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New hybrid chitosan–silicone-containing glycerohydrogels†

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New biologically active hybrid chitosan–silicone-containing glycerohydrogels based on chitosan and silicon glycerolates were synthesized by a sol–gel method; the effect of silicon glycerolates and chitosan concentrations on the process of gel formation was evaluated.

Polysaccharide biomacromoleculs such as chitin and chitosan are involved in the biomimetic silica sol–gel synthesis (silica biomineralization), where diatoms are preferred model organisms in silica biomineralization studies.[1–4]

Chitin and chitosan (the N-deacetylated form of chitin) are natural nontoxic, biodegradable and biocompatible polymers possessing physiological activity (wound healing, antibacterial, haemostatic, anti-inflammatory).[5–9] Chitosan is a polyelectrolyte in dilute acid solution (below pH 6.5) due to the presence of protonated amino groups (Figure 1).[10]

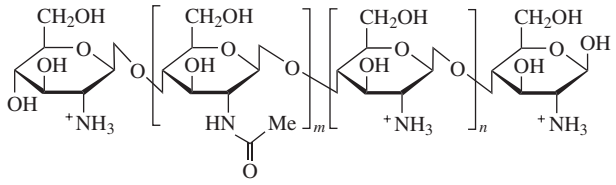

Figure 1 Molecular structure of chitosan in a charged form.

A combination of the biodegradability of a biopolymer with its bioactivity and the reinforcing properties of silica make it possible to prepare advanced biodegradable hybrid materials for biomedical applications such as wound dressing and as scaffolds in tissue engineering and for the sustained release of drugs.[6–8,11]

In the case of silica biomimetic studies, the silica precursor used in sol–gel processing is a crucial parameter. Silicon alkoxides $Si(OR)_4$,[1,2,4] colloidal silica SiO_2[8] and silicates such as Na_2SiO_3[3,10,12] have been used as precursors for biomimetic studies.

Tetrakis(2-hydroxyethyl) orthosilicate $Si(OCH_2CH_2OH)_4$ was involved as a biocompatible water-soluble precursor in the sol–gel synthesis of monolithic nanocomposite silica biomaterials, in particular, silica–chitosan hydrogels.[13,14] Such a sol–gel process was carried out in aqueous solution without a homogenizing solvent and a catalyst under ambient conditions. Unlike alcohols released during the hydrolysis and subsequent condensation of conventional precursors, such as tetramethyl and tetraethyl orthosilicate, ethylene glycol has no adverse effect on polysaccharides. Note that other silicon–polyol precursors, in particular, silicon glycerolates, were not used in the sol–gel synthesis of hybrid polysaccharide hydrogels.

Earlier, we used tetrafunctional silicon glycerolates[15,16] and combined di- and tetrafunctional silicon glycerolates[17,18] as biocompatible water-soluble precursors (Figure 2) to synthesize pharmacologically active organosilicon hydrogels.

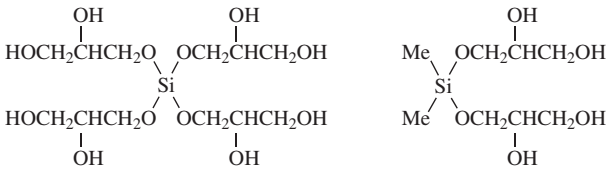

Figure 2 Silicon glycerolate precursors.

The gels obtained based on these precursors were nontoxic, and they exhibited pronounced wound-healing, regenerative and transcutaneous activity. It was interesting to apply the silicon glycerolates as precursors to the preparation of new hybrid chitosan–silicone-containing hydrogels with a wide spectrum of pharmacological activity.

The aim of this work was to synthesize new hybrid chitosan–silicone-containing glycerohydrogels by a sol–gel method and to investigate the influence of the concentrations of silicon glycerolates and chitosan on the process of gel formation.

For this purpose, we used tetrakis(2,3-dihydroxypropoxy)-silane $Si[OCH_2CH(OH)CH_2OH]_4$ **1** and combined dimethylbis-(2,3-dihydroxypropoxy)silane $Me_2Si[OCH_2CH(OH)CH_2OH]_2$ and tetrakis(2,3-dihydroxypropoxy)silane $Si[OCH_2CH(OH)CH_2OH]_4$ **2** synthesized at a molar ratio of 1 : 2 as precursors.‡ They were employed as glycerol solutions at a precursor : glycerol molar ratio of 1 : 3. The aqueous chitosan solutions contained 0.10, 0.25, 0.50 and 3.00 wt%.§

The influence of the precursor concentration on gel formation was studied using a 0.50 wt% solution of chitosan. Gels were formed at the concentrations of precursors **1** and **2** starting from 5 and 30 wt%, respectively. The hydrogels obtained were very weak and underwent syneresis on storage. Transparent (**1**) or semitransparent (**2**) monolithic hydrogels were prepared with increasing the concentration of precursors to 38 and 45 wt%, respectively.¶ These gels were stable at storage; when dispersed

† Dedicated to Academician Oleg N. Chupakhin on the occasion of his 80th Anniversary.

