Acidity of Alginate Aerogels Studied by FTIR Spectroscopy of Probe Molecules

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Summary: Supercritical drying of alginate gels is an efficient way to prepare aerogels with high surface area (>300 $m^2 \cdot g^{-1}$). FTIR spectroscopy allows to monitor the adsorption of NH_3 from the gas phase onto the acid sites of the alginate. Free carboxylic groups are effective Brønsted sites, whereas the divalent cations used in the ionotropic gelation present the properties of Lewis sites. The ratio between Brønsted and Lewis sites provides infomation on the role of pH in alginate gelation and suggests that non-buffered gelation by transition-metal cations is a mixed ionotropic-acid process.

Keywords: aerogel; alginate; ammonia adsorption; FTIR spectroscopy; supercritical drying

Introduction

Alginates are abundant polysaccharides produced by brown algae, mainly composed of (1-4) linked β-D-mannuronic (M) and α-L-guluronic (G) residues (Figure 1), in varying proportions, sequence and molecular weight.

The use of alginate in most applications lies in its ability to form heat-stable strong gels with divalent or trivalent cations, most generally Ca^{2+}. The strength of the gel has been attributed to the electrostatical interaction of the cation with the guluronic residues, affording a three-dimensional network described by an "egg-box" model.[1] Alginates are used as thickeners in the food industry as well as for the encapsulation of bioactive materials like drugs,[2,3] proteins,[4] living cells,[5] and enzymes.[6] Several levels of characterization have been provided by Infra-Red spectroscopy since the work of Aspinall who reported the typical IR frequencies of the functional groups of polysaccharides.[7] Semi-quantitative estimation of the M/G ratio of alginates had been proposed by Mackie.[8] The values obtained by infra-red spectroscopy were in very good agreement with the others established procedures. Sakugawa proposed a simplified method for the estimation of the composition of alginate by FTIR using the Ca and Mn salts.[9] Filippov[10] and Chandia[11] characterized by FT-IR several alginates enriched in guluronic acids or mannuronic acids by partial hydrolysis. Sartori correlated the peak wavenumber of the FTIR spectra quantitatively to the Na^+ and Ca^{2+} content.[12]

Although important information was obtained in these works, the characterization had been hampered by the sample preparation, either thick film typically 10–20 μm or KBr pellets. No information was provided on the interaction of the polysaccharide with probe molecules. In this work, such a technique, typically used for the characterization of inorganic catalysts,[13,14] has been applied to the study of the acidity of alginate aerogels. Supercritical CO_2 drying of hydrogels has been

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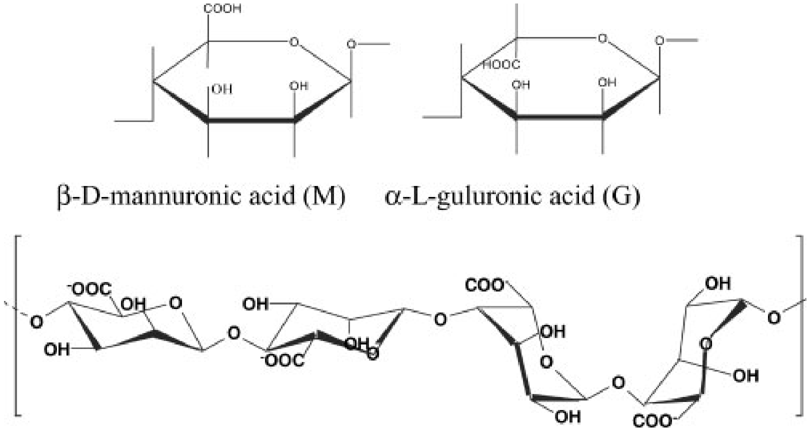

Figure 1.
Structure of alginate repeating units (M, mannuronic acid, and G, guluronic acid).

suggested as the best method to obtain an image of the wet materials in the solid state. This procedure releases the porous texture quite intact by avoiding the pore collapse phenomenon. The alginate aerogels beads obtained by this technique have a very high surface area close to 300 $m^2 \cdot g^{-1}$. The surface functions of the polysaccharide are thus accessible to probe molecules.[15,16]

Experimental

Preparation of the Alginate Gels

Alginate can form gel by ionotropic gelation where the cation coordinate to two carboxylic groups of the macromolecule or by lowering the pH. The properties of alginate gels are influenced by the ratio and sequencing of the uronic monomers,[19,20] the concentration of the cation in the maturation bath, and the time of maturation.[21,22]

Ionotropic gelation

The polymer solution (1% (w/w) solution of sodium alginate (Sigma medium viscosity, 35% guluronic content) in deionised water) was spread out at room temperature in a Petri dish and gently covered by a 0.24 M solution of $M(NO_3)_2$. Nitrates of Ca, Ba, Ni, Co, and Cu from Aldrich were used. The film was cured in the gelation solution during 22 hours. The resulting gels were shaped as film well-fitted to prepare self-supported disks for IR spectroscopy.

