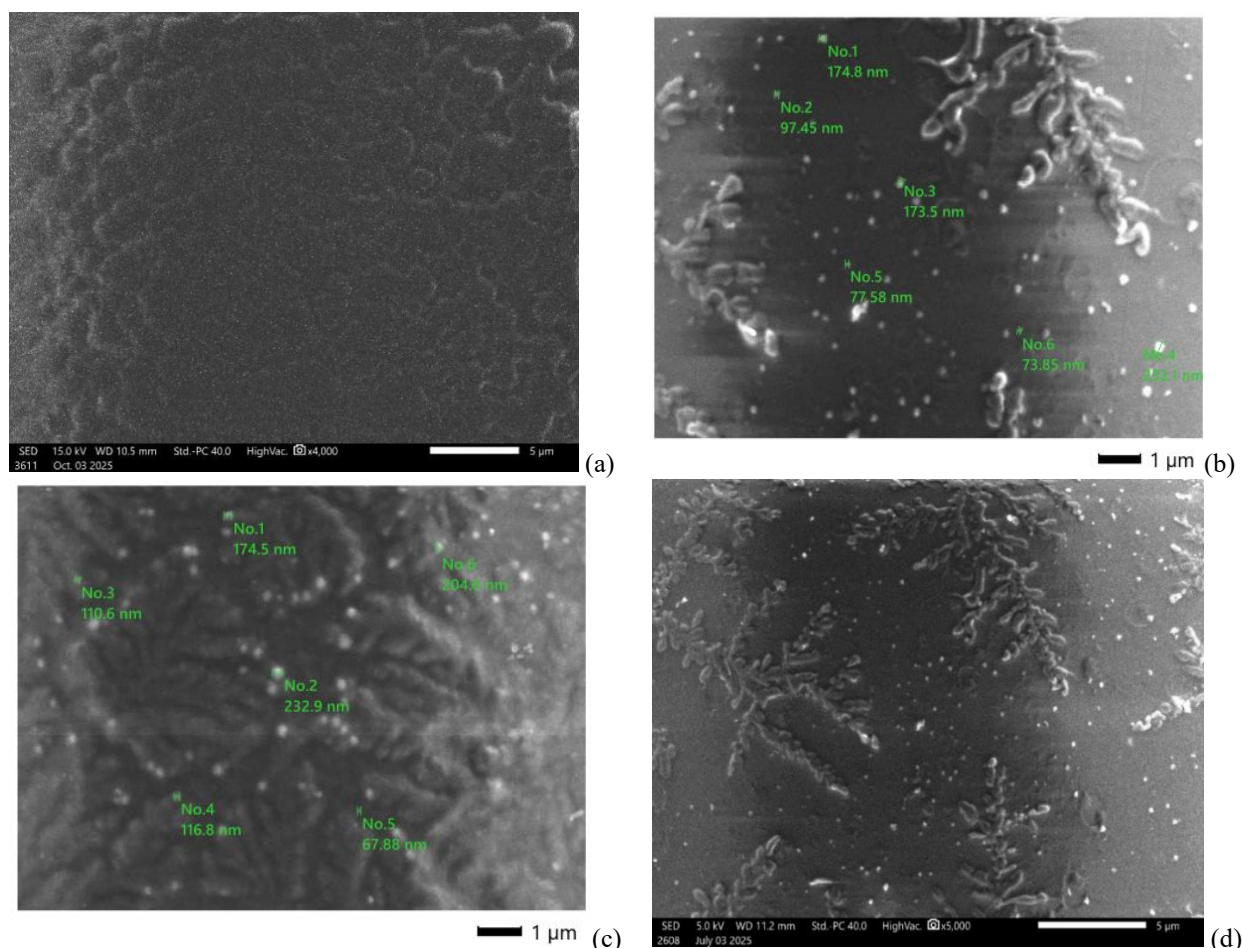


Fig. 5 SEM micrographs of morphology of the chitosan (a) and β-CD:AA (b, c, d) surfaces. (a) scale: $5\mu\text{m}$ ($\times 4,000$); (b) scale: $5\mu\text{m}$ ($\times 5,000$); (c-d) scale: $1\mu\text{m}$ ($\times 10,000$)

For example, on the surface of the β-CD:AA=1:2 mol sample at a $1\mu\text{m}$ scale (c), uniformly distributed spherical particles ranging in size from approximately 70 nm to 230 nm were detected. However, small aggregates were observed in some areas. This is likely due to the nanostructures forming during the modification process as a result of intermolecular hydrogen interactions between β-D-cyclodextrin and acrylic acid. Micrographs obtained at a $5\mu\text{m}$ scale clearly show branched aggregate structures consisting of clusters and spherical nanoparticles located on

their surfaces (d). This morphology indicates that the nanoparticles first formed and then, through their mutual association, formed into large aggregates.

