# Aerogel materials from marine polysaccharides

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Hydrocolloid-forming polysaccharides are natural polyelectrolytes able to form stable hydrogels largely used in the food and pharmaceutical industry. Gelling polysaccharides derived from seaweeds or wastes of the seafood industry include polymers with several functional groups: alginates (carboxylic groups), carrageenans (sulfonic groups) and chitosan (amino groups). This article deals with suitable methods to prepare dry materials which retain the dispersion of the polymer hydrogel, namely polysaccharide aerogels. The materials whose properties are herewith described satisfy most of the appropriate requirements for heterogeneous catalysts and supports: they are stable in most organic solvents, present a high surface area and diverse accessible surface functionalities. Their application as catalysts, catalyst supports or adsorbents provide a new opportunity to obtain useful materials from one of the less energy-intensive sources of biomass.



## Introduction

The introduction of renewable resources in the production of catalyst supports and adsorbents is only possible if the materials intended to replace oil-derived or energy-intensive solids present adequate properties, such as high surface area, appropriate surface chemistry and porosity, thermal and chemical stability, and low cost. The purpose of this perspective is to show at which extent aerogel formulation of hydrocolloids from seaweeds and seafood wastes renders them suitable for applications as high-surface area materials.

Hydrocolloid-forming polysaccharides are natural polyelectrolytes able to gelify water when added in tiny amounts. Hydrogels containing 1–2% polymer and 98–99% water can be shaped as self-standing spheres or films with good mechanical stability. This property is at the basis both of their natural function as water-storage agents for living organisms as well as of their main application as thickening agents for the food industry. The dispersion of the hydrophilic polysaccharide chains in the hydrogels render them easily exchangeable, and the variety of their functional groups can be put to advantage. Natural polysaccharides available on industrial scale include polymers with anionic functions, such as alginates (carboxylic groups) or carrageenans (sulfonic groups) derived from seaweed, or with cationic functions, such as chitosan (amino groups), obtained by deacetylation of chitin from seafood shells or fungi (schematic representation of the monomers in Fig. 1). Their availability in nature is virtually unlimited and industrial processes for their extraction already run at the plant scale. The present annual production of seaweed-extracted polysaccharides is 45 000 t $y^{-1}$.[1] Seaweeds are probably the least energy-intensive sources of biomass whose production can be easily increased without competition with existing food resources.[2]

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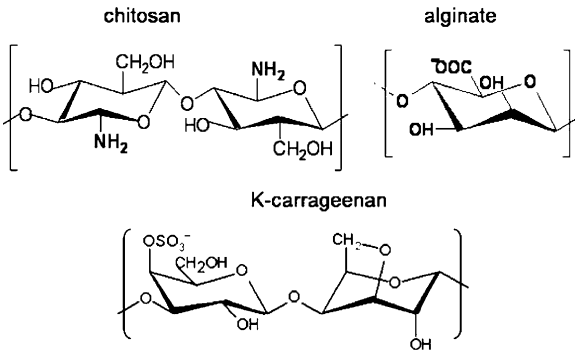


**Fig. 1** Repeating units of chitosan, alginate and κ-carrageenan.



The first application of polysaccharide hydrogels in catalysis, as support for enzymatic catalysts, is stemmed from their widespread use as growth media for bacterial growth. At present, alginates and carrageenans are the main enzyme supports in the brewing industry and for bio-labelled laundry powders.[1] More recently, natural polysaccharides have been tested as supports for metal catalysts.[3,4] They satisfy most of the appropriate requirements, as they are stable in most organic solvents and present numerous and diverse surface functionalities. The interactions of the functional groups with water reduce their effectiveness as catalysts in aqueous media. However, the use in catalysis of dried polysaccharide gels has suffered from diffusion limitations, due to the low surface area of the dried materials generally used, xerogels or cryogels.[5] Although the specific surface area is of major importance for a catalyst support, very few attempts to increase this parameter were reported in the literature. Clark and co-workers[6–8] described the use of expanded corn starch for liquid phase organic reactions. These authors formed an expanded starch gel network by gelatinisation in hot water, then by ageing the sample several weeks at low temperature and drying it through an organic solvent. Surface areas close to 200 $m^2\ g^{-1}$ were thus achieved.

In this perspective, we accept the common definition of xerogel as a hydrogel which has shrunk to a virtually non-porous state, while an aerogel is a gel which has been dried retaining the dispersion of the wet state. Evaporative drying methods bring about the collapse of the gel structure and do not allow preserving the accessibility of the functional groups. The technique of supercritical drying allows to avoid shrinkage of the gels and has already brought several breakthroughs in other fields of the materials science, *e.g.* for silica-based adsorbents, insulation materials, optical fibers or the preparation of carbon precursors.[9,10] In the case of polysaccharides, supercritical drying was originally used to prepare samples for electron microscopy.[11] Quite curiously, the investigation of the surface properties and of the applications of the polysaccharide aerogels as materials has only started in very recent years.

## Polysaccharide hydrocolloids

### Alginates

Alginates are a family of polysaccharides mainly produced by brown algae. They are constituted of (1 → 4) linked β-D-mannuronic (M) and α-L-guluronic (G) residues (Fig. 2), according to three types of sequences $(M)_m$, $(G)_n$ and $(M, G)_x$. Alginates differ by their M/G ratio. They are extensively used for the entrapment of biologically active materials.[12]

Most applications of alginates in drug release systems[13] and as supports in biocatalysis are based on their ability to form heat-stable strong gels with divalent or trivalent cations. Monovalent metal ions form soluble salts with alginate whereas divalent or multivalent cations, except $Mg^{2+}$, form gels. Cations show different affinity for alginate, and selective ion binding is the basis for the ability of alginate to form ionotropic hydrogels. The properties of the alginate gels are affected by the ratio and sequencing of uronic monomers,[14] the concentration of cations in the maturation bath and the time of maturation.[15,16] Alginates with high guluronic content give gels with a higher strength than alginate with high mannuronic content. This was attributed to the stronger affinity of the guluronic residues for divalent cations.[17–19] The structure of the ionotropic alginate gels has been described by the so-called "egg-box model", in which each divalent cation is cooordinated to the carboxyl and hydroxyl groups of four guluronate monomers from two adjacent chains of the polymer.[20–23] This structure, shared by other polysaccharide such as pectates, confers a high rigidity to the parallel aggregates of polymer chains.

A less frequently cited method to form an alginate gel is to lower the pH of a sodium alginate solution. The alginic acid gels formed in this way have been known and used since a long time,[13,24] but the understanding of these gels has been poor, as compared to the more extensively studied ionotropic gels. The effects of chemical composition and molecular weight on gel strength and gelling kinetics were studied.[17] Acidic gels, such

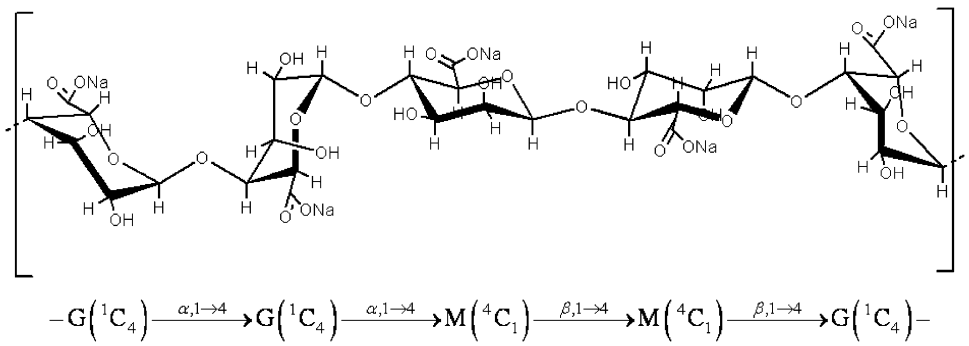


**Fig. 2** Bonding of the alginate repeating units: (M) mannuronate and (G) guluronate.

as ionotropic gels, present a higher mechanical strength for a high guluronic content. Small-angle X-ray scattering (SAXS) suggested the formation of junction zones with a high degree of multiplicity.[25,26]

### Carrageenans

Carrageenans are a group of natural polysaccharides extracted from red marine algae (Rhodophyceae). Their structure is constituted of a linear chain of alternating (1→3)-linked α-galactose-4-sulfate and (1→4)-linked 3,6-β-anhydrogalactose.[27] They come in three major types designated by means of the Greek letters κ, ι and λ. The main structural difference among them is in the sulfate group degree of substitution.

