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Porous Chitosan-Silica Hybrid Microspheres as a Potential Catalyst

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Chitosan/silica hybrids were synthesized and shaped as microspheres, which may or may not present a core/shell structure, depending on the experimental conditions. The core is constituted by a homogeneous hybrid, while the shell is pure silica. The amino groups of the biopolymer are still accessible as active sites for heterogeneous catalysis.

Introduction

Amphiphilic organic molecules are self-assembled into various supramolecular structures. These self-assemblies can be used as a template to create novel mesophasic inorganic materials. Taking inspiration from nature, many recent articles focused on the union of inorganic materials and supramolecular organic chemistry.[1–3] Shinkaï et al. recently described the preparation of novel silica fibers by using a cholesterol-based gelator,[4] which acts as a template in the TEOS polymerization process. The silicic solid obtained after calcinations demonstrated a helical structure. In the same time, Moreau et al. reported an ingenious approach to form chiral hybrid organic–inorganic solids by means of organogelators.[5] These compounds, although similar in structure to those of Shinkaï, differ in that the silica precursor had been covalently attached to the gelator structure. Another strategy dealt with the use of biopolymers such as polysaccharides to synthesize bioinspired hybrid materials.[6]

Among the polysaccharides, chitosan is a copolymer of linked β,(1→4),2-amino-2-deoxy-D-glucan and 2-acetamidodeoxy-D-glucan (Scheme 1). It is mainly used in medicine and food industries.[7]

The presence of hydroxyl and amino groups, which are excellent functional groups for the anchoring of a large variety of organometallic complexes, makes chitosan a good candidate as a precursor for heterogeneous molecular catalysts.[8,9] Moreover, the amino groups are potential base catalysts. Different approaches were

Scheme 1. Chemical Structure of Chitosan
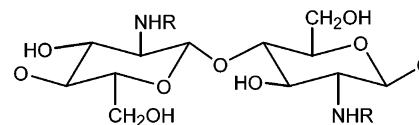
R: H, COCH$_3$

reported in the literature concerning the involvement of chitosan in polysaccharide/silica hybrids. Schulz et al. used chitosan as a template for the synthesis of mesoporous silica.[10] A fibrous material was obtained, the pore radius distribution being polymodal. The authors suggest that this special structure results both from the aggregation of the hydrated chitosan helices in bundles of parallel fibers with different size and from the gelation of the system. Hunt et al. synthesized chitosan/silica hybrid gels,[11] also. In this study, drying under supercritical CO_2 conditions led to aerogel with high surface area. Possible applications are envisaged like wastewater treatment using the chelating effect of the included chitosan. Monteiro et al. prepared new organic–inorganic hybrids from chitosan and aminosilylating agents.[12] Both chitosan and silylating agents have the amine groups cross-linked through linear glutaraldehyde units. The resulting dried hydrogels exhibited a lamellar-like surface morphology.

Our strategy was based on the application of the hybrid materials in catalysis. Our aim was, in particular, to benefit from the amino groups. We reported recently the efficiency of a supercritical drying by CO_2 on polysaccharide gels,[13] which favors the formation of porous materials. Hence, this procedure was adopted in the present work.

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Experimental Section

Synthesis of Hybrid Materials (Samples A, B, C, and D). Sample A: An aqueous solution of chitosan (50 mg) in 4 cm³ of a mixture of acetic acid/water (3/1) was mixed with 0.5 cm³ of tetraethoxysilane (TEOS). A gel was formed immediately, in less than 1 min. The gel matured overnight, and then it was washed several times with distilled water in order to remove acetic acid. The gel was then dehydrated by immersion in a series of successive ethanol–water baths (50 cm³) of increasing alcohol concentration (10, 30, 50, 70, 90, and 100%) for 15 min each. Then the hybrid material was dried under supercritical CO_2 conditions (74 bar, 31.5 °C) in a Polaron 3100 apparatus.

Sample B: A few milligrams of sample A were calcinated at 500 °C in order to remove the organic phase.

