ISSN-0011-1643 CCA-2409

Original Scientific Paper

Adsorption of Sulfated Polysaccharides on Mercury and γ -Al₂O₃ Particles in Sodium Chloride Solution and Seawater*

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Received February 5, 1996; accepted July 10, 1996

Adsorption of Dextran sulfate and natural sulfated polysaccharides from algal sources, ι -, κ - and λ -carrageenans on the mercury surface and on the γ -Al₂O₃ particles were studied by out of phase a.c. voltammetry. On the mercury surface, the adsorption of Dextran sulfate and adsorption of carrageenans are of similar extent. Comparative studies of the adsorption of polysaccharides on aluminium particles showed a considerable adsorption of i-carrageenan and Dextran-sulfate onto γ -Al₂O₃ particles ($\Gamma = 300 \text{ mg/g}$ and 100 mg/g, respectively). Adsorption studies were conducted at pH = 4 in 0.55 M NaCl and in natural seawater (pH = 7.9). Adsorption of i-carrageenans at the mercury/NaCl solution interface is stronger at the potential of -0.2 V than at -0.6 V vs. Ag/AgCl reference electrode. In seawater, the difference between adsorption of ι -carrageenan at two potentials is diminished for concentrations of polysaccharide lower than 10 mg/L, where the concentrations of calcium and magnesium ions present are high enough for neutralization of sulfate groups.

The results support the idea, that in seawater, adsorption on particles may play an important role in the transformation, removal and transport of polysaccharides to the sea bottom.

^{*} Dedicated to Marko Branica on the occasion of his 65th birthday.

INTRODUCTION

Natural polysaccharides derived from microbial and algal sources have an everyday application as stabilizers and emulsifiers in dairy products, toothpastes, cosmetics and a number of other products. 1,2 Recently, considerable attention has been paid to the investigation of polysaccharides since they are recognized as the main part of gelatinous aggregates observed during phytoplankton blooms, which can be regarded as one of the features of the phenomenon called marine snow. There are evidences^{6,7} that marine snow particles are formed by coagulation. Their relatively small fractal dimensions are consistent with their formation from coagulation processes. The chemical nature of particle surfaces is an important aspect of coagulation because it determines whether two particles will join to form a new larger particle after collision. The tendency of two particles to stick is expressed by using an experimentally determined parameter known as the collision efficiency factor or the stickiness. Formation of marine snow is widely studied, especially after it was recognized that with the small settling rate of single algal cells or their exudates it would not be possible to explain the rapid accumulation of organic matter on the seafloor after the spring algal bloom.4,5

Marine snow aggregates may be important for more than just their ability to move small algal cells rapidly to the seafloor, they could provide an isolated environment, different from the surrounding water. The role of coagulation in marine snow was studied by different models⁶ while the features of solid-liquid separations in natural aquatic systems were also discussed by O'Melia.⁷ Different studies were undertaken regarding the characterization and behaviour of polysaccharides in natural waters.^{8,9} It was found that polysaccharides produced by phytoplankton species of the Chaetoceros family were all sulfated and contained the sugars galactose, rhamnose and fucose in different proportions.¹⁰

Sulfated glycans were isolated from Rodophyta (red algae). Among these sulfated glycans are polyaccharides – carrageenans. They are based on linear chains of β -D-galactopyranose residues linked glycosidically through positions 1 and 3 (A units) and α -D-galactopyranose residues linked glycosidically through positions 1 and 4 (B units) (see Table I and Figure 1). These units are arranged in an alternating sequence $(AB)_n$. Carrageenans are classified according to the presence of 3,6-anhydro-D-galactose and the position and number of sulfated groups in different types of carrageenans $(\kappa$ -, λ -, μ -, ι -, ζ - and θ -), though a belief has been expressed that completely regular carrageenan structures do not exist in the nature. Regarding monomer composition of natural polysaccharides galactose was found to be the main part of the Adriatic sea mucilages, developed during phytoplankton blooms.