The dendritic structures observed in the micrographs are likely due to nonequilibrium growth and aggregation of particles due to intermolecular interactions between β -cyclodextrin and acrylic acid, which is characteristic of the aggregation mechanism. SEM micrographs of the polycomplexes CS: β -CD/AA=7:3 mass. and CS: β -CD/AA=3:7 mass, respectively, were obtained (Fig. 6-7).

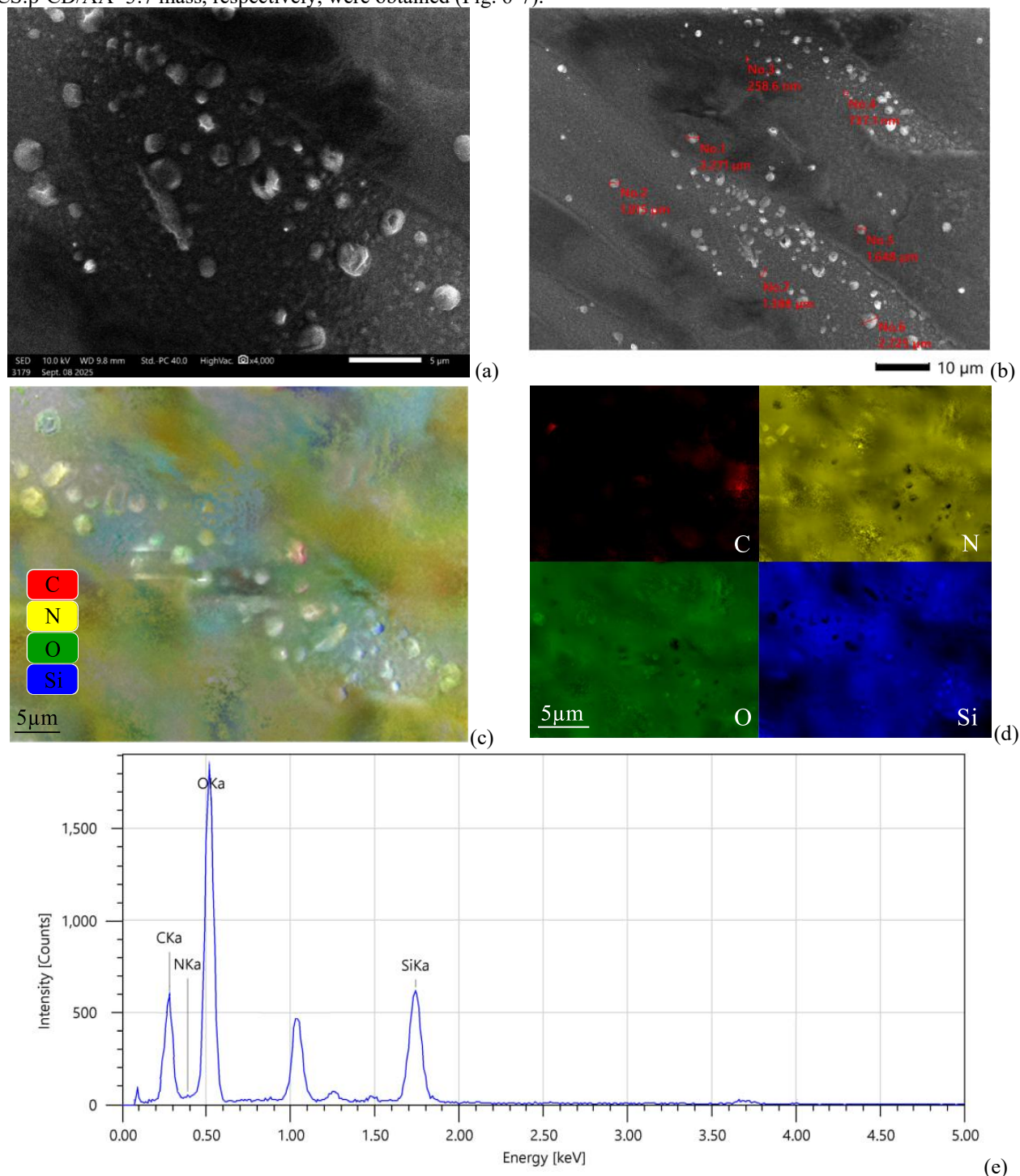


Fig 6 SEM microphotographs of the polycomplex CS: β -CD/AA=7:3 mass. (a-b), composite elemental mapping (c-d) and their EDS spectra (e)

It should be noted that upon obtaining the polymer complex, the morphology of the sample differs radically from that of the original components, both chitosan and β -CD/AA. The results indicate that the interaction of chitosan with the carboxyl-containing modification of β -D-cyclodextrin results in the formation of micro- and nanocapsules in the size range from 259 nm to 2.725 μ m. «Hollow» structures are likely formed through the «self-assembly» technique. The presence of nitrogen atoms in the composite elemental mapping indicates hydrophilization of β -D-cyclodextrin with chitosan macromolecules. The presence of silicon atoms in the EDS spectrum is apparently due to the use of a glass substrate when obtaining the SEM image.