Sample C: 4 cm³ of a mixture of acetic acid and water (3/1) was mixed with 0.5 cm³ of TEOS. Ten days later a gel was formed. It was washed, dehydrated by ethanol, and dried as described previously for sample A.

Sample D: 50 mg of chitosan dissolved in 16 cm³ of an aqueous acetic acid solution of 1% v/v was added to 120 mg of SDS (sodium dodecyl sulfate) and 0.05 mol of TEOS. Two phases were obtained and each phase was washed, dehydrated, and dried as described for sample A. One phase was only pure silica whereas the other one was hybrid chitosan/silica. The first one was not investigated.

Synthesis of Chitosan Microspheres. An aqueous solution of chitosan was obtained by dissolving 1 g of chitosan in 100 cm³ of a solution of acetic acid, 0.055 mol·dm⁻³. This corresponds to a stoichiometric amount of acid with respect to the amount of NH_2 functions. Total dissolution was obtained under stirring over one night at room temperature. This solution was dropped into a NaOH solution (4 N) through a 0.8-mm gauge syringe needle. The chitosan beads were stored in the alkaline solution for 2 h (these beads were directly used for microspheres **1**) and then dehydrated by immersion in a series of successive ethanol–water baths as for sample A (these beads were directly used for the synthesis of microspheres **2**). Then the microspheres were dried under supercritical CO_2 conditions (74 bar, 31.5 °C) in a Polaron 3100 apparatus (these beads were directly used for the synthesis of microspheres **3**).

Synthesis of Hybrid Microspheres 1. Hydrated chitosan microspheres (corresponding to 300 mg of dry chitosan) were put in an excess of tetraethoxysilane (0.12 mol of TEOS per gram of chitosan). The hydrolysis proceeded for 12 h. Fifty milligrams of NaF (1.2×10^{-3} mol) was added to catalyze the condensation of silica. The mixture matured for 12 h. Then the beads were washed by distilled water and then dehydrated by immersion in a series of successive ethanol–water baths (150 cm³) as for sample A. Additional absolute ethanol was used to accomplish complete exchange of the water in the microspheres by ethanol before drying under supercritical CO_2 (scCO₂) (74 bar, 31.5 °C) in a Polaron 3100 apparatus.

Synthesis of Hybrid Microspheres 2. Four grams of chitosan microspheres in ethanolic media were suspended in an excess of a mixture of 75% of tetraethoxysilane (TEOS) (6 cm³) and 25% of water (2 cm³). The condensation of silica was catalyzed by NaF (100 mg). The mixture matured for 24 h under stirring. Then the beads were washed and dried as for microspheres **1**.

Synthesis of Hybrid Microsphere 3. Fifty milligrams of chitosan microspheres dried by supercritical CO_2 were put in a mixture of 0.5 cm³ of TEOS, 490 mg of H_2O/EtOH/urea (1/5/1 in weight), and 1 cm³ of water. The mixture matured for 24 h. Then the beads were washed and dried as for microspheres **1**.

Characterization of Materials. (1) SEM images were obtained using an Hitachi S4500I microscope. (2) Nitrogen sorption isotherms were obtained with an ASAP 2010 Micromeritics apparatus. The surface area was determined using BET equation and micropores volume was calculated by the t-plot method (Harkins and Jura). (3) Thermogravimetric analysis was performed with a Netzsch TG 209 C apparatus.

Quantitative GC Analysis of the Remaining Salicylaldehyde in the Solution. In a typical experiment, 33.8 mg of microspheres was put in 2.5 cm³ of a solution of salicylaldehyde (0.03 mol·dm⁻³) in ethanol. The aldehyde amount remaining in solution was determined by GC analysis (FID, BP20, $T_{inj.}$ = 250 °C, $T_{det.}$ = 270 °C, oven 50–250 °C, 20 °C·min⁻¹) according to a calibration curve.