Acidic gelation

A 2% (w/w) solution of sodium alginate (10 cm^3) was cooled down to 2 °C. Ground maleic anhydride (0.2 g) was dispersed in the polymer solution and left to hydrolise until a hydrogel monolith was obtained.[17] The monolith was water-washed and dried. After supercritical drying, aerogel films were cut from the monolith.

Hydrogel drying

Two different procedures were followed in order to dry the films. Xerogel films were obtained after drying the hydrogel on glass plates in an oven at 50 °C. To obtain the aerogel films, intermediate alcogels were formed by immersion of the hydrogel films in a series of successive ethanol-water baths of increasing alcohol concentration (10, 30, 50, 70, 90, and 100%) during 15 min each.[18] The aerogel films were obtained by drying the alcogels under supercritic CO_2 conditions (74 bars, 31.5 °C) in a Polaron 3100 apparatus. Samples with different cations were prepared and labelled M-alg aerogel or M-alg xerogel according to the drying process and the nature of the cation.

Textural Properties

The samples were characterized by nitrogen adsorption-desorption at −196 °C.

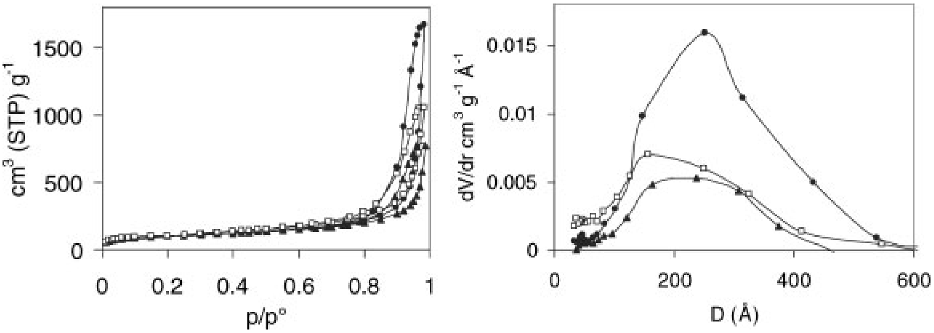

Figure 2.
Nitrogen adsorption-desorption isotherms at 77 K (left hand) and corresponding pore size distributions (right hand) of H-alg aerogel (empty squares), Ca-alg aerogel (filled triangles), and Cu-alg aerogel (filled circles).

Isotherms were recorded in a Micromeritics ASAP 2010 apparatus at $-196\,^{\circ}C$ after outgassing the sample at $80\,^{\circ}C$ under vacuum until a stable $3\cdot10^{-5}$ mbar pressure was obtained without pumping.

As expected when compared to the aerogel microspheres,[15] all isotherms of M-alg aerogels are of type IV at the borderline with type II in the IUPAC classification indicating the presence of large mesopores with a size distribution which continues into the macropore domain. The isotherms of three characteristic solids H-alg aerogel, Cu-alg aerogel, Ca-alg aerogel are given in Figure 2. The adsorption at low relative pressure allows evaluating the surface area of the sample by the BET method.[23] High surface areas comprised between 190 and 400 $m^2 \cdot g^{-1}$ are obtained, the values being reported in Table 1. The corresponding xerogels exhibit a surface area lower than 2 $m^2 \cdot g^{-1}$. The mesopore size distributions of the aerogels have been calculated according to the method of Broekhoff and de Boer[24] and are reported in Figure 2.

Infra-Red Spectroscopy

FTIR spectra were recorded on a Bruker Vector 22 spectrometer, equipped with a DTGS detector. Adsorption of ammonia was carried out using suitable IR cells equipped with KBr windows connected to a vacuum line (residual pressure $< 10^{-6}$ mbar).