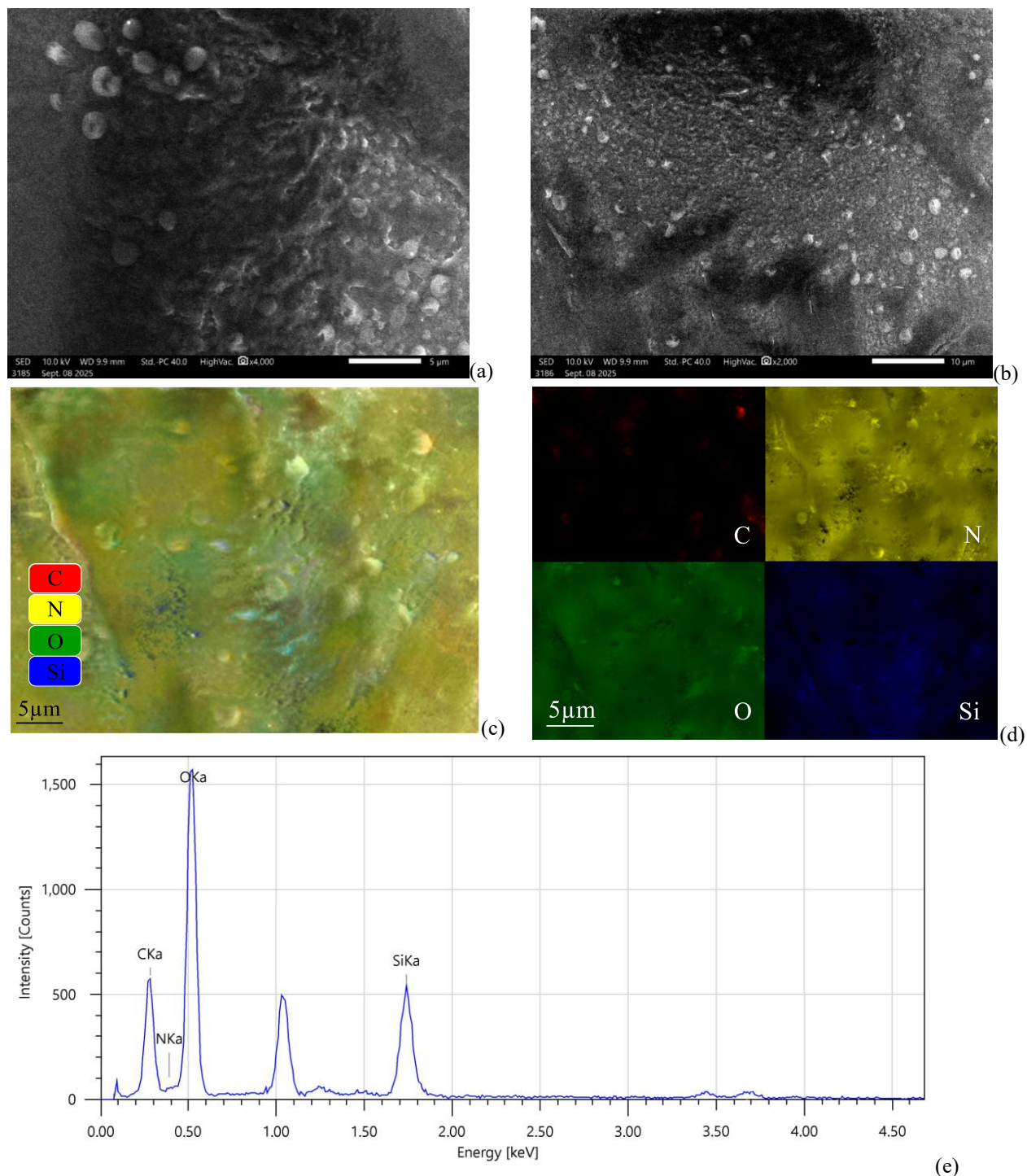


Fig. 7 SEM microphotographs (a-b), composite elemental mapping (c-d) and EDS spectra (e) of the CS:β-CD/AA=3:7 mass polycomplex

Therefore, the results show that the introduction of β-CD:AA into the CS macromolecule caused significant changes in the surface morphology of the original polymer. The morphology of the polycomplexes obtained at both mass ratios (7:3 and 3:7) differed from that of the original components. According to the micrographs, the nanoparticles were relatively homogeneously distributed throughout the matrix, and in some areas, small aggregates formed due to their dense arrangement. Instead of the previously characteristic cluster aggregates and spherical nanoparticles, the micrographs revealed structures with low electron density in the central region, indicating the formation of shell-like, i.e., "hollow" nano- and microcapsules. These results are consistent with the data of other authors [28,40-41].

