Electronic supplementary materials *Mendeleev Commun.*, 2024, **34**, 372–375

Structure and properties of hydrogels based on sodium alginate and synthetic polyacids

Marina Yu. Gorshkova, Irina F. Volkova, Etery S. Grigoriyan and Sergey P. Molchanov

Sodium alginate (SA) (Foodchem. International Corp., China) with a molecular weight of Mw = 1.7 × 10^3 kg/mol was used as received. Hereinafter, molecular weights were determined by GPC. The MVEMA (Polyscience Inc., United States) with a molecular weight of Mw = 41 kg/mol was used as received. The copolymer DIVEMA with molecular weight of Mw = 50 kg/mol was synthesized by radical cyclocopolymerization.[S1] LD (Spectrum, United States), sodium chloride, sodium hydroxide, and hydrochloric acid (reagent grade, LabTekh, Russia) were used as received. The 0.01M phosphate-buffered saline with pH 7.4 (PBS), buffers 0,01M HEPES, and 0.01M MES were obtained by diluting concentrates (Sigma Aldrich, United States), the tetraborate buffer with pH 10.2 was prepared from boric acid and sodium hydroxide. Bidistilled water additionally purified on a Milli Q system (Millipore, United States) was used for preparation of all solutions.

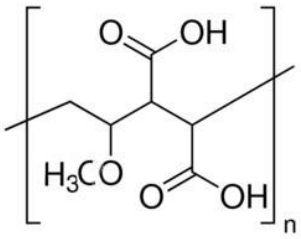

Figure S1 Structure of hydrolyzed MVEMA.

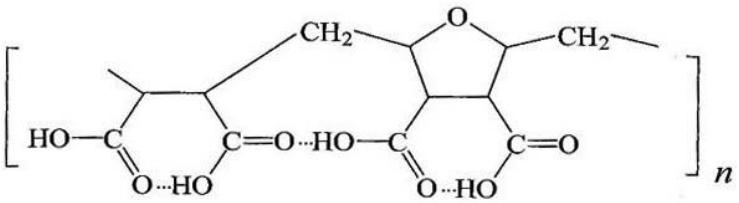

Figure S2 Structure of hydrolyzed copolymer DIVEMA.

The molecular weight of the polymers was determined by GPC on a Gilson instrument (United States, France) equipped with a Gilson RI-131 refractometric detector, a Rheodyne 7125 injector, a TSK gel GM PWXL column (Tosoh Bioscience, Japan), and the MultiChrom data acquisition and processing software (Ampersand, Russia). The tetraborate buffer with pH 10.2 containing 0.2 mol/L NaCl was used as a solvent and an eluent. The system was calibrated using PEO standards; analysis was done at room temperature.

The hydrogels were obtained by mixing aqueous solutions of the polymers. The calculated amount of copolymer aqueous solution (concentration 4 wt %) was added to an SA aqueous solution (concentration of 2 wt %) and resulting mixture was stirred for 10–15 min at room temperature and kept for 1 h. The mixture was poured onto a polypropylene substrate and dried at room temperature to a constant weight. Film samples subjected to heat treatment were exposed to temperature of 80°C for 24 h.

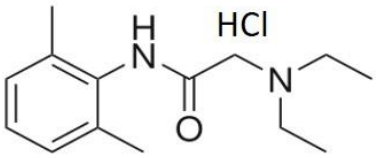

Figure S3 Structure of LD.

To prepare hydrogels containing 5 wt % LD in terms of the total weight of polymer components, the calculated amount of a LD aqueous solution with a concentration of 3.2 wt % was added to the SA solution under continuous stirring for 15 min and then the necessary volume of polyacid solution was poured in portions.

To estimate the swelling ratio (SR) of the hydrogels in water and buffers, the weighed portion of the sample (200 mg) was placed in a glass vessel and then 15 mL of liquid was poured. At certain time intervals the liquid was removed, the gel was carefully dried with a filter paper, and the wet hydrogel weight was determined. The SR was calculated by the formula

$SR = \frac{m_t - m_0}{m_0}$, where m_o and m_t is the weight of dry gel and wet gel, respectively.