Among them, κ-carrageenan only bears one negative charge per disaccharide unit. Furthermore, it presents the best properties of the three types of carrageenan for gelation.[28] The details of the mechanism of the salt-induced gelation continue to be a matter of debate, although there is general agreement on a two-steps mechanism of gelation implying a coil-to-helix transition followed by a rapid aggregation of helices. Helix formation and gelation are cation-specific. Some monovalent cations, such as potassium, rubidium and caesium, strongly promote the gelation of κ-carrageenan.[29,30] Gel beads may be formed by dropping a solution of κ-carrageenans into a cation solution. Due to the easy implementation of this soft immobilization technique, κ-carrageenans beads are often used as a matrix to entrap molecules of biological significance,[31,32] such as food products, enzymes, whole microbial, plant or animal cells. The loaded beads are then used as immobilized biocatalysts or for controlled release of entrapped molecules of pharmaceutical interest.

### Chitosan

Chitosan, obtained by deacetylation of chitin (poly-β-(1,4)-acetylglucosamine) from seafood industry wastes (squid pens or crab shells), is a linear copolymer of linked β-(1,4)-glucosamine. Deacetylated chitin is defined as chitosan when its degree of deacetylation is beyond 60%.[33–35] The degree of deacetylation and molecular weight of chitosan influence all the physico-chemical properties, solubility, viscosity and can determine the field of application of chitosan.

The interest for chitosan stems from its applications in biomaterials, drug-delivery systems,[36,37] food additives, water clarification, and as support for cells and enzymes.[38] Chitosan can form either chemical or physical hydrogel.[39,40] Chemical hydrogels are formed by irreversible covalent links, as in crosslinked chitosan hydrogels. Physical hydrogels are formed by various reversible links, as in ionically crosslinked hydrogels and polyelectrolyte complexes, or in entangled gels. The latter are formed by solubilisation of chitosan in an acidic aqueous medium, and precipitation in an alkaline solution, which is the simplest way to prepare a chitosan hydrogel.

Several methods have been used to modify chitosan either physically,[41–43] or chemically,[44–46] in order to improve its mechanical or chemical stability, hydrophilicity and metal sorption capacity. Chitosan functionalization relies on the nucleophilicity of the amine group. It readily reacts with electrophilic reagents such as aldehydes,[47] acid chlorides,[48] acid anhydrides and epoxides.[49] The most straightforward route to functionalize primary amine containing materials is the reaction with an aldehyde to form an imine (Schiff base formation).[50,51] These materials were used for the reduction of chromate,[52] phenol,[53,54] and nitroaromatic compounds.[55,56] Functionalisation of chitosan has provided catalysts for cyclopropanation of olefins,[57] oxidation of alkylbenzene,[58] Suzuki and Heck reactions.[50] A recent review summarized the main advances in this field published in recent years.[3]

The amino groups of chitosan provide it with the highest chelating ability among the natural polymers obtained from seafood wastes and natural substances.[59,60] The high exchange capacity of the hydrogels has allowed to introduce large amounts of transition metals but shrinkage during evaporative drying has severely limited the accessibility of the metal particles.

## Experimental

The preparation of polysaccharide aerogels implies several steps. The first step is the formation of a hydrogel by reaction of a polymer solution with a gelling agent. The procedures used for (a) the preparation of hydrogels (for several polysaccharides), and (b) the preparation of aerogels (using $CO_2$ supercritical drying) are successively described.

### Preparation of alginate beads

Sodium alginate is dissolved in distilled water at a concentration of 2% (w/w). The polymer solution is added dropwise at room temperature to a stirred $CaCl_2$ or $CuCl_2$ (Aldrich) solution (0.24 M) using a syringe with a 0.8 mm diameter needle. The microspheres are cured in the gelation solution for 15 h.

### Preparation of carrageenan beads

A 2.5% (w/w) κ-carrageenan stock solution is prepared by dispersing κ-carrageenan (*eucheuma cottonii* Sigma 90%) in ultrapure water at 80 °C for 30 min. The principle of bead formation is the thermo- and ionotropic gelation of κ-carrageenan hot droplets added into a cold saline (KCl) solution. The 2.5% stock solution of κ-carrageenan thermostated at 80 °C is added dropwise into a 0.6 M KCl solution at 5 °C under stirring using a syringe with a 0.8 mm diameter needle. The gel beads are aged for 12 h in this solution at 5 °C without stirring and finally washed with cold water.[61]

### Preparation of chitosan beads

Aqueous solutions of chitosan are obtained by dissolving 2.5 g of chitosan in 100 mL of a solution of 0.055 mol $L^{-1}$ acetic acid. Two types of chitosan have been used: one from α-chitin from crab shell (Aldrich, degree of acetylation of 8%, molecular weight 700 000 g $mol^{-1}$) and one from β-chitin from squid pen (Mahtani Chitosan PVT, degree of acetylation 5%, molecular weight 200 000 g $mol^{-1}$). Each solution is added dropwise into a NaOH solution (4 M) through a 0.8 mm syringe needle. The chitosan beads are stored in the alkaline solution for 2 h, and then filtered off and washed with water.

### Supercritical drying

The microspheres are dehydrated by immersion in a series of successive ethanol–water baths of increasing alcohol

 

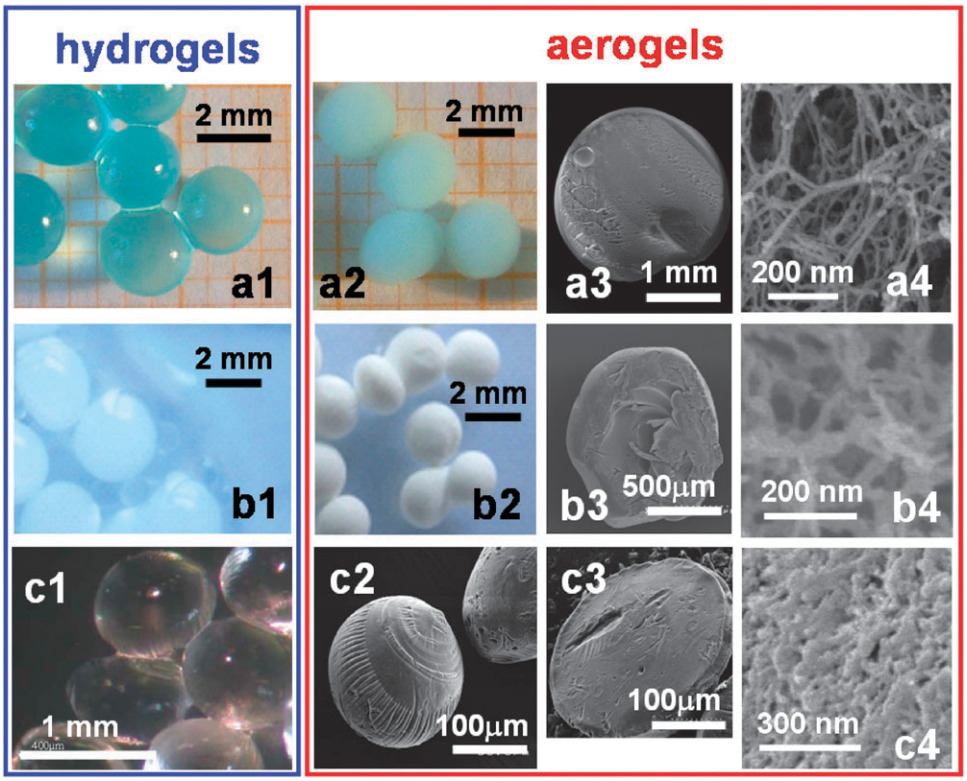


**Fig. 3** Optical and SEM images of hydrogel (first column) and aerogel spheres (second column) and of cross-sections of aerogel spheres (third and fourth columns) of Cu-alginate (row a), chitosan (row b) and carrageenan (row c).

concentration (10, 30, 50, 70, 90 and 100%) for 15 min each.[62] Finally, the microspheres are dried under supercritical $CO_2$ conditions (74 bar, 31.5 °C) in a Polaron 3100 apparatus.