To sum up, SEM micrographs will reveal the formation of chitosan/β-cyclodextrin-AA-based nanocapsules, characterized by a distinct nanoscale structure. These nano- and microstructures will serve as evidence of the successful implementation of this approach and will facilitate the assessment of their potential for application in targeted drug delivery. The formation of such nanostructures allows the use of such polycomplexes as nanocontainers for the transport of biologically active substances.

TEM

The morphological features of the β -CD:AA surface were examined through transmission electron microscopy at multiple magnifications, revealing microstructural modifications at detailed resolutions, namely, the formation of structural elements that differ from the initial reaction components, as well as a porous structure, confirming the effective formation of nanocapsules(Fig.8).

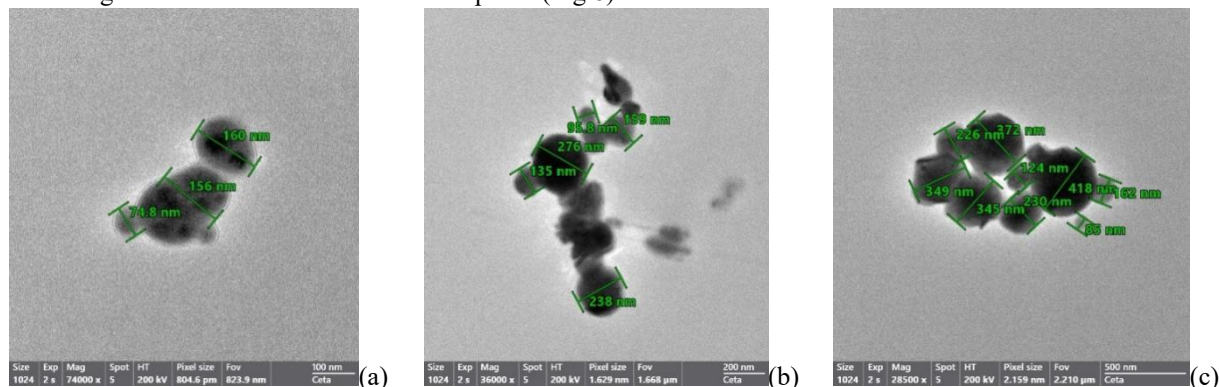


Fig. 8 TEM micrographs of β -CD: AA=1:2, mol: scale: (a) 100 nm; (b) 200 nm and (c) 500 nm