Results and Discussion

Infrared spectra of both xerogel and aerogel films of Cu-alginate are reported in Figure 3 (spectra *a* for the aerogel and *b* for the xerogel). Both spectra present bands at the same wavenumbers: a broad band with a maximum around 3460 cm^{-1} assigned to O–H stretching, bands in the 2960–2840 cm^{-1} region assigned to C–H stretching, C–O stretching of the carboxyl group between 1750 and 1400 cm^{-1}, several bending modes between 1500 and 1200 cm^{-1}, and C–O and C–C stretching modes between 1200 and 1000 cm^{-1}.[25,26] The main bands related to the acidic functional group are the asymmetric C–O

Table 1.
Surface area and cation content of the aerogels.

sample	S_{BET} ($m^2 \cdot g^{-1}$)	% weight M^{2+}	% weight Na^+
Cu-alg aerogel	356 ± 5	12 ± 0.1	0.006
Co-alg aerogel	298 ± 5	10.5 ± 0.1	0.007
Ca-alg aerogel	374 ± 5	7.9 ± 0.1	0.01
H-alg aerogel	391 ± 5	—	<0.005

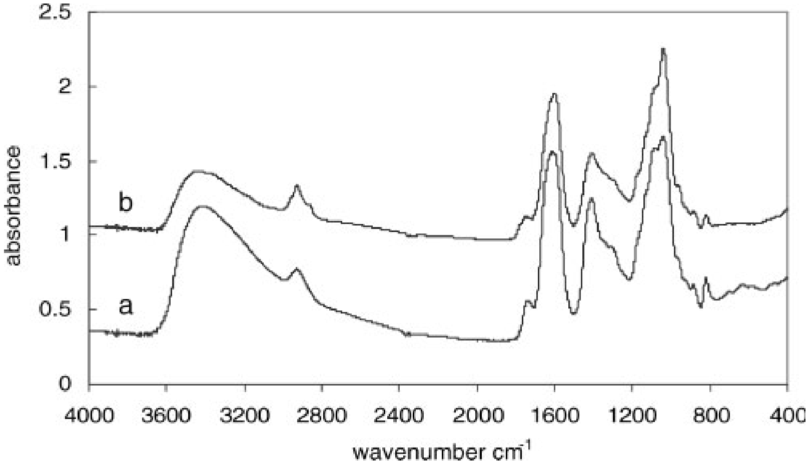

Figure 3.
FTIR spectra of Cu-alg xerogel (a) and Cu-alg aerogel (b). Spectra offset for clarity.

stretching of free (1745 cm^{-1}) or interacting carboxyl groups (1595 cm^{-1}). The ratio of absorbance of the bands suggests that most carboxyl groups are salified in the aerogel as well as in the xerogel. All the Na^+ cations have been exchanged, the amount of residual sodium is negligible (Table 1).

Ammonia was chosen as a molecule to probe the acidic functions, due to its small size, near to the size of the water molecule, and to its ability to differentiate between Lewis and Brønsted acid sites.[27] NH_3 is molecularly adsorbed on electron acceptor sites (Lewis acids), like as metal cations, and is transformed to NH_4^+ by proton exchange with a Brønsted acid site. The spectra of ammonia adsorption on Cu-alg xerogel and aerogel samples are reported in Figure 4.

When ammonia is contacted with the xerogel film, the appearance of the spectrum is not modified and no reaction is evidenced, whatever the pressure of NH_3 imposed (Figure 4 left hand). On the contrary, when ammonia is contacted with the aerogel film, the free carboxylic acid band disappears for a pressure of ammonia of only 1 mbar (Figure 4 right hand). More detailed information is provided by difference spectra reported in Figure 5, in which the absorbance adsorption is subtracted, wavelength by wavelength, from the absorbance after adsorption of ammonia.

In the case of the Cu-alg xerogel, no bands are eroded when ammonia is introduced in the cell, and the positive difference bands at 1610, 965, and 930 cm^{-1} are typical of unreacted molecular ammonia.

In the case of the Cu-alg aerogel, a negative difference band at 3545 cm^{-1} corresponds to the disappearing of the O–H stretching of the COOH groups, salified by ammonia. The strong bands at 3325 cm^{-1} and in the region 3260–3170 cm^{-1} correspond to the N–H stretching modes of molecularly adsorbed NH_3 on the Cu^{2+} cations.