### Characterization of the solids

Scanning electron micrographs of the aerogel beads were obtained using a Hitachi S-4500 apparatus. $N_2$ adsorption isotherms have been measured on a Micromeritics ASAP 2010 apparatus on samples outgassed at 50 °C. Surface areas were measured by the BET method by assuming a molecular area of $N_2$ of 0.162 $nm^2$. Mesopore size distributions have been evaluated from the desorption curves by the correlation of Broekhoff and de Boer for cylindrical pores.[63] The availability of reference silicas with mesopores of known size and DFT calculations have shown that this method provides more reliable results than the traditional BJH method.[64,65] In the size range of the pores of polysaccharide aerogels, the BJH method underevaluates pore size by about 30%.

The total pore volume of the aerogels has been evaluated by their mass/volume ratios. The macropore volume has been calculated as the difference between total pore volume and mesopore volume.

## From hydrocolloids to porous materials

The method of drying of the hydrocolloid controls the properties of the dried material. In the case of evaporative drying, most secondary structures of the hydrogels are compliant enough to be drawn together by capillary tension. In this case, the xerogel formed is virtually non-porous and has the same density of the dried polysaccharide.[66,67]

Among the alternative drying methods, freeze drying is largely applied in the pharmaceutical and food industry to reduce water activity and the susceptibility of the materials to bacterial attack. Freeze-drying usually allows to retain the volume of the hydrogel, as, for instance, in the case of alginate microbeads formed in a water/liquid $CO_2$ emulsion.[68] However, the secondary structures of the gels are extensively altered by the growth of ice crystals before the sublimation step.[69,70] Fast-freezing and pressure-freezing methods allow a better preservation of the fine structure of the gels. However, also in these cases significant structural damage can be observed for samples of thickness higher than a tenth of millimeter.[71]

Drying by supercritical $CO_2$ is an effective method to retain the structure of hydrogels. In the case of polysaccharides, the effectiveness of the drying method depends on the mechanical properties of the polymer. Optical and SEM pictures of beads of several polysaccharide hydrogels (Ca-alginate, chitosan from α-chitin, and κ-carrageenan) and the corresponding aerogels are reported in Fig. 3. It can be observed that the beads of alginate aerogel are slightly smaller than the beads of the parent hydrogel, while the formation of the aerogel has induced a significant shrinkage of the beads of chitosan and especially of the carrageenan hydrogel.

The micrographs of the cross-section of the aerogel beads reported in Fig. 3 provide some further information on the structure of the material. The distribution of the polymer is quite homogeneous through the beads of aerogel, except a

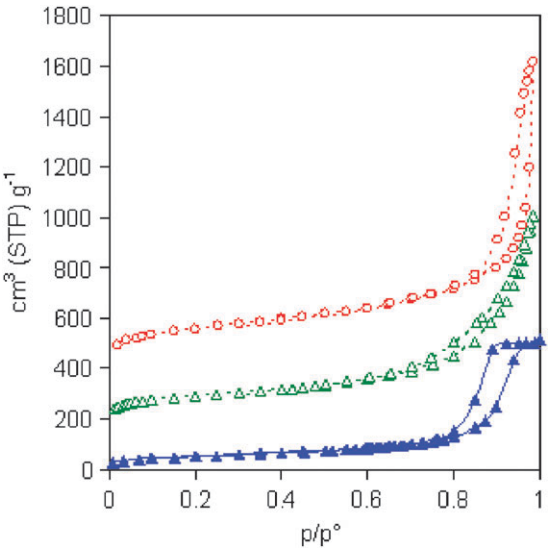


**Fig. 4** $N_2$ sorption isotherms at 77 K for Ca-alginate (○), chitosan (△) and carrageenan (▲) aerogels. The isotherms are shifted by 200 $cm^3\ g^{-1}$ for clarity.

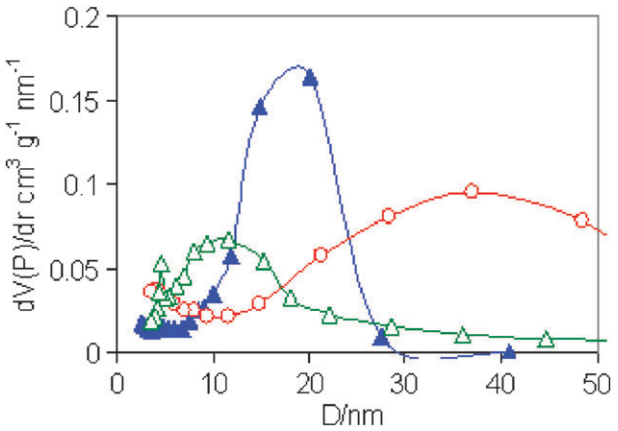


**Fig. 5** Pore size distributions for Ca-alginate (○), chitosan (△), and carrageenan (▲) aerogels.

slightly denser outer crust formed when the parent drop of polysaccharide solution has contacted the solution of the gelling agent.[72] At higher magnification, it can be observed that the alginate and chitosan aerogels present a network of isolated fibrils which corresponds to the structure of the polymer in the parent hydrogel. As a consequence of its severe shrinkage, the carrageenan aerogel presents a more compact structure, with cavities in the size range of mesopores (smaller than 50 nm).

The decrease in gel volume during the preparation of the aerogel is reported in Table 1 for the three different polysaccharide materials. The shrinkage of alginate hydrogels during the aerogel preparation induces a 7% decrease in bead size, corresponding to a 20% decrease in gel volume. The shrinkage takes place essentially in the alcohol exchange of the hydrogel and is negligible during the $CO_2$ supercritical drying of the alcogel.[66] Other polysaccharides present a more substantial shrinkage when submitted to the same procedure of supercritical drying. The likely explanation for this effect is the flexibility of the polymer structures in the gel. The entropic effect due to the presence of a solvent allows to keep the polymer strands well apart in the hydrogel. Upon drying, the enthalpic advantage of a reduction of the exposed surface is high enough to draw together the strands of the most flexible polymers, also in the absence of the tension of a solvent meniscus. The rigidity of the alginate fibrils is high enough to resist this effect, probably due to the peculiar egg-box structure which rigidifies the aggregates of parallel polymer chains.

If the pore size of the hydrogels is currently measured by diffusion methods,[73] the pore size of the aerogels can be measured by equilibrium methods. Intrusion mercury porosimetry is the most common method to evaluate the size of macropores (pores larger than 50 nm) but this technique requires the application of high pressure levels and the interpretation of the results can be questioned in the case of flexible or fragile solids, such as polymers and aerogels.[74–77] The size of mesopores (pores with size between 2 and 50 nm) is usually evaluated by adsorption volumetry. This technique provides useful pore size values if it is taken into account that most methods of pore size evaluation are calibrated on cylindrical pores and can provide biased results for pores of different geometry.[78] It has also been observed that compliant materials can somewhat bend upon the traction of liquid nitrogen menisci.[79,80]

The $N_2$ adsorption–desorption isotherms of the three polysaccharide aerogels are reported in Fig. 4. All the isotherms are type IV, with a significant hysteresis corresponding to the presence of mesoporosity. No significant microporosity is observed. The mesopore size distributions for the three materials are reported in Fig. 5 and the textural data calculated from the isotherms are reported in Table 1. Alginate and chitosan aerogels present broad mesopore distributions with no evidence of continuity with macroporosity (beyond 50 nm). Carrageenan aerogel presents a narrower mesopore distribution centered around 19 nm.