TEM micrographs of β -CD:AA reveal spherical nanoparticles across a wide range of sizes ($d = 75$ -420 nm) in the surface morphology. Aggregates formed by particle agglomeration were also observed. Elemental analysis and EDS spectra revealed that the composition of the β -CD:AA modifications consists of 84.53% C, 1.5% N, and 13.97% O, corresponding to the chemical composition of β -CD:AA (table 10). Additionally, the surface morphology of the CS: β -CD/AA polycomplexes was examined by TEM (Fig. 8).

Table 10 TEM EDS elemental analysis of β -CD: AA=1:2, mol

Z	Element	Family	Atomic Fraction (%)	Atomic Error (%)	Mass Fraction (%)	Mass Error (%)	Fit Error (%)
6	C	K	87.77	1.93	84.53	2.38	1.13
7	N	K	1.34	0.28	1.5	0.32	7.52
8	O	K	10.89	1.95	13.97	2.41	1.02

A micrograph of the surface morphology of the CS: β -CD/AA=7:3 mass. polycomplexes shows the formation of spherical nanoparticles ranging in size from 92 nm to 170 nm (Fig. 9).

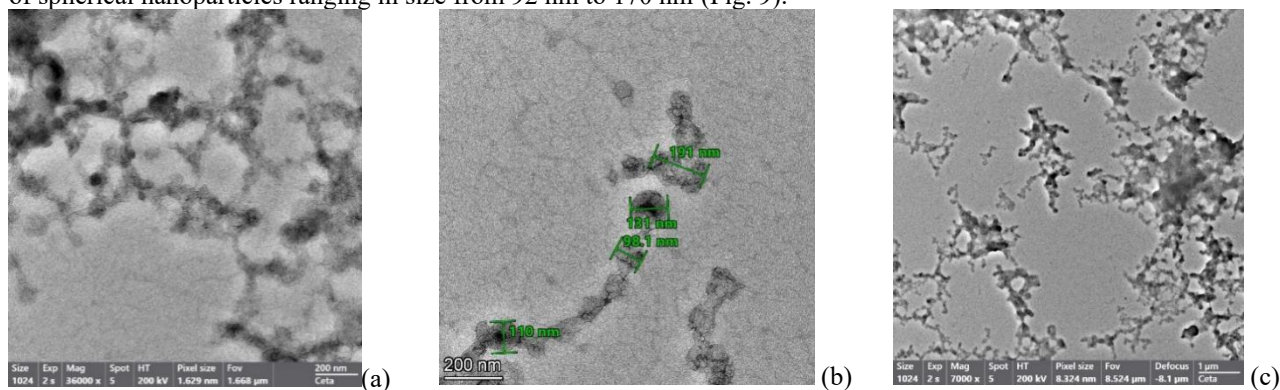


Fig. 9 TEM micrographs of CS: β -CD/AA=7:3, mass. polycomplexes obtained self-assembly: 200 nm (a); 50 nm (b);500 nm (c) and 1 μ m (d)

The spherical shape of the particles confirms that their structure is stable. A TEM micrograph reveals that in some regions, the particles have a chain or cluster shape. This is likely due to interactions between the particles. The presence of Si, S, and Al in the EDS spectrum is related to the chemical composition of the substrate on which the film was obtained (Table 11).

Table 11 TEM EDS elemental analysis of CS: β -CD/AA=7:3, mass

Z	Element	Family	Atomic Fraction (%)	Atomic Error (%)	Mass Fraction (%)	Mass Error (%)	Fit Error (%)
6	C	K	71.86	2.98	60.13	3.33	2.96
8	O	K	18.06	3.01	20.13	3.32	1.08
13	Al	K	0.31	0.07	0.58	0.13	7.96
14	Si	K	9.61	1.72	18.81	3.05	0.72
16	S	K	0.16	0.04	0.36	0.08	13.09

The formation of new structures has been observed, indicating interactions between chitosan and β -D-cyclodextrin within the capsules, which contribute to their stability. It was found that the structure of the CS/CD

polycomplexes exhibits a porous architecture and a well-developed, homogeneous surface compared to unmodified β -D-cyclodextrin.