The negative difference bands at 1745 and 1675 cm^{-1} correspond to the disappearing of the asymmetric C–O stretching of free COOH groups, and are accompanied by the increase of the 1600 cm^{-1} band of $COO^-NH_4^+$. The band at 1480 cm^{-1} is due to a deformation mode of NH_4^+ and the band at 1400 cm^{-1} to the symmetric C–O stretching of the carboxylate groups.

Despite the fact that the amount of cations in the gelling bath was is in a high excess, residual COOH were present, albeit in very small amount, when Cu^{2+} was used to gelify alginate. This incomplete salification of alginate was rather surprising: Haug and Smidrød did show the copper to present a very high affinity for alginate.[28] This fact prompted us to study the gelification of alginate with other cations. The

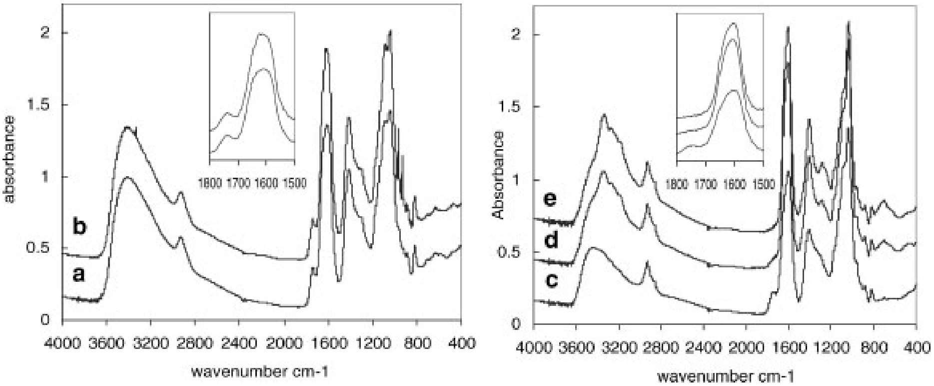

Figure 4.
Adsorption of NH_3 on Cu-alg xerogel (left hand); $p(NH_3)$ (mbar) (a): 0; (b): 94; on Cu-alg aerogel (right hand) $p(NH_3)$ (mbar) (c): 0; (d):1; (e): 6. Spectra offset for clarity. The region 1500–1800 cm^{-1} is magnified in the insets.

FTIR spectra of Cu, Co, Ca ionotropic alginate aerogels and of the acidic aerogel are reported in Figure 6.

The band at 1743 cm^{-1} corresponds to the C–O stretching of free COOH groups. In the case of ionotropic aerogels, residual COOH are observed, in very small amount, only when the transition metal cations Co^{2+} and Cu^{2+} are used. When calcium was used, all the acidic functions are salified. The different chemical affinity of the cations with the carboxylic anion, as measured at very low cation/alginate ratio,[28] does not explain the behaviour of the system at the high cation/alginate ratio of the gelling system. The driving factor for the level of cation exchange of the alginate seems to be the difference in pH value of the various cationic solutions. These values, reported in Table 2, can be classified into two groups, one where the pH is above the pKa value of alginate (4.2),[29] the other where the pH is below. The calcium cations belong to the former group, while the transition metal cations constitute the second.

In order to enhance this pH effect, an acidic gel was prepared at pH 1.94 (labelled H-alg aerogel). The ratio of absorbance of

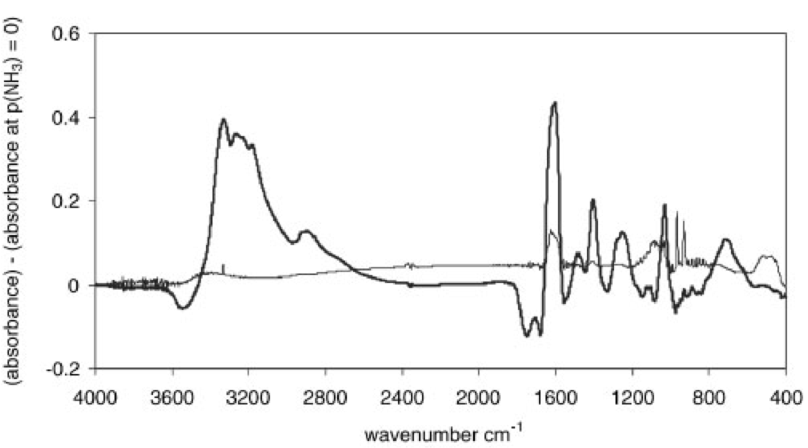

Figure 5.
Difference spectra for the adsorption of NH_3 on (thick line) Cu-alg aerogel at $p(NH_3) = 6$ mbar and (thin line) Cu-alg xerogel at $p(NH_3) = 33$ mbar.