The mesopore volume from the $N_2$ sorption isotherms can be compared with the macropore volume calculated from the volume and the mass of the aerogel beads. The data, reported in Table 1, indicate that the mesopore volume represents a tiny fraction of the total volume of alginate and chitosan aerogels. The micrographs of Fig. 3 clearly indicate that the main

**Table 1** Textural properties of polysaccharide aerogels and volume shrinkage in the aerogel preparation

| Polysaccharide aerogel | Volume shrinkage (%) | Surface area/$m^2\ g^{-1}$ | Average pore size/nm | Mesopore volume/$cm^3\ g^{-1}$ | Macropore volume/$cm^3\ g^{-1}$ |
|---|---|---|---|---|---|
| Ca-alginate | 20 | 570 | 38 | 1.16 | 38 |
| Chitosan | 60 | 330 | 11 | 0.40 | 15 |
| κ-Carrageenan | 95 | 200 | 19 | 0.76 | 0 |

 

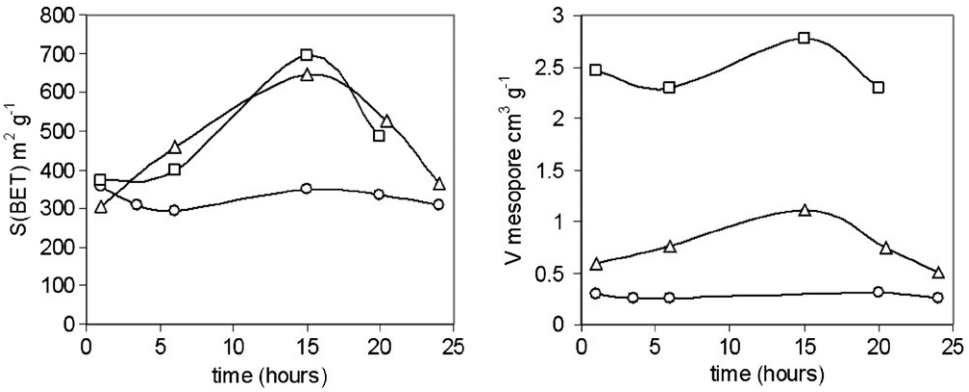


**Fig. 6** Influence of the guluronate/mannuronate ratio and ripening time on (left) the surface area and (right) the mesopore volume of Ca-alginate aerogels. Guluronate fraction: 74% (□), 45% (△), and 20% (○). The lines are guides for the eye.



porosity of these materials is in a size range larger than 50 nm, beyond the limits of detection by $N_2$ sorption. In these materials, the mesoporosity can to be attributed to widely spaced zones of contact between fibrils and the aerogels have to be considered as macroporous.

The aerogel of carrageenan presents a completely different texture. The mesopore volume corresponds to the whole porosity of the material (see Table 1), in good agreement with the micrograph of a cross-section of the aerogel (Fig. 3), in which only cavities smaller than 50 nm are visible. In this case, the pores correspond to residual cavities which are still accessible after a nearly complete collapse of the hydrogel structure.

It can be observed that the macroporous aerogels of alginate and chitosan present a high surface area (see Table 1). If the surface area is attributed to the outer surface of the isolated fibrils which constitute the materials, the average size of the fibrils can be evaluated at 40 Å for the Ca-alginate aerogel and 100 Å for the chitosan aerogel. If it is assumed that the polymer chains in the fibrils are essentially oriented in the direction of the fibril axis and present the same cross-section as the crystallized polymer,[81–84] it is possible to calculate an average number of chains per fibril: 32 for the Ca-alginate aerogel and 170 for the chitosan aerogel.

## Control of the structural properties

### Alginate

If we consider that the aerogel is the image on the dried state of the parent hydrogel, the parameters which modify the dispersion of the hydrogels also affect the textural properties of the aerogel. As described previously, the properties of the alginate gels are affected by the ratio and sequencing of uronate monomers (mannuronate or guluronate),[14] the concentration of cations in the gelling bath and the time of ripening in the gelling solution.[15,16]

The first parameter, the ratio and sequencing of uronic monomers is in fact controlled by the natural source of the polymer. The aerogel from guluronate-rich alginates presents a more compact nanostructure than the mannuronate-rich sample,[66] in agreement with the better mechanical properties of guluronate-rich hydrogels.[14] Velings and Mestdagh studied the kinetics of gelling of a mannuronate-rich Ca-alginate and found that the hydrogels reached stable mechanical properties after some hours ripening in the gelling solution.[15] The evolution of the surface area of Ca-alginate aerogels with the ripening time in a 0.24 M $Ca^{2+}$ solution is reported in Fig. 6 for three alginates with different guluronic ratios. It can be observed that after 15 h ripening the aerogels present a common stable value of surface area (about 350 $m^2\ g^{-1}$) independently of their guluronic content. This common value is reached in different ways for alginates with different guluronate content. The aerogels of mannuronate-rich alginate increased with ripening time and reached the final value after about 6 h, in good agreement with the gelling kinetics of Velings and Mestdagh. Quite curiously, the surface area of the aerogels of the guluronate-rich alginates reached a maximum of more than 500 $m^2\ g^{-1}$ at 3 h ripening time and decreased towards 350 $m^2\ g^{-1}$ for longer ripening time. This result suggests that the organisation of the guluronic-rich hydrogels passes through an intermediate state which is more dispersed that the final one.

The surface area is related to the size of the fibrils which form the aerogel. For a gelling time of a few hours, the surface area of the aerogel is virtually independent from the composition of the alginate. For longer gelling time, the surface area of the most mannuronate-rich sample (20% guluronate fraction) remains stable at 300–350 $m^2\ g^{-1}$. In the case of the samples with higher guluronate content, the surface area increases with the time of gelling up to a maximum of 600–700 $m^2\ g^{-1}$ at 15 h and decreases for longer gelling time. This behaviour is probably due to the interplay of the parameters affecting the size of the fibrils in the hydrogel and their coalescence in the drying process. Probably, increased gelling time allows the guluronate-rich samples to develop a better resistance to fibril coalescence, with a corresponding increase of the surface area of the aerogel. It is likely that coalescence of fibrils in the hydrogel itself accounts for the decrease of surface area for gelling time longer than 15 h.

The mesopore volume of the aerogel provides information on the regions in which the distance between aerogel fibrils is lower than about 50 nm. The mesopore volume depends on the composition of the alginate in a much more critical way than the surface area. Indeed, the mesopore volume of the aerogels is strictly related to the guluronate content (Fig. 6,

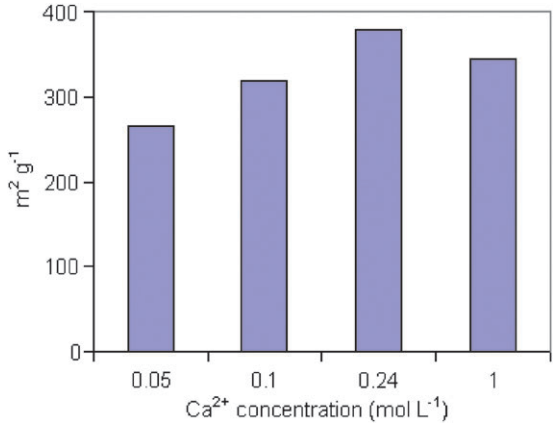


**Fig. 7** Influence of the concentration of the $Ca^{2+}$ gelling solution on the surface area of alginate aerogel.

right). For instance, for a gelling time of 6 h the mesopore volumes are 0.25, 0.76 and 2.15 $cm^3\ g^{-1}$ for alginates with guluronate fractions of, respectively, 20, 45 and 74%. This result has to be related to the higher rigidity of the guluronate-rich fibrils, which prevent their adhesion and allows them to shelter a significant volume in their contact regions.

The effect of the concentration of $Ca^{2+}$ cations in the gelling solution on the surface area of the aerogel is reported in Fig. 7 for a mannuronate-rich (20% guluronate) alginate after 6 h ripening. The surface area increases with the $Ca^{2+}$ concentration and reaches a shallow maximum at a concentration of 0.24 mol $L^{-1}$.

### Chitosan

The textural properties of the aerogels are largely controlled by the viscosity of the polymer solution,[85] which in turn depends on the molecular weight, the deacetylation degree and the natural source of the parent chitin. For a given degree of deacetylation, the type of chitin affects the distribution of the residual acetylated glucosamines, the crystallinity of the polymer and its gelling properties.[86] Generally, aerogels with higher surface area are obtained from hydrogels of chitosan obtained by deacetylation of α-chitin from crab shell than by deacetylation of β-chitin from squid pen.