The surface morphology of the CS: β -CD/AA=3:7 mass. polycomplexes films were also studied (Fig. 10).

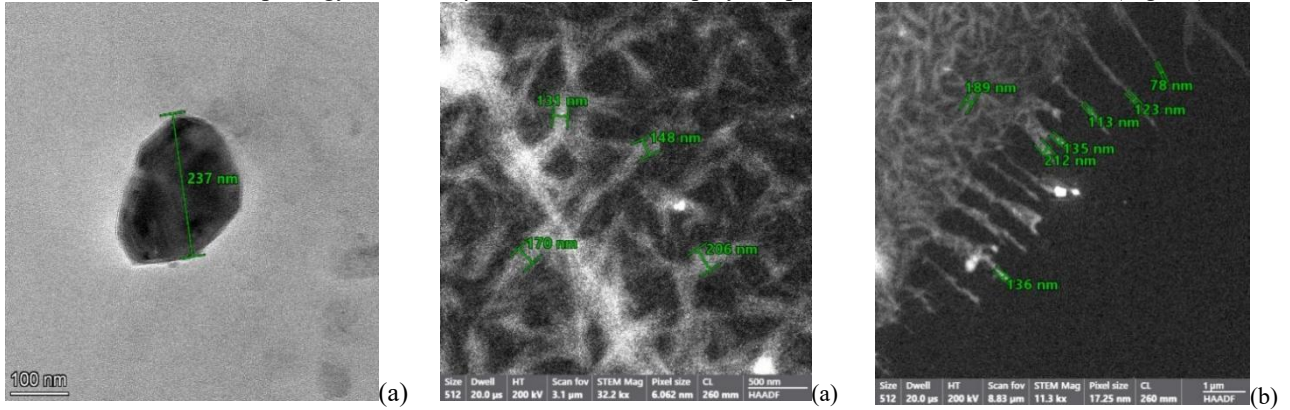


Fig. 10 TEM micrographs of CS: β -CD/AA=3:7, mass. polycomplexes obtained self-assembly: 100 nm (a); 500 nm (b) and 1 μ m (c)

It should be noted that the surface morphology of the CS: β -CD/AA=3:7 mass. and CS: β -CD/AA=7:3 mass. Polycomplexes are identical. However, with a decrease in the amount of chitosan in the polycomplex, an increase in the size polydispersity of the spherical nanoparticles is observed: $d=71$ -237 nm. The fusion of the spherical particles formed relatively large aggregates. The presence of Si in the EDS spectrum is related to the chemical composition of the substrate from which the film was obtained (Table 12).

Table 12 TEM EDS elemental analysis of CS: β -CD/AA=3:7, mass

Z	Element	Family	Atomic Fraction (%)	Atomic Error (%)	Mass Fraction (%)	Mass Error (%)	Fit Error (%)
6	C	K	69.56	2.99	59.26	3.17	0.34
8	O	K	19.53	3.16	22.16	3.51	0.18
7	N	K	8.78	1.64	14.32	2.54	0.84
14	Si	K	2.14	0.41	4.26	0.81	0.69

Noticeable, unlike the modified β -D-cyclodextrin, in chitosan: β -D-CD complexes, a networked structure was developed. Thus, the results of studying the morphology of the obtained samples using TEM indicate the origin of microstructural changes and the formation of porous structures, which confirms the successful formation of nanocapsules. These results are in good agreement with the findings reported by other authors[42-46].

Furacilin loading and encapsulation efficiency

The degree of furacilin loading into the polycomplex was calculated based on the amount of free furacilin in the supernatant obtained after the loading process. The concentration of furacilin in the supernatant was determined by UV spectrophotometry at a wavelength of 375 nm. Here, $\lambda=375$ nm corresponds to the maximum absorption of furacilin and was selected for quantitative determination of the drug [47-48]. For this purpose, furacilin solutions with concentrations ranging from 20 to 200 μ g/mL were prepared, their UV spectra were recorded, and a calibration curve was constructed to determine the furacilin loading efficiency in acetate buffer (pH 4.75) (Fig. 11).