Mass loss of the gels upon swelling for 24 h at room temperature due to the partial dissolution of hydrogel components in media was calculated from an initial weight of the sample taken in swelling and a weight of dried residue after measuring the swelling ratio by the formula

$\text{Loss} = 100 - \frac{m_{res} \times 100}{m_o}$, (wt%), where m_o is an initial weight of gel sample and m_{res} is a weight of dried residue of gel.

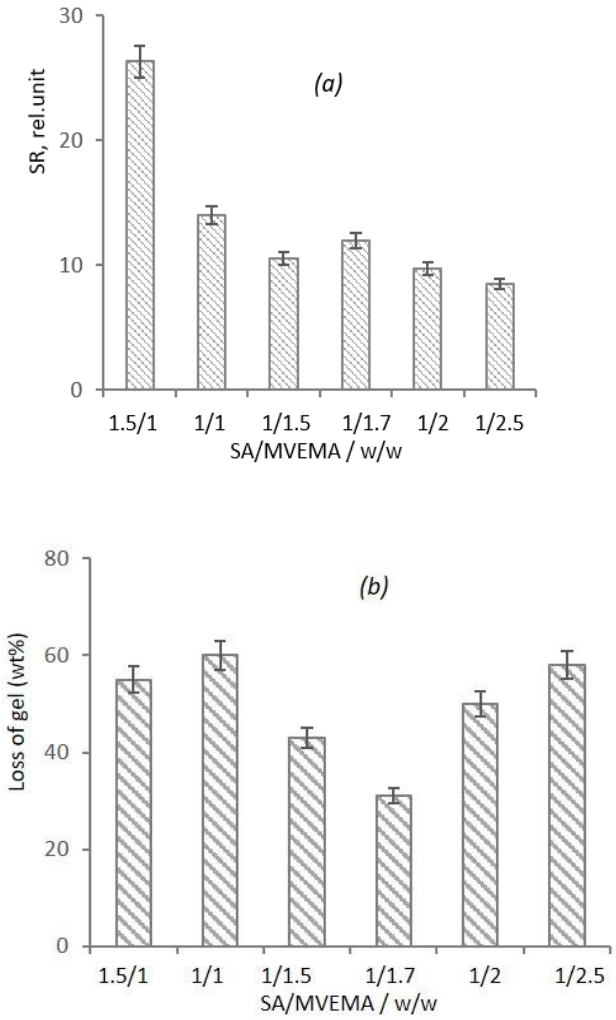

Figure S4 SR values: *(a)* (in 60 min of swelling) and part of soluble fractions (loss of gel in 24 h) of SA/MVEMA/80 gels, *(b)* with different weight ratio of components upon swelling in PBS at 23°C.

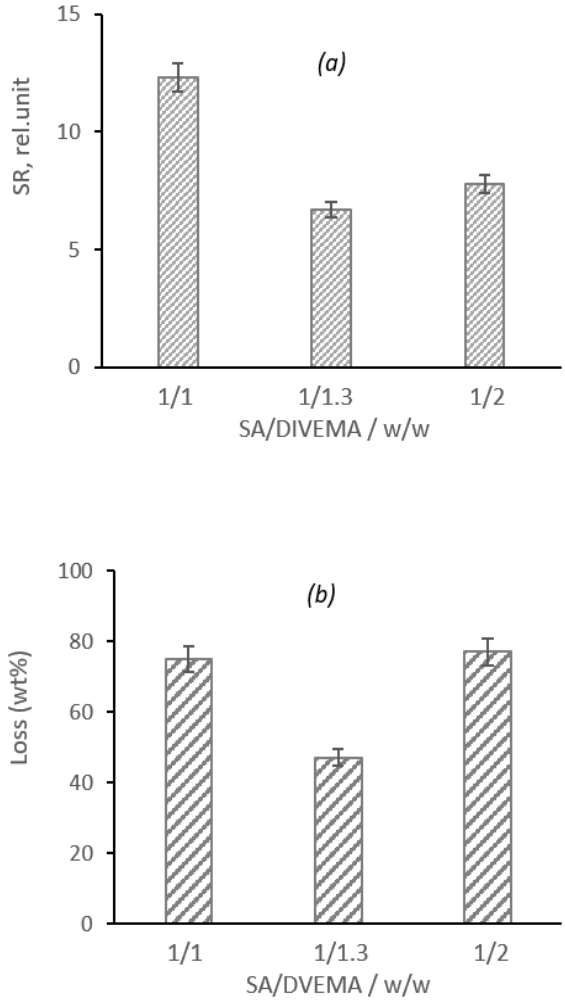

Figure S5 SR values: *(a)* (in 60 min) and part of soluble fractions (loss of gel in 24 h) of SA/DIVEMA/80, *(b)* with different weight ratio of components upon swelling in PBS at 23°C.