The modification of the medium of gelation by replacing the alkali aqueous solution by an alkali water–alcohol solution strongly influences the textural properties of the corresponding aerogels. When the ethanol fraction of the gelling solution increases from 0 to 70% (v/v), the surface area of the aerogel increases from 125 to 220 $m^2\ g^{-1}$. According to the study of Clayer *et al*,[87] the amphiphilic structure of the alcohol contributes to maintain the critical balance between the hydrophobic and hydrophilic interactions. The increase of the specific surface area can be connected to the assumption that alcohol, by supporting the hydrophobic interactions to the detriment of the hydrogen bonds, prevents the collapse of the chains in the drying phase.

Important modifications of the textural properties are observed when gelling is induced by cross-linking agents, also at very low levels of reticulation.[88] Chemical gelling is obtained reacting chitosan with 1,1,3,3-tetramethoxypropane (TMP), a

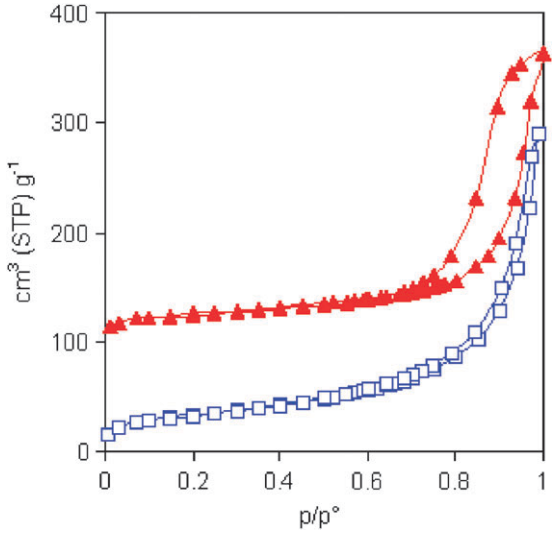


**Fig. 8** $N_2$ sorption isotherms of aerogels of chitosan (□) and cross-linked chitosan (▲). The isotherms are shifted by 100 $cm^3\ g^{-1}$.

"masked dialdehyde", followed by reduction of the polymeric Schiff base with an excess of cyanoborohydride.[89] In Fig. 8, the $N_2$ sorption isotherms of the aerogels of an alkali-gelled chitosan from β-chitin and the same chitosan gelled by TMP (2% molar cross-linker with respect to the number of amine groups) are compared. The aerogel of the alkali-gelled chitosan presents a surface area of 120 $m^2\ g^{-1}$ and a small mesopore volume with a broad pore size distribution which goes beyond the mesopore size limit of 50 nm. The aerogel of the cross-linked chitosan presents a surface area of 95 $m^2\ g^{-1}$ and a mesopore volume of 0.32 $cm^3\ g^{-1}$ with a narrower pore size distribution centered around 16 nm.

## Accessibility of the functional groups

Does the high surface area of the polysaccharide aerogels corresponds to a good accessibility of their functional groups? The interaction of the functional groups with selective reactive gases provides information on the accessibility of the active sites.

### Ammonia adsorption on alginic acid and alginate aerogels

FTIR spectroscopy of adsorbed $NH_3$ has been used to investigate the acidity of alginate aerogel films prepared in the presence of different cations or gelified in acidic medium.[90,91] Ammonia was chosen as the probe molecule due to its ability to differentiate between Lewis and Brønsted acid sites. The spectra of ammonia adsorption on the aerogel and xerogel, reported in Fig. 9 after equilibration with 30 mbar of $NH_3$, dramatically differ in the case of an alginic acid gel. In the adsorption of ammonia on the alginic acid aerogel, the characteristic band of the free carboxyl groups at 1735 $cm^{-1}$ disappears and is replaced by the C–O stretching bands of carboxylate at 1620 and 1575 $cm^{-1}$. This demonstrates that the carboxylic acids of the alginate are salified by $NH_4^+$ as effective Brønsted sites. The nearly complete disappearance of the carboxylic acid band indicates that nearly all the acid sites are accessible to the probe molecule. When the xerogel is equilibrated with the same

 

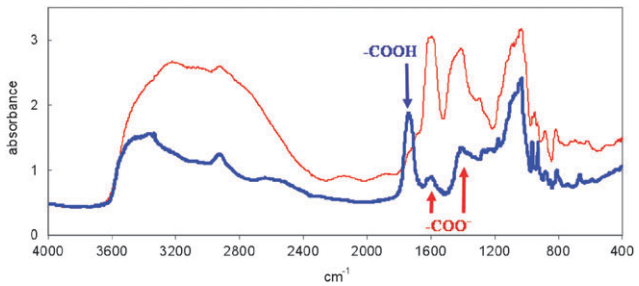


**Fig. 9** FT-IR spectra of aerogel (thin line) and xerogel (thick line) of alginic acid exposed to $NH_3$ vapor at 30 mbar pressure.

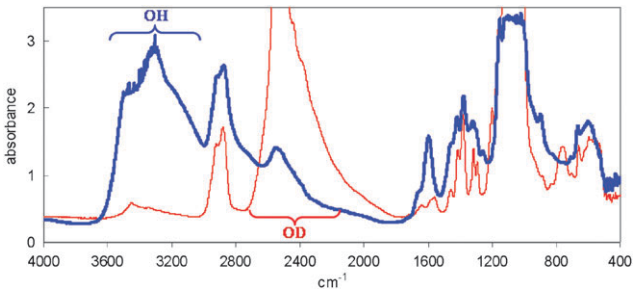


**Fig. 10** FT-IR spectra of aerogel (thin line) and xerogel (thick line) of chitosan exposed to $D_2O$ vapour at 15 mbar pressure.



pressure of $NH_3$, virtually no modifications of the 1735 $cm^{-1}$ carboxylic acid band are observed. The different accessibility of the acid functions in xerogel and aerogel can be explained by their different surface area. The surface area of the alginic acid aerogel was 390 $m^2\ g^{-1}$, while the surface area of the xerogel was lower than 2 $m^2\ g^{-1}$.

Ammonia interacts with alginate aerogel according to two distinct mechanisms: molecular adsorption on electron-acceptor sites (Lewis acid) or proton transfer from a Brønsted acid site and electrostatic bonding of the resulting ammonium cation to an anionic site of the adsorbent. Spectroscopic evidence shows that in alginate gel formed by alkaline earth cations, all carboxylic groups are salified. In this case, no proton exchange sites are available and the adsorption of ammonia takes place by interaction with the available Lewis acids, namely the alkaline earth cations. In the case of the ionotropic gels formed by transition metal cations, a significant fraction of carboxylic groups are not salified and ammonia adsorption takes place both on the divalent cation and by proton transfer from the available Brønsted sites.[90,91]

### Accessibility of the surface sites of chitosan aerogel

Transmission FT-IR spectroscopy has been used to monitor the deuteration by $D_2O$ vapour at room temperature of wafers of aerogels and xerogels of chitosan from β-chitin.[92] The spectra of aerogel and xerogel equilibrated with 15 mbar $D_2O$ differ in a striking manner (Fig. 10). In the case of the aerogel, the O–H and N–H stretching bands in the region 3430–3180 $cm^{-1}$ have nearly disappeared and have been replaced by the O–D and N–D stretching bands in the 2530–2370 $cm^{-1}$ region. In the case of the xerogel, equilibration with the same pressure of $D_2O$ for a much longer time marginally eroded the O–H and N–H bands and gave rise to very weak O–D and N–D bands. The difference of the surface area of the two solids (175 $m^2\ g^{-1}$ for the aerogel and 5 $m^2\ g^{-1}$ for the xerogel) accounts for the nearly complete deuteration of the alcohol and amine groups of the aerogel and the low reactivity of the xerogel. The deuterating agent is a probe of the accessibility of the materials to small polar molecules, and the difference observed indicates that the chitosan aerogel can be more effective than a xerogel for applications in catalysis or adsorption.