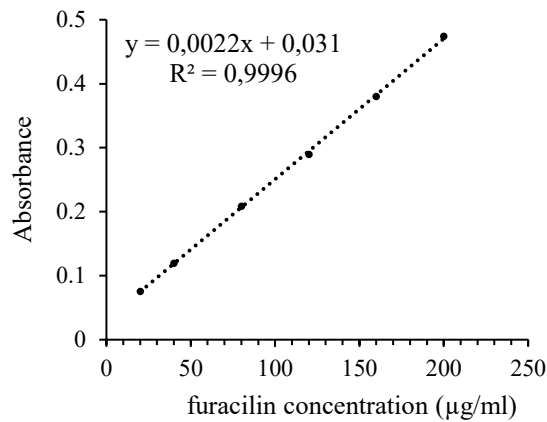


Fig. 11 Dependence of the optical density of furacilin solution on its concentration in acetate buffer (pH 4.75)

The calibration curve shows that with increasing furacilin concentration, the absorption intensity at $\lambda_{\max}=375$ nm also increases. According to the calculation results, the amount of free furacilin in 20 mL of supernatant was 1.9204 mg. Since the initial amount of furacilin was 4.00 mg, the amount of furacilin loaded into the polycomplex

was 2.0796 mg, and based on Eqs. (4) and (5), the encapsulation efficiency (EE) was 51.99%. The loading capacity (LC) of furacilin in the polycomplex was 5.20%.

It should be noted that drug-loading efficiency depends on the chemical structure of the loaded drug, loading conditions, and other factors. Therefore, while the loading efficiencies reported in [49-51] were higher than that obtained in the present study, the results reported in the works of these authors [25, 27, 52-53] were lower than the values obtained in our study.

According to the literature, the loading of furacilin into the CS:β-CD/AA polycomplex can occur as a result of several types of intermolecular interactions. The relatively hydrophobic fragments of the furacilin molecule can form a *host-guest inclusion complex* with the hydrophobic inner cavity of β-CD [54]. This inclusion complex is one of the main mechanisms for the encapsulation of drug molecules by β-CD and increases the solubility and stability of the drug substance [55-57]. In addition, the nitro (-NO₂), carbonyl (C=O), and amine (-NH₂) functional groups in the furacilin molecule [58] can form hydrogen bonds with the polymer matrix [59].

Study of the prolonged release of furacilin from the CS:β-CD/AA polycomplex

To determine the concentration of furacilin released in PBS (pH 7.4), a calibration curve was constructed based on solutions in the range of 10, 20, 30, 40, 50, 60, 80, and 100 μg/mL (Fig. 12). The resulting calibration equation is: $y = 0.002x - 0.0005$, $R^2 = 0.9989$.

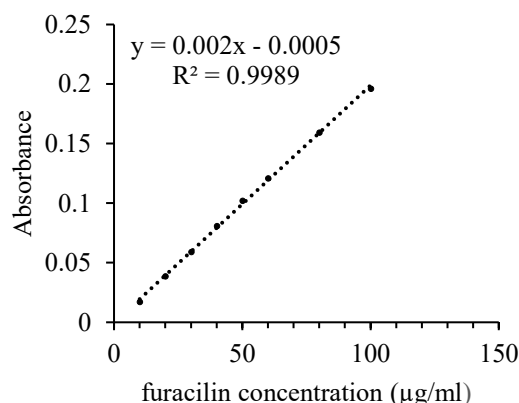


Fig. 12 Dependence of the optical density of furacilin solution on its concentration in phosphate buffer (pH 7.4)

As part of the study, the kinetics of furacilin release from the drug delivery system based on the synthesized CS:β-CD/AA=3:7 mass. polycomplex were studied (Table 13).

Table 13. Kinetics of furacilin release from the polycomplex

No.	Time, h	Absorbans (A)	C, μg/mL	Cumulative amount, mg	Release, %
1	2	0.0113	5.90	0.295	14.18
2	4	0.017	8.75	0.4375	21.03
3	6	0.0236	12.05	0.6396	30.75
4	24	0.0395	20.00	1.0612	51.02

In the UV spectra, an increase in the absorption intensity at 375 nm was observed over time, indicating an increase in furacilin release from the polycomplex (Fig. 13b).