TABLE I				
»Idealized Repeating Units« (AB_n) and some data about the				
carrageenans investigated				

1. Identi- fying letter	2. A units (β-D-Galp)	3. B units (α-D-Galp)	4. Properties regarding formation of gels	5. Source of isolation
κ	4-sulfate	3,6-anhydro	rigid gels	Euchema cottonii
λ	2-sulfate (70%) unsulfated (30%)	2,6-disulfate	non-gelling form	Gigartina aciculaire and G. pistulata
ı	4-sulfate	3,6-anhydro 2-sulfate	flexible and compliant gels	Euchema spinosa

^{1,2,3 -} Ref. 8.

Complexation properties of λ -carrageenan towards Pb, Cd and Zn ions were investigated by voltammetry. The authors concluded that the reduction processes for these metal ions in the presence of λ -carrageenan are reversible and diffusion controlled and that the complexes have a labile behaviour on the time scale of the techniques used. It was supposed that the adsorption process of λ -carrageenan is present to a very limited extent, since this is the prerequisite for the method used for the calculation of the stability constants. They estimated the stability constants for the complexes to be in the range $\log K = 3.06-4.68$.

Here, we investigated the adsorption of selected sulfated polysaccharides, ι -, κ -, λ -carrageenans and Dextran sulfate on the mercury surface and on well defined aluminium particles in sodium chloride solution (0.55 M, pH = 4.0) and in seawater (pH = 7.9). Mercury surface was chosen as a model of hydrophobic surface. Biological debris, decomposed organic material and siloxane surfaces of silicates may be taken as models of natural hydrophobic surfaces. Aluminium oxide particles are taken as a model of hydrophilic surface. Oxide surfaces are quite common in natural waters as a result of the presence of different mineral particles. This investigation could give more insight into the processes which appear during the transformation of organic matter in natural waters. A high adsorption of carrageenans on hydrophobic and hydrophilic surfaces even at a few mg/L of added polysaccharides could explain the fast transformation of organic matter, sulfated polysaccharides in particular, from dissolved to colloidal-particulate state during phytoplankton blooms and in the situations when polysaccharide concentrations are increased. The concentration range of polysaccharides investigated is too

^{4,5 -} Sigma-Chemicals-Catalogue, 1994.

K - Carrageenan

$$CH_2OH$$
 O_3SO
 O_4
 O_4
 O_5OO_5
 O_5OO_5
 O_6
 O_7
 O_8
 O_8

Figure 1. Molecular structure of carrageenans (κ -, λ -, ι -) and Dextran sulfate.

low to suppose the formation of large aggregates in the solution. In the presence of particles or in the vicinity of mercury electrode the formation of aggregates could occur due to the increased concentrations in particular case.

EXPERIMENTAL

For the measurement of the amount of adsorbed polysaccharides the electrochemical method, phase selective a.c. voltammetry (PSACV) was applied. In phase selective a.c. voltammetry, a direct voltage is applied across the cell (i.e. between working and reference electrode) with superimposed sine-wave alternating voltage of small amplitude. 12,13 By using this method, either the capacitive current $-i_C$ (i.e. the current needed to build up a certain potential on the electrode) or the Faradaic current i_f (i.e. the amount of current needed for electron transfer at the interface) can be measured selectively since i_C is always shifted by 90° with respect to the applied alternating potential, i.e. current and potential are out of phase while i_f is in phase with respect to the alternating potential $\varphi = 0^{\circ}$. The capacitive behaviour of Hg/H₂O interface is mainly governed by the dielectric properties of the molecules in the adsorption layer. Organic molecules which adsorb on the electrode have a lower dielectric constant than water molecules, causing changes-decrease of the elec-

tric double layer capacitance. 13 Total capacitance of the electrode-solution interface is obtained by using the expression

$$\Theta = \frac{c_0 - c_{\theta}}{c_0 - c_{\text{org}}} \tag{1}$$

where Θ is the surface coverage $\Theta = \Gamma/\Gamma_{\max}(2)$ of the electrode with respect to organic adsorbates, and subscripts 0, Θ and org refer to the surface with no, partial or complete coverage of the electrode with organic molecules, respectively. Further, $i_{\rm C}$ is directly proportional to c, which gives a direct relation between the phenomenon of investigation, surface coverage (Θ) and measurable quantity $(i_{\rm c})$:

$$\Theta = \frac{i_{c,0} - i_{c,\theta}}{i_{c,0} - i_{c,\text{org}}}.$$
(3)