Catalytic Test. In a typical experiment 0.5 mmol of lauric acid and 0.5 mmol of glycidol were injected in a flask containing 30 mg of hybrid beads in 4 cm³ of toluene. Zero time of reaction was taken when temperature reached 70 °C. Reactants and products amounts were determined by GC analysis (FID, HP5, $T_{inj.}$ = 250 °C, $T_{det.}$ = 275 °C, oven 50–250 °C, 15 °C·min⁻¹) with octanol as the internal standard according to a calibration curve established with authentic samples.

Results and Discussion

Our first experiments were directly derived from the work of Shinkai[4] and Hunt.[11] The materials were prepared by mixing chitosan in acetic acid with tetraethoxysilane (TEOS).[14] Sample A corresponds to the hybrid material obtained immediately after supercritical CO_2 (scCO₂) drying. Sample B is the calcinated sample A. Sample C is the blank corresponding to the pure silica prepared under the same conditions as the previous samples. Sample D corresponds to the hybrid prepared with SDS (sodium dodecyl sulfate). The four samples were observed in SEM, and nitrogen sorption was done in order to confirm the textural properties.

The study of the SEM images shows the differences between the hybrid material (Figure 1a) and the witness sample (Figure 1c). Nanoparticles were obtained in the case of silica alone, although long fibers were present in the case of the hybrid material. When calcinated (Figure 1b), the structure of the mineral oxide is similar to the hybrid before calcination. This means that the chitosan organized the silica and that the sol–gel transcription was efficient. The initial ratio silica/chitosan of 2.7 was maintained in the hybrid according to the thermogravimetric analysis (silica/chitosan = 2.6).

The nitrogen adsorption/desorption isotherms of samples A, B, C, and D are of type IV in the IUPAC classification, characteristic of a mesoporous material. The data were reported in Table 1.

Although surface areas were very high, two phases were observed by MEB. One was constituted of pure silica, and the other was a hybrid containing 30 wt % chitosan. Unfortunately, the results suffered from a lack of reproductivity in the composition of the hybrid. Some of the experimental parameters were modified in order to overcome this problem: (i) Use of diluted acetic acid to avoid the degradation of the chitosan. In this case the hybrid contains 37 wt % chitosan (Figure 2a). (ii) Presence of SDS (sodium dodecyl sulfate) as a surfactant (sample D). The hybrid contains 67 wt % chitosan (Figure 2b). (iii) Change in the drying process. Acetone was used instead of ethanol before drying under scCO₂ conditions. In all the cases some hybrids were obtained which differed in their composition, texture, and surface area. They were still not satisfying in terms of composition homogeneity, particularly during the preparation of sample D. Another pure silica phase has been
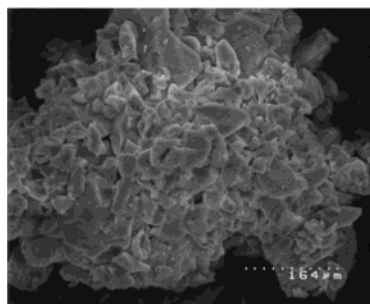
(14) An aqueous solution of chitosan is prepared by dissolving 200 mg of chitosan in 12 cm³ of acetic acid and 3 cm³ of distilled water. Then 2 cm³ of tetraethoxysilane (TEOS) and a few drops of distilled water were added. The mixture was stirred for 5 min.

(a)

(b)
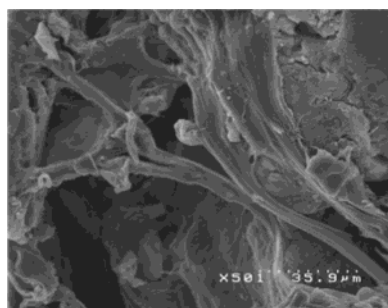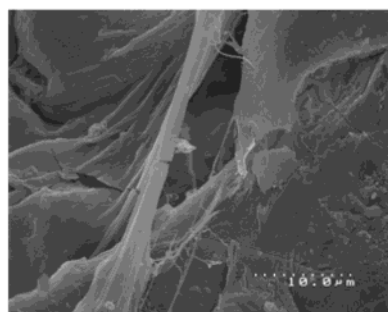
Figure 1. SEM images of chitosan/silica hybrid (a) before calcination, (b) after calcination, and (c) pure silica witness.