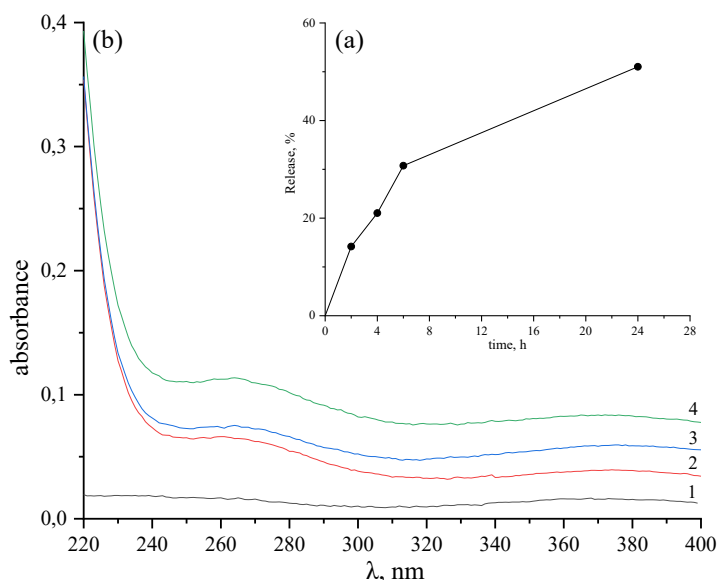


Fig. 13 Release kinetics (a) and Release of furacilin from the CS:β-SD/AA polycomplex: UV spectra at various time points (b): (1) 2h; (2) 4 h; (3) 6 h; (4) 24 h

The release rate of furacilin was 14.18, 21.03, 30.75, 37.80, and 51.02% at 2, 4, 6, 12, and 24 h, respectively. The cumulative release of furacilin increased over time. The release rate reaching 51.02% at 24 h indicated the prolonged release of furacilin from the polycomplex.

Thus, based on the experiments, the structure of the polycomplex-based nanocapsule allows for its controlled release (prolonged release).

Conclusion

To conclude, the synthesis of acrylic acid-modified β-D-cyclodextrin derivatives at various molar ratios, with the optimal stability and particle size observed at a 1:2 molar ratio. Dynamic light scattering (DLS) analysis of this sample indicated that 63.2% of particles had a size of 93.9 nm, confirming the formation of stable, spherical nanoparticles. Subsequently, polycomplexes were synthesized using chitosan and β-D-cyclodextrin modified with acrylic acid in mass ratios of 7:3 and 3:7, based on the 1:2 molar ratio derivatives. It was established that at a CS:β-CD/AA mass ratio of 6:4, the zeta potential reached a minimum of +1.7 mV, indicating maximum binding between chitosan and the cyclodextrin modification.

Hydrodynamic analysis revealed particle sizes of 172.6 nm (76.4%) and 106.1 nm (67.5%) for these polycomplexes. SEM imaging confirmed the formation of hollow nanocapsules, while TEM analysis showed sizes ranging from 70.9 to 237 nm, with spherical aggregates observed.

The encapsulation efficiency (EE) of furacilin in a CS:β-CD/AA-based polycomplex and its loading capacity (LC) were studied, yielding 51.99% and 5.2%, respectively. The drug loading process into the polycomplex was investigated at pH 4.75 in an acetate buffer medium. Extended release was studied at pH 7.4 in a phosphate buffer medium, revealing that 51.02% of the furacilin was released within 24 hours.

Thus, these experiments demonstrate that CS:β-CD/AA-based polycomplexes are of fundamental and applied interest for targeted drug delivery.

Author Contributions: Zaynura Jakhonkulova: Investigation, validation, formal analysis, writing-original draft preparation. Noira Vokhidova: Conceptualization, data curation, formal analysis, methodology, validation, writing-review and editing. Sayyora Rashidova: Supervision, validation

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Conflicts of Interest: The authors declare no conflict of interest

Abbreviations

The following abbreviations are used in this manuscript:

CS	Chitosan
β -CD	β -D-cyclodextrin
AA	Acrylic acid
PEG	Polyethyleneglycol
EE	Encapsulation efficiency
LC	Loading capacity
PBS	Phosphate-buffered saline

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