‡ Tetrakis(2,3-dihydroxypropoxy)silane **1** and combined dimethylbis-(2,3-dihydroxypropoxy)silan and tetrakis(2,3-dihydroxypropoxy)silane **2** were synthesized by the transesterification of tetraethoxysilane (cp grade, Russia) or dimethyldiethoxysilane (Sigma-Aldrich) and tetraethoxysilane in an excess of glycerol (analytical grade, Russia) at a reagent molar ratio of 1.0 : 7.0 or 0.5 : 1.0 : 8.0, respectively.[15,17] Starting ethoxysilanes were distilled at atmospheric pressure. Ethanol was removed at atmospheric pressure and then *in vacuo*. Ethoxy groups were completely replaced, as monitored by ^{1}H NMR spectroscopy (D_2O). The synthesized products were transparent viscous liquids readily soluble in water.

§ Aqueous solutions containing 0.10, 0.25, 0.50, and 3.00 wt% chitosan (Sonat; deacetylation degree, 0.82; molecular weight, 2.5×10^5) were prepared at pH 5.5–6.0 and kept for a day before use.

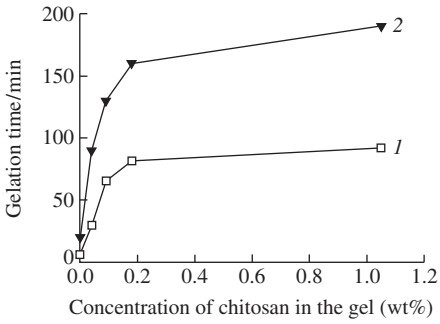

Figure 3 Dependence of gelation time on the concentration of chitosan in the gel for precursors (*1*) **1** and (*2*) **2**.

the gels were easily converted to an ointment-like state. Note that the silicon–glycerol precursors were completely compatible with chitosan, and phase separation or precipitation was not observed. A further increase in the precursor concentration resulted in the formation of very hard and strong hydrogels (precursor **1**) or phase separation (precursor **2**); at the same time, the gelation time was significantly increased.

Figure 3 shows the dependence of the gelation time on the concentration of chitosan in the gel in a range from 0.03 to 1.05 wt% (the initial concentration of chitosan in solution varied from 0.10 to 3.00 wt%); the concentrations of precursors **1** and **2** were 38 and 45 wt%, respectively.

Thus, a considerable deceleration of gelation occurred already when the concentration of chitosan in the gel was 0.03 wt%; however, with increasing concentration to more than 0.18 wt%, the gelation time did not change (precursor **1**) or increased insignificantly (precursor **2**). The gels obtained were stable in storage; they possessed the required consistency and did not injury the skin and mucous membranes when applied.

When silicon glycerolates interact with water or chitosan solution, Si–OH groups are formed due to reversible hydrolysis. The excess of glycerol in the system shifts the equilibrium back to the starting components. Then, silanol condensation occurs to generate Si–O–Si groups. The polycondensation processes result in the formation of a polymeric three-dimensional network, which is accompanied by the loss of fluidity and the formation of polymeric gels.[16,17] Since chitosan is a polyfunctional compound, its OH, $^{+}NH_3$ and NHCOMe groups can form numerous intermolecular bonds including hydrogen bonds with the C–OH and Si–OH groups of the precursors.[3,13] Based on published data,[10,13] we assume that chitosan is a template for the production of the polymer hydrogels. Note that chitosan retards gelation (Figure 3) unlike the formation of biocomposites based on positively charged chitosan macromolecules and negatively charged colloidal particles of SiO_2,[19] where polyelectrolyte complexes were formed. Polyelectrolyte complexes with chitosan have been also considered in the case of anionic polysaccharides.[20]

¶ To prepare the hydrogels, the required amounts of a chitosan solution (or water) and a precursor in an excess of glycerol were placed in a probe tube and carefully stirred to homogenization. Then, the probe tube was tightly sealed and kept in a thermostat at 80 °C. The gelation time was determined visually by the loss of fluidity in the system. Gels were characterized by elemental analysis, refractometry and IR spectroscopy.