The experiments were performed with a stationary hanging mercury drop electrode (HMDE) in a three electrode cell (Metrohm VA 663) connected to a Metrohm E-506 polarograph. The phase selective a.c. voltammetry (frequency 75 Hz, amplitude 10 mV, phase angle 90°) was used for measurements of the amount of adsorbed polysaccharide on the mercury electrode, as well as for the determination of the amount of polysaccharide that remained in solution after the adsorption on \(\gamma \)-Al₂O₃. The mercury drop surface area was 5.4×10^{-3} cm⁻². Transport to the electrode surface was achieved by stirring the solution. Experiments were performed in 0.55 M NaCl solution and seawater as electrolyte. Sodium chloride salt was purified from organic contaminants by heating the salt at 723 K. After being prepared as a saturated solution (5.5 M), it was further cleaned with charcoal. The measurements were performed at two electrode potentials E = -0.6 V and -0.2 V vs. Ag/AgCl. The potential of -0.6 V vs. Ag/AgCl is close to the electrocapillary maximum of mercury in 0.55 M NaCl solution and favours the adsorption of neutral molecules. At the potential of -0.2 V, the mercury surface is positively charged and favours the adsorption of negatively charged molecules.

The chemicals used were Dextran sulfate (molecular weight 5×10^5 , Figure 1) was in the form of sodium salt with addition of phosphate buffer to give aqueous solutions of pH 6–8 and it was supplied from Pharmacia (Uppsala-Sweden). Carrageenans (1-, κ -, λ -) were from Sigma (USA) isolated from red algae (see Table I and Figure 1). Mean molecular weight value for carrageenans is reported in the literature as 3.5×10^5 to 7×10^5 . Al $_2O_3$ was Aluminium oxid C, Degussa AgG, Germany, which was also used in several adsorption studies carried out earlier. The oxide was characterized as nonporous, with an average particle size of 20 nm and a specific BET surface of 100 ± 15 m² g⁻¹. The concentrations of γ -Al $_2O_3$ in the solutions were 2 g/L. Adsorption experiments with Dextran, Dextran sulfate and ι -carrageenan on γ -Al $_2O_3$ particles were carried out as batch experiments: electrolyte solution (0.55 M NaCl), γ -Al $_2O_3$ suspensions to make 2 g/L and aliquots of polysaccharide solutions were prepared and equilibrated for 24 hours on a laboratory shaker, at room temperature. The suspensions were afterwards filtered on prewashed 0.45 μ m membrane filters, Millipore, for determination of the residual polysaccharide by phase selective a.c. voltammetry. As there is always some degree of uncertainty in-

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volved in filtration of polymeric material, the experimental samples were compared to blanks, which were treated in exactly the same manner, except that no γ -Al₂O₃ particles were added. Adsorption was calculated by (a) an internal comparison to experimental blanks and (b) by determination of the residual polysaccharide using the external calibration curve for the same polysaccharide.

RESULTS AND DISCUSSION

In Figure 2, a.c. voltammetric curves for adsorption of different concentrations of λ -, ι - and κ -carrageenans are presented. A decrease of capacitive current is noticed at the starting potential E=-0.2 V, in comparison to the value obtained for electrolyte without adsorbable organic molecules (curve 0). Characteristic peaks on the voltammetric curves are observable at approx

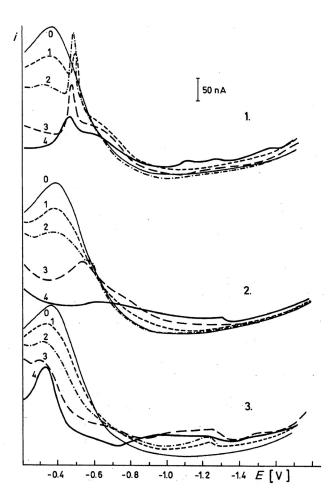


Figure 2. *i-E* voltammetric curves for ι -, λ -, κ -carrageenans for the concentrations of: 0) 0 mg/L; 1) 2.2 mg/L 2) 5.5 mg/L 3) 11 mg/L 4) 27.5 mg/L in 0.55 M NaCl at pH = 4.0; for 60 s of accumulation with stirring at -0.2 V vs. Ag/AgCl ref. electrode.