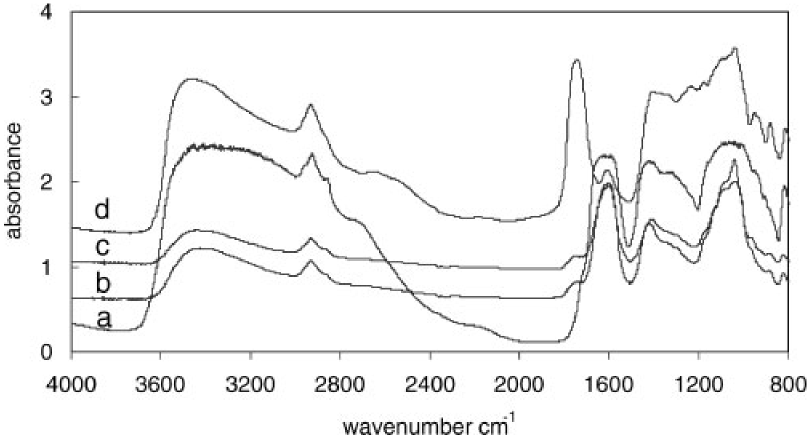

Figure 6.
FTIR spectra of M-alg aerogels: M = Ca (a); Co: (b); Cu: (c); H: (d). Spectra offset for clarity.

Table 2.
pH value of the gelling solutions.

sample	pH (20 °C)
Ca-alg aerogel	4.96
Co-alg aerogel	4.20
Cu-alg aerogel	3.51
H-alg aerogel	1.94

the bands at 1740 cm^{-1} and 1600 cm^{-1} suggests that most carboxyl groups are free in the H-alg aerogel (Figure 6d).

These data suggest that when the solution pH is below the pKa of alginate, an acidic gelation occurs along with the ionotropic gelation, leading to the presence of the free carboxylic acids.

When ammonia is contacted with these aerogel films, the formation of $COO^{-}NH_4^{+}$ on free carboxylic acid groups occur very easily with a pressure of ammonia of only 1 mbar (Figure 7) for the M-alg aerogel. Although these acid groups were not complexated by the metal cation, they are easily accessible to the probe molecule. The observations reported for Cu-alg aerogel are consistent with the behaviour of Co-alg aerogel.

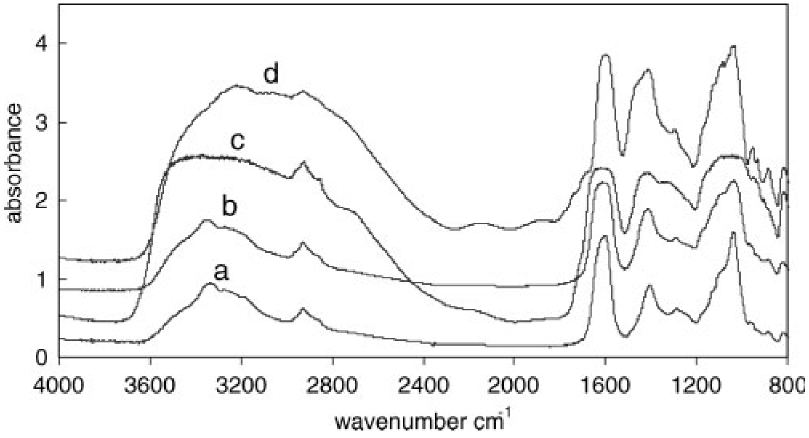

Figure 7.
FTIR spectra of NH_3 adsorption on M-alg aerogels. Co, 6 mbar NH_3 (a); Cu, 6 mbar NH_3 (b); Ca, 4 mbar NH_3 (c); H, 33 mbar NH_3 (d). Spectra offset for clarity.

Conclusions

FTIR spectroscopy of ammonia adsorbed on alginate aerogels reveals that the solids can be considered as acids. The carboxylic groups are effective Brønsted sites when the gelling agent is a transition metal cation or, especially, in the case of acidic gelling. The metal cations act as Lewis acid sites. All the Brønsted sites are accessible to the probe molecule in the supercritically-dried aerogels, while acid sites are inaccessible in the xerogel form of alginates.

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