(c)

Table 1. Textural Characterization of Hybrid Materials

sample	surf. area $(m^2 \cdot g^{-1})$	vol. micropores $(cm^3 \cdot g^{-1})$
A	300	0.029
B	500	0.061
C	910	0.123
D	590	0.072

separated. Moreover, the morphologies of the particles were not well-defined (Figure 2). Homogeneity and reproducibility are prerequisite conditions for a catalytic application.

In the one-pot synthesis, it was difficult to control both the self-assembling of the polymer and the rate of silica condensation. We decided thus to synthesize the hybrid in two steps. The first one induces an auto-assembling of the polymer chains as a gel. The second one concerns the silica condensation. To control the morphology of the final hybrid material, chitosan is shaped as microspheres according to a classical procedure[12] and then tetraethoxysilane (TEOS) was added as a silica precursor with NaF as a catalyst to improve
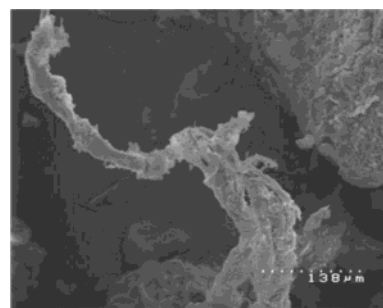
(a)
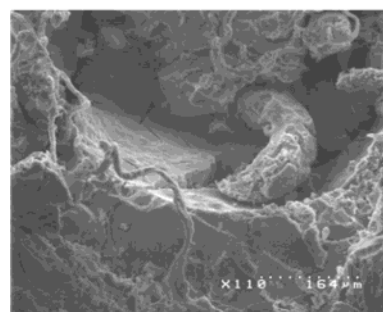
(b)

Figure 2. SEM of chitosan/silica hybrid: (a) synthesis in diluted acetic acid and (b) synthesis in the presence of SDS.

the formation of silica by sol−gel through a specific nucleophilic assistance.[15] For microspheres **3**, the condensation of silica was conducted in the presence of urea as porogen agent. In this case the presence of NaF was not necessary because the amine groups catalyze the silica condensation.[16] The structure of the hybrid microspheres depends on the maturation time. The SEM pictures of the hybrid microsphere **1** (Figure 3, a_1−b_1) reveal the fibrous character of the hybrid, very similar to the pure chitosan beads with a preservation of the bead morphology. The beads contain 19% of chitosan (TG analysis). The homogeneity of the hybrid composition is evidenced by EDX analysis. On the contrary, the SEM picture of the hybrid microsphere **2** (Figure 3, a_2), containing 16% of chitosan, shows a core/shell structure. The core is porous (Figure 3, b_2) and here, again, compared well with the pure chitosan beads.

The radial distribution of the SiO_2 percentage is reported in Figure 4 for the microsphere **2**. The SEM observation is confirmed; the shell is constituted of pure silica, while the core is a homogeneous chitosan/silica hybrid at least until very close to the center of the sphere. The value of 80% silica is in agreement with the thermogravimetric analysis. Microspheres **3** present the same core/shell structure.

To study the mineral part of the material, calcination of the organic part of the hybrid **2** was conducted at 500 °C under air. The microscopy pictures show that the texture of the core of the bead is preserved during the calcination (Figure 5). The EDX analysis confirms that silica is the only component.