 $-0.4~\mathrm{V}$ and $-0.5~\mathrm{V}$ for ι - and λ -carrageenans, and approx. $-0.3~\mathrm{V}$ for κ -carrageenan and could probably be ascribed to the presence of sulfate groups in the polysaccharides and their interaction with the mercury surface, *i.e.* their reorientation and/or some faradaic process. For example, on the voltammetric curve of Dextran sulfate, a sharp peak similar to the one of ι -carrageenan was observed too. In Figure 3, we can see how the structure of the adsorbed layer of ι -carragenan changes with increasing concentration of polysaccharide. At a concentration range of 10 mg/L, we observe the characteristic peak at $-0.4~\mathrm{V}$ while at a concentration of 200 mg/L at which the adsorption pla-

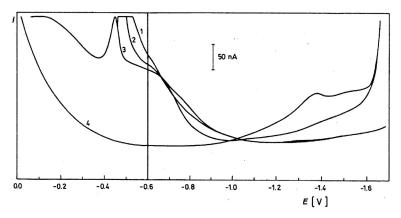


Figure 3. *i-E* voltammetric curves for *i*-carrageenan for the concentrations of: 1) 0 mg/L 2) 5 mg/L 3) 10 mg/L and 4) 200 mg/L in 0.55 M NaCl, at pH = 4.0 for 60 s of accumulation with stirring at -0.6 V vs. Ag/AgCl ref. electrode.

teau is already achieved, we can observe the high suppression of the capacity current, while the peak at -0.4 V has disappeared. The experiment in Figure 3 was performed so that the accumulation with stirring (60 s) was performed at the potential of -0.6 V and then the scan direction was turned to negative direction so that voltammetric curves were recorded from -0.6 V to -2.0 V. In the second run, after an accumulation period (60 s), scan direction was turned to positive direction and so the positive part of the voltammetric curves were recorded, *i.e.* from -0.6 V to 0 V. On that way the whole voltammetric curve was recorded from \sim 0 V to -2.0 V. For 200 mg/L of ι -carrageenan at the potential of -1.5 V, a small wave is observable which could be ascribed to the faradaic process. ¹⁸ On the positive side of the voltammetric curve, no adsorption wave is observable, it is probably it is masked by the oxidation wave of mercury. From the i-E voltammetric curves presented in Figure 2 and in Figure 3 for the different concentrations of polysaccharides the nonequilibrium adsorption isotherms were constructed

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as shown in Figure 4 and Figure 5. In Figure 4, nonequilibrium adsorption isotherms on mercury surface for Dextran sulfat, ι -, κ - and λ -carrageenans are presented for 60 s of accumulation time at -0.6 V and/or -0.2 V at pH 4.0 in 0.55 M NaCl. For ι -carrageenan, stronger adsorption was obtained at the potential of -0.2 V, at which the mercury surface is positively charged and favours the adsorption of negatively charged carrageenans. It is important to mention that pK of sulfate groups in Dextran sulfate is low, less than two, as reported by the supplier Pharmacia Fine Chemicals, and similar is to be expected for carrageenans. For λ -carrageenan and Dextran sulfate, we measured the adsorption only at the potential of -0.2 V, while at the potential of -0.6 V for lower concentrations of λ -carrageenan, a wave appeared