The immersion in organic low surface-tension solvents does not affect the texture of the aerogels. The accessibility of the primary amine functions of chitosan may be quantified in organic solvent by formation of a salicylaldimine Schiff base upon treatment with salicylaldehyde. Aerogel beads of chitosan from β-chitin in contact with salicylaldehyde become yellow and their UV-visible diffuse reflectance spectra exhibit the absorption band at 318 nm characteristic of the Schiff base. Quantitative GC analysis of the remaining salicylaldehyde in the solution allows to calculate the fraction of reacted amino groups, which corresponds to the fraction of accessible sites. In the aerogel formulation, up to 70% of the amine groups are accessible *vs.* 27% for the xerogel form.[93] The amine functionality of the polysaccharide framework could display intrinsic catalytic activity in base-catalyzed reactions.

## Applications in catalysis

Several properties of the polysaccharide aerogels render them promising materials for catalysis: their high surface area, the high density of their functional groups (up to 5.8 mmol $g^{-1}$ amino groups for chitosan, 5.6 mmol $g^{-1}$ carboxylic groups for alginic acid, 2.8 mmol $g^{-1}$ sulfate groups for κ-carrageenan), and the absence of diffusional limitations in their macroporous framework. The accessibility of the functional groups suggests that they can behave as organic solid acid or base catalyst depending on the nature of the polymer. With surface areas as high as 500 $m^2\ g^{-1}$, polysaccharides can compete with inorganic solids as supports for organometallic or metal catalysts. Shaping of the catalyst can be easily performed by adapting the method of contacting the polysaccharide solution and the gelling agent.[4,94–96] Up to date millimeter-size aerogel spheres have been used but formulations as membranes or hollow fibers are possible.[5]

### Polysaccharide aerogels as catalyst supports

The effectiveness of alginate aerogel microspheres as a catalyst support was evidenced in the reaction of substitution of an allyl carbonate with morpholine catalyzed by the hydrosoluble trisulfonated triphenylphosphine palladium(0) complex $Pd(TPPTS)_3$. The stability of the obtained catalyst was investigated in terms of textural stability and catalytic activity.[97]

The catalysts were prepared by impregnation of the outgassed aerogel beads by an aqueous solution of $Pd(TPPTS)_3$ complex synthetized *in situ*. Direct impregnation by water can originate a capillary tension able to draw together the alginate fibrils and destroy the texture of the gel. To avoid this effect, the aerogel beads were impregnated with anhydrous ethanol for 1 h before contacting the catalyst solution. After 30 min ripening at 40 °C, the beads were dehydrated by immersion in

two successive baths of anhydrous ethanol and dried under supercritical $CO_2$ conditions. This method of impregnation with the catalyst solution allowed to retain the textural properties of the materials. For example, the aerogel of an alginate with 20% guluronic residue with an initial surface area of 335 $m^2\ g^{-1}$ loses less than 1.5% of its surface area when loaded with 0.37% w/w Pd. The aerogel of an alginate with 74% guluronic residue retained its initial surface of 450 $m^2$ per gram of alginate when loaded with a similar amount of catalyst.[97]

The catalytic tests of allylic substitution of methylallyl carbonate with morpholine were performed in acetonitrile at 50 °C with 1% Pd/methyl allyl carbonate molar ratio. The course of the reaction was monitored by periodic samplings and quantitative analysis by gas chromatography (BP20 column). Quantitative yield was reached within 30 min and the same result was achieved in three successive catalytic runs. The catalytic activity was not affected by the guluronic/mannuronic ratio of the alginate. Considering the hydrophilic properties of both the support and the catalyst, the effect of water addition was studied, as was performed for aqueous-phase catalysts supported on freeze-dried polysaccharides.[98,99] In the case of catalysts prepared by impregnation of lyophilized polysaccharides, there was no catalytic activity in the absence of water. The activity increased with the absorbed amount of water, suggesting that the catalytic sites, not accessible in the dried material, became accessible due to the swelling of the support by water. On the contrary, in the case of catalysts supported on alginate aerogel, water had no effect on the activity: the high dispersion of the polymer allowed a good accessibility to the catalytic sites independently of the level of hydration.[97] Similar results in terms of conversion and recycling of the catalyst were obtained in ethanol.

The same procedure of catalyst synthesis was applied to other polysaccharides, such as κ-carrageenan and chitosan, to obtain some data on the influence of the chemical structure of the support. The differences in turnover numbers (mol product (mol Pd h)$^{-1}$), close to 500 for alginates, 190 for carrageenan and only 40 for chitosan, indicated that the activities were correlated to the electrostatic properties of the support. Carrageenans only bear one sulfate group per two saccharide monomers, while alginate presents one carboxylic group per monomer. In chitosan aerogels, 70% of the amino groups of the polysaccharide are accessible, and the sensitivity of the allylic substitution reaction to the pH of the medium could explain the very low activity obtained when the Pd complex was supported on this basic polysaccharide. This explanation does not rule out the possibility of a direct inhibition of the palladium complex by the amino groups of chitosan.

### Polysaccharide aerogels as organic catalysts

The amine functionality borne by the chitosan framework can display intrinsic catalytic activity in base catalyzed reactions. This was evidenced by the use of chitosan as a catalyst for fatty acid addition to glycidol, leading to monoglyceride formation.[93] The reactions were performed in toluene at 70 °C with 0.5 mmol of lauric acid, 0.5 mmol of glycidol and 30 mg aerogel beads of chitosan from β-chitin. A yield of 66% was reached in 24 h. This result can be compared with the inactivity of lyophilised chitosan tested in the same conditions. The activity of the chitosan aerogel was comparable to the activity of amine-functionalized silicas.[100]

When chitosan was used as a component of organic-silica composites dried in $CO_2$ supercritical conditions, the amino groups were still accessible and developed the same catalytic properties as in the case of the all-chitosan aerogel. In the case of a composite with a homogeneous distribution of silica and chitosan, the conversion of lauric acid reached 94% in 50 h with a selectivity of 98% in monoglyceride also after three consecutive reaction runs.[101] In the case of core–shell materials, the catalytic activity was not significant, due to a very low accessibility of the amino groups.

For the chitosan–titania composites, the inorganic component of the hybrid played a synergistic role in the catalytic activity.[102] When titania was generated inside the chitosan alcogel beads, the $CO_2$ supercritical drying of the material afforded solids with 25% wt titania with surface areas close to 480 $m^2\ g^{-1}$. The coexistence of amino groups as nucleophilic activators and titania nanoparticles as electrophilic activators led to a highly active catalyst for lauric acid addition to glycidol. Conversion of 90% with a selectivity of 100% was reached in 5 h. The catalyst was recycled three times without significant loss in activity or selectivity.

### Chitosan aerogels as bifunctional catalysts

The immobilisation of water-soluble metallophthalocyanine complexes (MPcS) on chitosan aerogel microspheres afforded new bifunctional catalysts which have been used for the aerobic oxidation of β-isophorone.[103] Chitosan performed a dual role, as support of the metal complex and as the organic base necessary for the reaction. Chitosan-based metal phthalocyanine (M = Fe, Co) catalysts were prepared by incipient wetness impregnation of chitosan aerogel beads with an aqueous solution of sulfonated phthalocyanines of Fe or Co. The solids containing phthalocyanine complexes were characterized by diffuse reflectance UV-Vis spectra. Interestingly, FePcS supported on the alginate was in a dimeric form, while the amount of monomeric and dimeric species was comparable in the case of supported CoPcS, indicating a lower tendency of CoPcS to form aggregates on the support. The amount of CoPcS dimeric species increased with the loading of phthalocyanine complexes.

The catalysts were tested in the heterogeneous oxidation of β-isophorone resulting in the formation of ketoisophorone (KIP), allylic alcohol and α-isophorone. CoPcS@chitosan performed much better than FePcS@chitosan and provided 62% conversion and 48% KIP selectivity (respectively 48 and 17% for Fe, 1% mol catalyst). On recycling, about 90% of the previous activity was retained and the KIP selectivity remained fairly constant.

## Perspectives

Aerogel formulations open the way to the exploitation of the surface properties and the high dispersion of the large family of polysaccharides for reactions at the interface between the polymer and a gas or an organic solvent. Applications in catalysis, adsorption and chemical sensing can take advantage

 

of the reactivity of the functional groups of the polymers or of catalytic sites in electrostatic or covalent interaction with the polysaccharide. The easy shaping of the gels into the desired morphology is another desirable property. The mechanical properties of these organic aerogels are still under study, but they are expected to be significantly less brittle than inorganic or hybrid materials with the same degree of dispersion.