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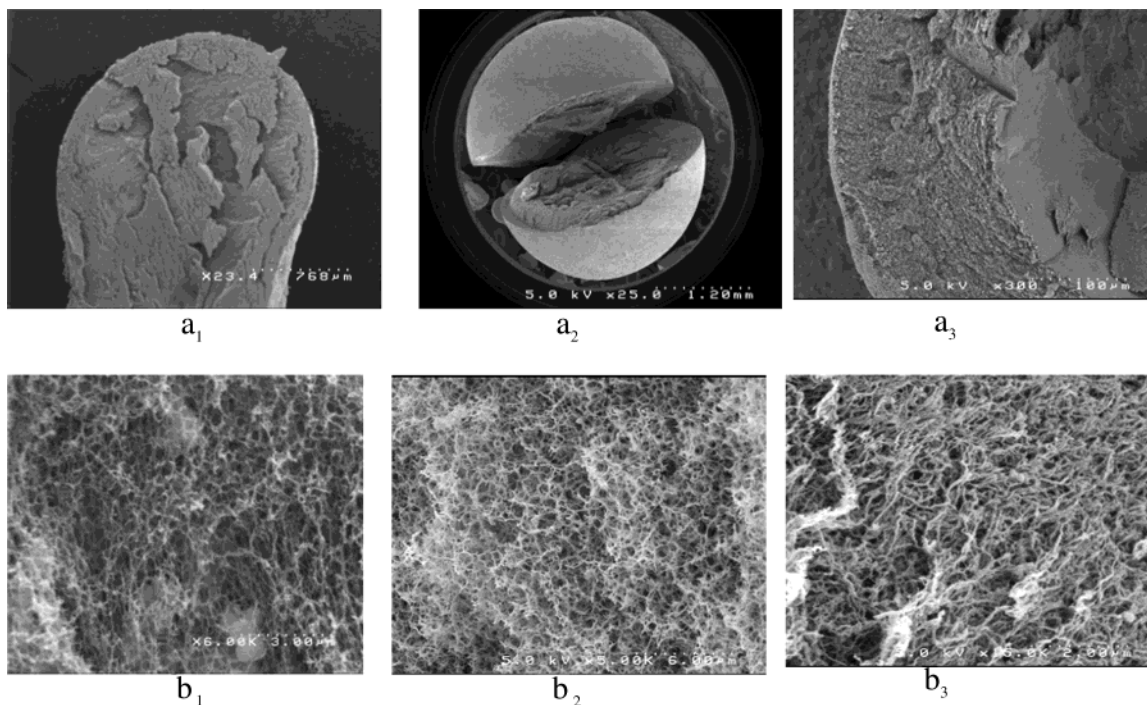
Figure 3. SEM of hybrid beads **1** (a$_1$−b$_1$), **2** (a$_2$−b$_2$), and **3** (a$_3$−b$_3$).
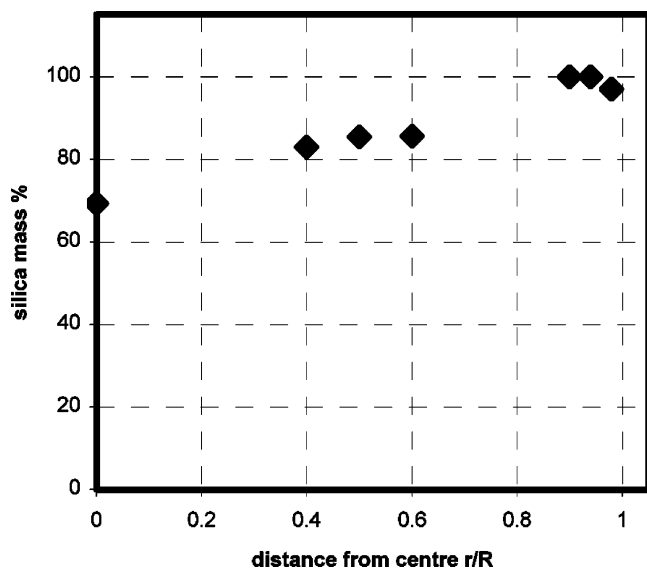

Figure 4. EDX of the hybrid microsphere **2**.

Figure 5. SEM of the hybrid microsphere **2** after calcination.

The textural properties of the hybrids were deduced from the classical nitrogen sorption volumetric analysis of microspheres degassed for 10 h at 80 °C. The isotherms reflect the presence of both micropores and macropores, as well as the absence of mesopores (Figure 6). The external surface areas obtained with the t-plot slope and volumes of micropores are reported in Table 2. It should be noted that the results are reproducible, even though the surface area was not so high as those observed with samples A and D.