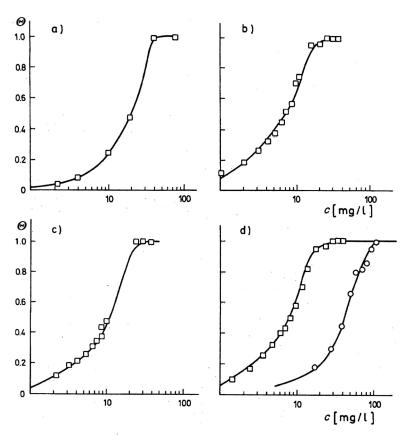


Figure 4. Adsorption isotherms for (a) Dextran sulfate, (b) λ -carrageenan (c) κ -carrageenan and (d) ι -carrageenan in 0.55 M NaCl, pH = 4.0, for 60 s of accumulation with stirring at the potentials of -0.2 V vs. Ag/AgCl ref. el (\square) and -0.6V vs. Ag/AgCl ref. el. (O).

which interfered with the measurement, as well as for Dextran sulfate in the a whole concentration range.

In Figure 5, the adsorption isotherms for carrageenans in natural seawater at pH = 7.9 are presented. The difference between the adsorption of ι -carrageenan at two potentials (-0.2 and -0.6 V) is smaller than in 0.55 M NaCl solution especially for lower concentrations of carrageenan. This could be explained by the fact that Ca²⁺ and Mg²⁺ ions partially neutralized the negative charge of sulfo groups on polysaccharide. At a higher concentration range of polysaccharide, i.e. > 10 mg/L for ι -carrageenan, the concentrations of Ca²⁺ (10⁻² mol/L) or Mg²⁺ (5 × 10⁻² mol/L) ions in seawater present are not enough to neutralize negative charges, so the difference between the adsorption curves at two potentials becomes apparent. The influence of cations on the adsorption of polyelectrolytes, such as for example polyacrylic acids, has already been observed. The formation of a complex in the solution prior to the adsorption on different surfaces was supposed to occur, which then enhances the adsorption to the surfaces. ¹⁹⁻²³

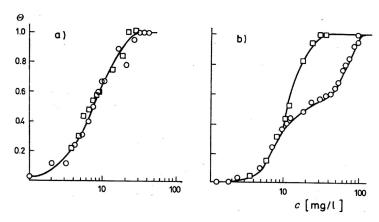


Figure 5. Adsorption isotherms for a) λ - (\square) and κ - (0) and b) ι - (\square ,0) carrageenans in seawater, pH = 7.9, for 60 s with stirring at the potentials of:

a) -0.2 V vs. Ag/AgCl ref. el.

b) -0.2V vs. Ag/AgCl ref. el. (\square) and -0.6 V vs. Ag/AgCl ref. el. (O)

In Figure 6, the equlibrium adsorption isotherm (at pH = 4.0) in 0.55 M NaCl solution for Dextran sulfate on γ -Al₂O₃ particles is presented as the dependance between the amount of substance in the solution vs. the adsorbed amount of Dextran sulfate per gram of added adsorbents. We can see that the surface density Γ of Dextran sulfate is about 100 mg/g γ -Al₂O₃. At the same time Dextran T-500 with the same molecular mass as Dextran sulfate, but without sulfate groups present was adsorbed to a much lower ex-

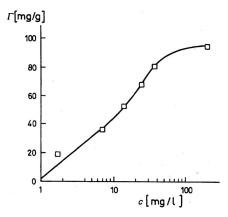


Figure 6. Adsorption isotherm of Dextran sulfate in 0.55 M NaCl (pH = 4.0) on γ -Al₂O₃ after 24 hours of equilibration time. Residual concentrations in solution were determined by a.c. voltammetry. Surface density Γ is plotted vs. the residual concentration of Dextran sulfate in solution.

tent (Γ = 30 mg/g γ -Al₂O₃), showing the importance of sulfate groups for the adsorption onto hydrophilic surface. In Figure 7, the equilibrium adsorption isotherm for ι carrageenan on γ -Al₂O₃ (pH = 4.0) in 0.55 M NaCl solution is presented. Here, the maximum of adsorbed ι -carrageenan is about 300 mg/g γ -Al₂O₃. This is about a three times higher concentration of adsorbed polysaccharide than in the case of Dextran sulfate. If we take into account the adsorption density of ι -carrageenan (300 mg/g γ -Al₂O₃) and Dex-