Aerogel formulation allows the extension to hydrocolloid derivatives of the techniques classically used for the characterization of inorganic solids, in particular those implying a high vacuum environment. Can these techniques provide a better understanding of the organisation of polymers in aqueous and natural systems? A positive answer requires the aerogel to be a good image of the parent hydrogel. This point is currently under investigation and polysaccharide gels are being studied by small-angle X-ray scattering (SAXS) at different steps of conversion from gel to aerogel in order to determine the relations between the polymer organization at the nanoscale in the hydrogel and the final dry aerogel.

Albeit polysaccharide aerogels are very promising materials, some drawbacks of the preparation method and of the properties of the final materials have to be addressed. The main limitation of the aerogel preparation method is the need for an intermediate solvent in order to obtain an effective replacement of water by liquid $CO_2$. Also if relatively environment-friendly solvent can be used, the minimisation of their use is needed to optimise the economical and environmental impact of the aerogel preparation. Moreover, the sensitivity of the texture of the aerogels to the presence of menisci of high surface-tension liquid puts limitations to the conditions of manufacturing, application and storage.

Also the high surface/mass ratio, a desirable property for materials such as catalysts and adsorbents, can be a missed blessing in operating conditions. The high surface area is often related to a very high porosity. This is a desirable property as far as diffusion properties are concerned but can bring about a low surface/volume ratio, implying that very high vessel volumes may be needed for catalytic reactors or adsorption contactors. As a consequence, the control of the porosity of the aerogel at optimum values between the contradictory requirements of diffusional properties and available volume is a main issue in aerogel preparation. Finally, one has to take into account the thermal stability of these organic polymers, whose decomposition often begins below 200 °C.

Nevertheless, aerogels of natural polysaccharides are promising candidates for many applications, as they couple the textural properties of highly accessible materials with the versatile chemistry of hydrocolloids. The appeal of such less energy-intensive materials than those presently used as catalyst and catalyst supports is a main driving force for the development of biomass-derived replacements.

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## References

1. D. J. McHugh, *A Guide to the Seaweed Industry*, FAO, Rome, 2003, p. 105.
2. G. P. Towler, A. R. Oroskar and S. E. Smith, *Environ. Prog.*, 2004, **23**, 334.
3. E. Guibal, *Prog. Polym. Sci.*, 2005, **30**, 71.
4. E. Guibal, T. Vincent and F. Peirano-Blondet, *Ion Exchange and Solvent Extraction*, ed. A. K. S. Gupta, CRC Press, Boca Raton, FL, 2007, p. 151.
5. T. Vincent and E. Guibal, *Environ. Sci. Technol.*, 2004, **38**, 4233.
6. S. Doi, J. H. Clark, D. J. Macquarrie and K. Milkowsi, *Chem. Commun.*, 2002, 2632.
7. V. L. Budarin, J. H. Clark, R. Luque, D. J. Macquarrie and R. J. White, *Green Chem.*, 2008, **10**, 382.
8. R. J. White, V. L. Budarin and J. H. Clark, *ChemSusChem*, 2008, **1**, 408.
9. A. C. Pierre and G. M. Pajonk, *Chem. Rev.*, 2002, **102**, 4243.
10. R. W. Pekala, *J. Mater. Sci.*, 1989, **24**, 3221.
11. C. Ouverx, N. Velings, M. M. Mestdagh and M. A. V. Axelos, *Polym. Gels Networks*, 1998, **6**, 393.
12. N. O. Dhoot and M. A. Wheatley, *J. Pharm. Sci.*, 2003, **92**, 679, and references therein.
13. H. H. Tønnesen and J. Karlsen, *Drug Dev. Ind. Pharm.*, 2002, **28**, 621.
14. O. Smidsrød, *Faraday Discuss. Chem. Soc.*, 1974, **57**, 263.
15. N. Velings and M. M. Mestdagh, *Polym. Gels Networks*, 1995, **3**, 311.
16. C. Ouverx, N. Velings, M. M. Mestdagh and M. A. V. Axelos, *Polym. Gels Networks*, 1998, **6**, 393.
17. K. I. Draget, G. Skjak-Braek and O. Smidsrǿd, *Carbohydr. Polym.*, 1994, **25**, 31.
18. X. Wang and H. G. Spencer, *Polymer*, 1998, **39**, 2759.
19. C. K. Siew, P. A. Williams and N. W. G. Young, *Biomacromolecules*, 2005, **6**, 963.
20. G. T. Grant, E. R. Morris, D. A. Rees, P. J. C. Smith and D. Thom, *FEBS Lett.*, 1973, **32**, 195.
21. E. R. Morris, D. A. Rees, D. Thom and J. Boyd, *Carbohydr. Res.*, 1978, **66**, 145.
22. I. Braccini and S. Pérez, *Biomacromolecules*, 2001, **2**, 1089.
23. L. Li, Y. Fang, R. Vreeker and I. Appelqvist, *Biomacromolecules*, 2007, **8**, 464.
24. I. L. Mongar and A. Wassermann, *J. Chem. Soc.*, 1952, 492.
25. K. I. Draget, *Annu. Trans. Nord. Rheol. Soc.*, 2001, **8**, 149.
26. K. I. Draget, B. T. Stokke, Y. Yuguchi, H. Urakawa and K. Kajiwara, *Biomacromolecules*, 2003, **4**, 1661.
27. J. N. Barbotin and J. E. Nava Saucedo, in *Polysaccharides*, ed. S Dumitriu, Marcel Dekker, New York, 1998, p. 749.
28. D. Slootmaekers and M. Mandel, *Int. J. Biol. Macromol.*, 1990, **13**, 17.
29. C. Rochas and M. Rinaudo, *Biopolymers*, 1980, **19**, 1675.
30. J. Borgstrom, L. Piculell, C. Viebke and Y. Talmon, *Int. J. Biol. Macromol.*, 1996, **18**, 223.
31. P. K. Walsh, F. V. Isdell, S. M. Noone, M. G. O'Donovan and D. M. Malone, *Enzyme Microb. Technol.*, 1996, **18**, 366.
32. O. Sipahigil and B. Dortunc, *Int. J. Pharm.*, 2001, **228**, 119.
33. G. A. F. Roberts, *Chitin Chemistry*, Macmillan, London, 1992, p. 274.
34. P. Sorlier, A. Denuzière, C. Viton and A. Domard, *Biomacromolecules*, 2001, **2**, 765.
35. M. N. V. Ravi Kumar, R. A. A. Muzzarelli, C. Muzzarelli, H. Dashiwa and A. J. Domb, *Chem. Rev.*, 2004, **104**, 6017.
36. P. Gerentesa, L. Vachoud, J. Dourya and A. Domard, *Biomaterials*, 2002, **23**, 1295.
37. V. R. Sinha, A. K. Singla, S. Wadhawan, R. Kaushik, R. Kumria, K. Bansal and S. Dhawan, *Int. J. Pharm.*, 2004, **274**, 1.
38. J. G. Winterowd and P. A. Sandford, in *Food Polysaccharides and their Applications*, ed. A. M. Stephen, Marcel Dekker, New York, 1995, p. 441.
39. J. Berger, M. Reist, J. M. Mayer, O. Felt, N. A. Peppas and R. Gurny, *Eur. J. Pharm. Biopharm.*, 2004, **57**, 19.
40. J. Berger, M. Reist, J. M. Mayer, O. Felt and R. Gurny, *Eur. J. Pharm. Biopharm.*, 2004, **57**, 35.