Two hydrogels containing ~1.0 wt% chitosan and 38 wt% precursor **1** or 45 wt% precurcor **2** synthesized in an excess of glycerol passed preliminary animal testing in accordance with a published procedure.[12] The gels are nontoxic and exhibit pronounced wound-healing, regenerative activity and an anti-edema effect.

Thus, the novel biologically active hydrogels were obtained by a sol–gel method based on chitosan and silicon glycerolates; the effect of the concentrations of starting components on the gelation process was revealed. The gels can be recommended for medical applications.

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References

1 H. Ehrlich, D. Janussen, P. Simon, V. V. Bazhenov, N. P. Shapkin, C. Erler, M. Mertig, R. Born, S. Heinemann, T. Hanke, H. Worch and J. N. Vournakis, *J. Nanomater.*, 2008, doi:10.1155/2008/670235.
2 M. Sumper and E. Brunner, *ChemBioChem*, 2008, **9**, 1187.
3 K. Spinde, M. Kammer, K. Freyer, H. Ehrlich, J.V. Vournakis and E. Brunner, *Chem. Mater.*, 2011, **23**, 2973.
4 M. Wysokowski, T. Behm, R. Born, V.V. Bazhenov, H. Meißner, G. Richter, K. Szwarc-Rzepka A. Makarova, D. Vyalikh, P. Schupp, T. Jesionowski and H. Ehrlich, *Mater. Sci. Eng., C*, 2013, **33**, 3935.
5 R. Jayakumar, R. L. Reis and J. F. Mano, *E-Polymers*, 2006, **035**.
6 R. Jayakumar, N. Nwe, S. Tokura and H. Tamura, *Int. J. Biol. Macromol.*, 2007, **40**, 175.
7 Y. Maeda, R. Jayakumar, H. Nagahama, T. Furuike and H. Tamura, *Int. J. Biol. Macromol.*, 2008, **42**, 463.
8 K. Madhumathi, P. T. Sudheesh Kumar, K. C. Kavya, T. Furuike, H. Tamura, S. V. Nair and R. Jayakumar, *Int. J. Biol. Macromol.*, 2009, **45**, 289.
9 W. Janvikul, P. Uppanan, B. Thavornyutikarn, J. Krewraing and R. Prateepasen, *J. Appl. Polym. Sci.*, 2006, **102**, 445.
10 V. Pedroni, P. C. Schulz, M. E. Gschaider de Ferreira and M. A. Morini, *Colloid Polym. Sci.*, 2000, **278**, 964.
11 E. J. Lee, D. S. Shin, H. E. Kim, H. W. Kim, Y. H. Koh and J. H. Jang, *Biomaterials*, 2009, **30**, 743.
12 T. Coradin, R. Brayner, C. Gautier, M. Hemadi, P. J. Lopez and J. Livage, in *Biomineralization: from Paleontology to Materials Science*, eds. J. L. Arias and M. S. Fernandez, Editorial Universitaria S.A., 2007, pp. 419–430.
13 Y. A. Shchipunov and T. Yu. Karpenko, *Langmuir*, 2004, **20**, 3882.
14 Y. A. Shchipunov, T. Yu. Karpenko, A. V. Krekoten and I. V. Postnova, *J. Colloid Interface Sci*., 2005, **287**, 373.
15 T. G. Khonina, O. N. Chupakhin, L. P. Larionov, T. G. Boyakovskaya, A. L. Suvorov and E. V. Shadrina, *Pharm. Chem. J.*, 2008, **42**, 609 (*Khim.-Farm. Zh*., 2008, **11**, 5).
16 T. G. Khonina, A. P. Safronov, E. V. Shadrina, M. V. Ivanenko, A. I. Suvorova and O. N. Chupakhin, *J. Colloid Interface Sci*., 2012, **365**, 81.
17 E. Yu. Larchenko, T. G. Khonina, O. N. Chupakhin and L. P. Larionov, *Perspektivnye Materialy*, 2011, **2** (13), 978 (in Russian).
18 T. G. Khonina, E. V. Shadrina, A. A. Boyko, O. N. Chupakhin, L. P. Larionov, A. A. Volkov and V. D. Burda, *Russ. Chem. Bull., Int. Ed*., 2010, **59**, 75 (*Izv. Akad. Nauk, Ser. Khim*., 2010, 76).
19 Y. A. Shchipunov, N. A. Ivanova and V. Silant'ev, *Green Chem*., 2009, **11**, 1758.
20 Y. A. Shchipunov, N. A. Ivanova and S. A. Sarin, *Mendeleev Commun.*, 2009, **19**, 149.

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