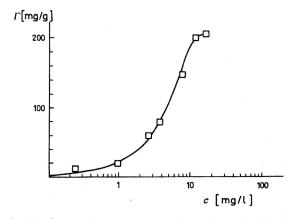


Figure 7. Adsorption isotherm of ι -carrageenan in 0.55 M NaCl (pH = 4.0) on γ -Al₂O₃ after 24 hours of equilibration time. Residual concentrations in solution were determined by a.c. voltammetry. Surface density Γ is plotted vs: the residual concentration of ι -carrageenan.

tran sulfate (100 mg/g $\gamma\text{-}Al_2O_3$), as well as the percentage of sulfur present (7 and 17%, respectively), we obtain the same adsorption density expressed in units of sulfur groups / m^2 (3.95 \times 10^{18} units / m^2 for $\iota\text{-carrageenan}$ and 3.19×10^{18} units / m^2 for Dextran sulfate). The adsorption is comparable to the adsorption of polyacrilic acids 23 at pH = 4.3 and the adsorption of humic acid to the same surface. 17 The adsorption of carrageenans (polysaccharides) to a considerable extent on hydrophilic particles could be an additional explanation for the fast removal of extracellular products of algal blooms in the water column, since it is known that the major part of exudates are polysaccharides. 3,9,10,24 In the particular case, the concentration range of sulfo polysaccharides is too low to allow coagulation in the solution and formation of gellatinous aggregates. 1,6 Adsorption studies of carrageenans in a higher concentration range (several grams per liter) are in progress.

CONCLUSIONS

The comparative study of the adsorption of polysaccharides on the mercury surface and γ -Al₂O₃ particles has shown that on the mercury surface the adsorption of Dextran sulfate and sulfated carrageenans are of similar extent. In 0.55 M NaCl solution, a stronger adsorption for ι -carrageenan was obtained at the potential of -0.2 V, where mercury surface is positively charged and favours the adsorption of negatively charged molecules. In seawater, the presence of calcium and magnesium ions partially neutralizes the negative charge of carrageenans by the formation of complexes in the solution and the difference between the adsorption at different potentials becomes less obvious.

A considerable adsorption of Dextran sulfate and ι -carrageenan on γ -Al $_2$ O $_3$. particles were obtained. That could be one of the possible ways of fast removal of polysaccharides appearing during phytoplankton bloom and their transport to the sea bottom.

Acknowledgments. – The authors acknowledge financial support of the Croatian Ministry of Science and Technology and the USA-Croatian Grant JF 145.

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SAŽETAK

Adsorpcija polisaharida sa sulfatnim skupinama na živi i česticama γ-Al₂O₃ u natrijevu kloridu i morskoj vodi

Marta Plavšić, Božena Ćosović i Jasminka Škudar

Adsorpcija dekstran-sulfata i prirodnih polisaharida sa sulfatnim skupinama ι -, λ - and κ -karagena, izoliranih iz alga, ispitivana je voltametrijom izmjenične struje. Na živinoj površini adsorpcija dekstran-sulfata i karagena približno je jednaka. Usporedna ispitivanja adsorpcije na česticama aluminijeva oksida pokazala su znatnu adsorpciju ι -karagena i dekstran-sulfata (Γ = 300 mg/g, odnosno 100 mg/g). Adsorpcija je mjerena kod pH = 4.0 u 0,55 M NaCl i u morskoj vodi pH = 7,9. Adsorpcija ι -karagena na granici faza živina površina/ elektrolit jača je na potencijalu -0.2 V nego na -0.6 V prema referentnoj Ag/AgCl-elektrodi. U morskoj vodi razlika u adsorpciji na ta dva potencijala smanjena je pri koncentracijama polisaharida manjima od 10 mg/L, gdje je koncentracija prisutnih Ca²+ i Mg²+ iona dovoljno velika da neutralizira sulfatne skupine karagena. Dobiveni rezultati u skladu su s idejom da je adsorpcija polisaharida na čestice u morskoj vodi važna za promjenu njihovih svojstava, uklanjanje i transport na morsko dno.