The role of urea as a pore-forming agent was successful as confirmed by the lower microporous character of the microspheres **3**. Moreover, in this case the external surface area was increased.

The choice of chitosan was dictated by the possibility that the amino functions could display intrinsic catalytic activity provided that the reactants easily access those
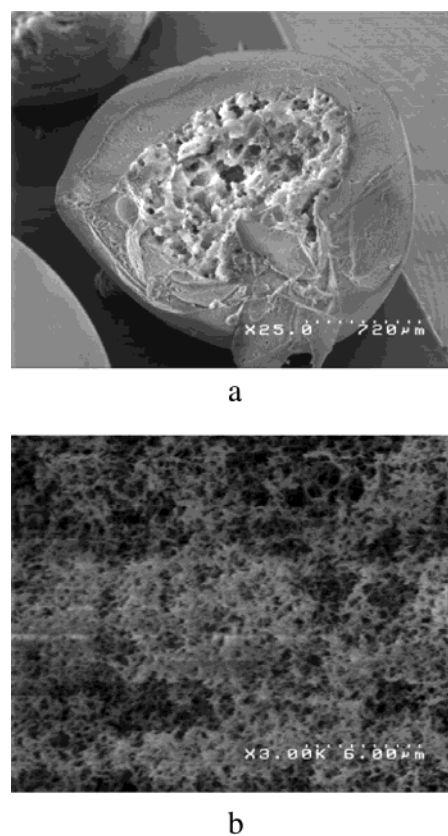
sites. To check this point, we quantified the accessibility of the primary amine functions in the hybrid by the formation of a Schiff base with salicylaldehyde, as described in ref 13. The experiment was conducted in ethanol. We verified that salicylaldehyde does not adsorb either on the silicic surface part of the hybrid or on the organic part behaving as polar OH groups with proof experiments on pure silica and cellulose. When
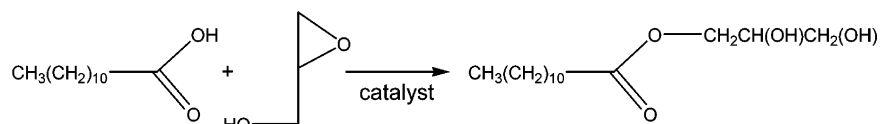
Scheme 2. Monoglyceride Synthesis

$$CH_3(CH_2)_{10}\text{—COOH} + \text{(epoxide/glycidol)} \xrightarrow{\text{catalyst}} CH_3(CH_2)_{10}\text{—C(=O)—O—CH}_2CH(OH)CH_2(OH)$$

Table 2. Textural Characterization of Microspheres

sample	surf. area $(m^2 \cdot g^{-1})$	vol. micropores $cm^3 \cdot g^{-1}$
chitosan	114	0.02
microsphere 1	73	0.127
microsphere 2	56	0.029
microsphere 3	149	0.016

Table 3. Percentage of Accessible Amine Groups for the Different Microspheres
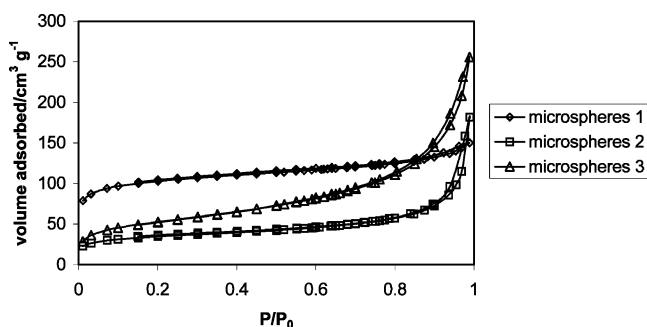
microspheres	% accessible amine groups (23 h)
chitosan	48
1	70
2	7
3	49

Figure 6. Nitrogen sorption isotherms.