41. R. Maruca, B. J. Suder and J. P. Wightman, *J. Appl. Polym. Sci.*, 1982, **27**, 4827.
42. H. Q. Shi, W. B. Tsai, M. D. Garrison, S. Ferrari and B. D. Ratner, *Nature*, 1999, **398**, 593.
43. X. Z. Shu and K. J. Zhu, *J. Microencapsulation*, 2001, **18**, 237.
44. Y. Koyama and A. Taniguchi, *J. Appl. Polym. Sci.*, 1986, **31**, 1951.
45. M. M. Amiji, *Carbohydr. Polym.*, 1997, **32**, 193.
46. X. Zeng and E. Ruckenstein, *J. Membr. Sci.*, 1998, **148**, 195.
47. G. A. F. Roberts and G. K. Moore, *Makromol. Chem.*, 1989, **190**, 951.
48. Y. C. Wei, S. M. Hudson, J. M. Mayer and D. L. Kaplan, *J. Polym. Sci., Part A: Polym. Chem.* , 1992, **30**, 2187.
49. X. Zeng and E. Ruckenstein, *Ind. Eng. Chem. Res.*, 1998, **37**, 159–165.
50. J. J. E. Hardy, S. Hubert, D. J. Macquarrie and A. J. Wilson, *Green Chem.*, 2004, **6**, 53.
51. K. Justi, M. C. M. Laranjeira, A. Neves, A. S. Mangrich and V. F. Favere, *Polymer*, 2004, **45**, 6285.
52. T. Vincent and E. Guibal, *Ind. Eng. Chem. Res.*, 2002, **41**, 5158.
53. Y. An, D. Yuan, M. Y. Huang and Y. Y. Jiang, *Macromol. Symp.*, 1994, **80**, 257.
54. T. M. Tang, M. Y. Huang and Y. Y. Jiang, *Macromol. Rapid Commun.*, 1994, **15**, 527.
55. H. S. Han, S. N. Jiang, M. Huang and Y. Y. Jiang, *Polym. Adv. Technol.*, 1996, **7**, 704.
56. T. Vincent and E. Guibal, *Langmuir*, 2003, **19**, 8475.
57. H. Wang, W. Sun and C. Xia, *J. Mol. Catal. A: Chem.*, 2003, **206**, 1995.
58. Y. Chang, Y. P. Wang and Z. X. Su, *J. Appl. Polym. Sci.*, 2002, **83**, 2188.
59. R. A. A. Muzzarelli, *Natural Chelating Polymers*, Pergamon Press, Oxford, 1973, p. 83.
60. G. A. F. Roberts, in ref. 33; p. 206.
61. O. Sipahigil and B. Dortunc, *Int. J. Pharm.*, 2001, **228**, 119.
62. A. Martinsen, I. Storrø and G. Skjåk-Bræk, *Biotechnol. Bioeng.*, 1992, **39**, 186.
63. J. C. P. Broekhoff and J. H. de Boer, *J. Catal.*, 1968, **10**, 377.
64. A. Galarneau, D. Desplantier, R. Dutartre and F. Di Renzo, *Microporous Mesoporous Mater.*, 1999, **27**, 297.
65. A. V. G. Neimark and P. I. Ravikovitch, *Microporous Mesoporous Mater.*, 2001, **44**, 697.
66. R. Valentin, K. Molvinger, F. Quignard and F. Di Renzo, *Macromol. Symp.*, 2005, **222**, 93.
67. F. Di Renzo, R. Valentin, M. Boissière, A. Tourrette, G. Sparapano, K. Molvinger, J. M. Devoisselle, C. Gérardin and F. Quignard, *Chem. Mater.*, 2005, **17**, 4693.
68. S. Partap, I. Rehman, J. R. Jones and J. A. Darr, *Adv. Mater.*, 2006, **18**, 501.
69. D. Casson and A. N. Emery, *Enzyme Microb. Technol.*, 1987, **9**, 102.
70. B. P. Hills, J. Godward, M. Debatty, L. Barras, C. P. Saturio and C. Ouwerx, *Magn. Reson. Chem.*, 2000, **38**, 719.
71. D. Serp, M. Mueller, U. von Stockar and I. W. Marison, *Biotechnol. Bioeng.*, 2002, **79**, 243.
72. B. Thu, O. Gåserød, D. Paus, A. Mikkelsen, G. S. Skjåk-Bræk, R. Toffanin, F. Vittur and R. Rizzo, *Biopolymers*, 2000, **53**, 60.
73. G. J. Kennedy and J. H. Knox, *J. Chromatogr. Sci.*, 1972, **10**, 549.
74. G. W. Scherer, D. M. Smith and D. Stein, *J. Non-Cryst. Solids*, 1995, **186**, 109.
75. T. Wognier, L. Duffours, J. Phalippou, P. Delord and V. Gibiat, *J. Sol–Gel Sci. Technol.*, 1997, **8**, 789.
76. R. Pirard and J. P. Pirard, *J. Non-Cryst. Solids*, 1997, **212**, 262.
77. R. Pirard, S. Blacher, F. Brouers and J. P. Pirard, *J. Mater. Res.*, 1995, **10**, 2114.
78. F. Rouquerol, J. Rouquerol and K. S. W. Sing, *Adsorption by Powders and Porous Solids*, Academic Press, San Diego, CA, 1999.
79. G. Reichenauer and G. W. Scherer, *J. Non-Cryst. Solids*, 2000, **277**, 162.
80. G. Reichenauer and G. W. Scherer, *Colloids Surf., A*, 2001, **187**, 41.
81. E. D. T. Atkins, I. A. Nieduszynski, W. Mackie, K. D. Parker and E. E. Smolko, *Biopolymers*, 1973, **12**, 1865.
82. E. D. T. Atkins, I. A. Nieduszynski, W. Mackie, K. D. Parker and E. E. Smolko, *Biopolymers*, 1973, **12**, 1879.
83. W. Mackie, S. Pérez, R. Rizzo, F. Taravel and M. Vignon, *Int. J. Biol. Macromol.*, 1983, **5**, 329.
84. K. Ogawa, T. Yui and K. Okuyama, *Int. J. Biol. Macromol.*, 2004, **34**, 1.
85. A. Tourrette, L. Vachoud, C. Tourné-Péteilh, S. Bégu, J. M. Devoisselle and F. Quignard, *Adv. Chitin Sci.*, 2006, **9**, 284.
86. G. Lamarque, C. Viton and A. Domard, *Biomacromolecules*, 2004, **5**, 992.
87. A. Clayer, L. Vachoud, C. Viton and A. Domard, *Macromol. Symp.*, 2003, **200**, 1.
88. R. Valentin, PhD thesis, Université Montpellier 2, Montpellier, France, 2005.
89. D. Capitani, A. A. De Angelis, V. Crescenzi, G. Masci and A. L. Segre, *Carbohydr. Polym.*, 2001, **45**, 245.
90. R. Valentin, R. Horga, B. Bonelli, E. Garrone, F. Di Renzo and F. Quignard, *Macromol. Symp.*, 2005, **230**, 71.
91. R. Valentin, R. Horga, B. Bonelli, E. Garrone, F. Di Renzo and F. Quignard, *Biomacromolecules*, 2006, **7**, 877.
92. R. Valentin, B. Bonelli, E. Garrone, F. Di Renzo and F. Quignard, *Biomacromolecules*, 2007, **8**, 3646.
93. R. Valentin, K. Molvinger, F. Quignard and D. Brunel, *New J. Chem.*, 2003, **27**, 1690.
94. M. Amaike, Y. Senoo and H. Yamamoto, *Macromol. Rapid Commun.*, 1998, **19**, 287.
95. T. Vincent and E. Guibal, *Solvent Extr. Ion Exch.*, 2000, **18**, 1241.
96. F. L. Mi, S. S. Shyu, Y. B. Wu, S. T. Lee, J. Y. Shyong and R. N. Huang, *Biomaterials*, 2001, **22**, 165.
97. R. Valentin, K. Molvinger, C. Viton, A. Domard and F. Quignard, *Biomacromolecules*, 2005, **6**, 2785.
98. F. Quignard, A. Choplin and A. Domard, *Langmuir*, 2000, **16**, 9106.
99. P. Buisson and F. Quignard, *Aust. J. Chem.*, 2002, **55**, 73.
100. A. Cauvel, G. Renard and D. Brunel, *J. Org. Chem.*, 1997, **62**, 749.
101. K. Molvinger, F. Quignard, D. Brunel, M. Boissière and J. M. Devoisselle, *Chem. Mater.*, 2004, **16**, 3367.
102. A. El Kadib, K. Molvinger, C. Guimon, F. Quignard and D. Brunel, *Chem. Mater.*, 2008, **20**, 2198.
103. A. B. Sorokin, F. Quignard, R. Valentin and S. Mangematin, *Appl. Catal., A*, 2006, **309**, 162.


 