To study the kinetics of swelling and the rate of LD release in PBS solution a weighed potion of gel (200 mg) was placed in a glass vessel containing 15 mL of liquid and kept without stirring at room temperature. The choice of the ratio gel/ buffer in kinetic experiments on swelling of hydrogels and the LD release was made to be able to determine the concentration of the released LD by spectrophotometry. To estimate the LD rate release from the gel the weighed sample containing 5 wt% LD was placed in a given volume of the medium, the liquid phase was taken at certain time intervals, its optical density was measured, and it was brought back. The concentration of LD was determined by UV spectroscopy at wavelength of 271.4 nm on a Specord M 40 spectrophotometer (Carl Zeiss Jena, Germany) with the Soft Spectra data acquisition and processing software using preliminarily obtained LD calibration plots. The drug release (LD%) were calculated using the formula

$LD\% = {LD_t}/{LD_{ini}} \times 100, (\%)$, where LD_t is the current amount of LD in solution (mg) and LD_{ini} is the initial amount of LD (mg) in an experimental sample.

Thermal analysis. Thermogravimetric analysis was carried out on a TGA/DSC 3+ Mettler Toledo simultaneous thermal analyzer (Switzerland) using 150 μL Al2O3 ceramic crucibles, in the range of 25–700°C at a rate of 10°C/min in an atmosphere of nitrogen at a flow rate of 50 mL/min.

SEM images were taken on Thermo Phenom XL G2 (Thermo Fisher Scientific, Netherlands) for freeze-drying gels.

AFM studies. Atomic force microscopy (AFM) was used to study surface morphology of the gel film samples on micron and submicron scales (5–70 μm). A scanning probe microscope Horiba Smart SPM (France) with the semi-contact mode was used. Silicon cantilevers NSG30 Golden (TipsNano, Switzerland) were applied. The range of oscillations of the cantilever far from the surface of the compound sample was 20–25 nm. The radius of curvature of the probe tip was 5–10 nm. The determination and comparison of the surface roughness and porosity of the test samples were carried out using the Gwyddion software (version 2.62, Czech Metrology Institute, Brno, Czech Republic) from their AFM images.

The roughness was determined based on the automatically determined depth of dots on the image by the average roughness R_a

$$R_a = \frac{1}{l_r}\int_0^{l_r} |z(x)|\,dx$$

where l_r is the length of the baseline and z(x) is the deviation from the baseline.

Determination surface porosity (by inverting the surface morphology) was performed using Grain Analysis section. The detection, separation and sizing of depressions (hereinafter referred to as pores) were carried out using the “watershed algorithm”. All AFM-images were processed at the same procedure settings to obtain comparable statistical parameters. The operation "Summary information" was used for statistical processing of the results. Number of pores, average pore area, average pore size, and average pore volume were registered and compared. Since the settings of the "watershed algorithm" are not connected to the linear dimensions of the image, but are set in pixels and relative values, the dimensions of the pores are not constant for all scales and depend on the size of the image-scanning field. The comparison of the statistical parameters of pores of hydrogel films was carried out exclusively within the same size of the scanning field. The

characteristics were compared within the same polymer between films of different processing: without additional heating, after additional heat treatment at 80°C, after additional heating at 80°C with drug LD. Sample fragments were cut out from the hydrogel films and fixed with the outer side up on the microscope sample holders with double-sided adhesive tape. The AFM probe was positioned in several places on the surface in the most homogeneous areas. AFM study was carried out on areas of 70 μm x 70 μm, 20 μm x 20 μm, 10 μm x 10 μm, 5 μm x 5 μm.

References

S1 I. F. Volkova, M. Yu. Gorshkova, P. E. Ivanov and L. L. Stotskaya, *Polym. Adv. Technol.*, 2002**, 13**, 1067. DOI: 10.1002/pat.234.