Table 4. Catalytic Formation of α-Monolaurin by Lauric Acid Addition on Glycidol

catalyst	% conv.[a]	tr = 24 h % yield	% sel.[b]
microspheres 1	66	10	15
microspheres 3	53	5	9

[a] Glycidol conversion. [b] Glycidol selectivity in α-monoglyceride formation.

Table 5. Catalytic Formation of α-Monolaurin by Lauric Acid Addition on Glycidol with Microspheres 1

run	reaction time	conv. (%)[a]	sel. (%)[b]
1st run	24 h	66	15
	50 h	82	65
2nd run	24 h	45	22
	72 h	85	70
3rd run	24 h	46	22
	72 h	94	96

[a] Glycidol conversion. [b] Glycidol selectivity in α-monoglyceride formation.

Table 4 shows the results of the catalytic activity of the hybrid microspheres. Microspheres **2** were not investigated for catalysis due to the very low accessibility to the amine groups. The reaction was carried out in toluene at 70 °C, a temperature compatible with thermal stability of the chitosan. All the experiments have been done for the ratio amine/substrate of 10 considering only the accessible amine groups.

The different activities of these materials suggest that the environment of the basic sites would be affected by the synthesis procedure.

The recycling of the materials was tested for the microspheres **1**, which presented the best catalytic performance. The results are given in Table 5.

The data reported in Table 5 show that the conversion of glycidol catalyzed by the hybrid microspheres is higher for the first run after 24 h than for the runs that followed. The selectivity in the monoglyceride formation increases with the conversion and also with the reuse order. This increase was previously observed with chitosan alone[13] and with amine-grafted silicas.[17] This phenomenon has been previously attributed to the slowing down of the competitive glycidol polymerization demonstrated after selective passivation of the residual silanol in the case of functionalized silicas. Indeed, the change in the selectivity versus conversion in the present work could result from progressive adsorption of the polymeric glycidol or monoglyceride onto the hydroxyl groups as the reaction goes along. This effect reduced the activity of these later being either the surface silanols of silica or the alcohol groups of the polysaccharide framework. This assumption is consistent with the lower selectivity and slightly higher activity of the catalyst observed at the beginning of the catalytic test or immediately after each recycling step that liberates these polar hydroxy groups by solvent washing.

contacted with the aldehyde, the hybrid microspheres became yellow, a color characteristic of the Schiff base and UV−Vis reflectance spectrum of the resulting materials after several washings with ethanol and drying exhibits the characteristic bands of the Schiff base. Quantitative GC analysis of the remaining salicylaldehyde in the solution revealed the ratio (%) of the amino groups of chitosan which have been involved in the Schiff base formation (Table 3). The results given in Table 3 correspond to a time contact of the salicylaldehyde with the microspheres of 23 h. The values are standardized versus the chitosan percent in the hybrid determined from the thermogravimetric measurement. The urea has been removed during the supercritical drying.

By comparison with the pure chitosan beads, the percent of accessible amine groups is higher than that in the hybrid microspheres **1** that features the most homogeneous composition. On the contrary, the amine groups of the microspheres **2** and **3**, which have a significant core/shell structure, are not or less accessible.

The potentiality of such a hybrid in catalysis is emphasized in the reaction of the fatty acid addition on glycidol, leading to monoglyceride formation (Scheme 2).

Such a catalytic reaction has been developed in the laboratory as a model reaction to evaluate the catalytic properties of amine-containing materials.[13,17]

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Conclusion

The work reported here is based on the use of natural polymers for the preparation of organic−inorganic hybrids. This would allow us to take advantage of three properties of biopolymers: (i) their tendency to self-assembly, (ii) their chemical functionality, and (iii) their natural asymmetry. With use of a source of silica, a new nanostructured hybrid material was formed, which could own both a mineral structure and surface properties of biopolymers and, on the other hand, both high porosity and controlled morphology. The potential of such materials in catalysis being demonstrated, other catalytic applications are in progress and uses of these hybrid solids for drug delivery or chromatography[18] are currently under investigation.

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