Control and formation mechanism of extended nanochannel geometry in colloidal mesoporous silica particles

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ABSTRACT: A large class of colloidal multi-micron mesoporous silica particles have well-defined cylindrical nanopores, nanochannels which self-assembled in the templated sol-gel process. These particles are of broad interest in photonics, for timed drug release, enzyme stabilization, separation and filtration technologies, catalysis, etc. Although the pore geometry and mechanism of pore formation of such particles has been widely investigated at the nanoscale, their pore geometry and its formation mechanism at a larger (extended) scale is still under debate. The extended geometry of nanochannels is paramount for all aforementioned applications because it defines accessibility of nanochannels, and subsequently, kinetics of interaction of the nanochannel content with the particle surrounding. Here we present both experimental and theoretical investigation of the extended geometry and its formation mechanism in colloidal multi-micron mesoporous silica particles. We demonstrate that disordered (and consequently, well-accessible) nanochannels in the initially formed colloidal particles gradually align and form extended self-sealed channels. This knowledge allows to control the percentage of disordered versus self-sealed nanochannels, which defines accessibility of nanochannels in such particles. We further show that the observed aligning the channels is in agreement with theory; it is thermodynamically favored as it decreases the Gibbs free energy of the particles. Besides the practical use of the obtained results, developing a fundamental understanding of the mechanisms of morphogenesis of complex geometry of nanopores will open doors to efficient and controllable synthesis that will, in turn, further fuel the practical utilization of these particles.

KEY WORDS: mesoporous silica; control and self-assembly mechanism; pore geometry;
One of the primary and most elusive goals of modern nanotechnology is to develop synthetic methodologies for the efficient and precise self-assembly of complex functional objects. Self-assembly is a preferential method of nano manufacturing because of its intrinsic robustness and relatively low cost. Layer by layer assembly of large layered structures\(^1\) is a prominent example of such approach. Self-assembly of colloidal three-dimensional silica shapes of almost biological morphological complexity which feature well-defined nanoporosity were previously reported \(^2-8\). It was shown that co-assembly of organic liquid crystals with inorganic precursor of some inorganic oxides, in particular, silica resulted in the formation of multi-micron-size particles of high morphological complexity\(^9,10\). The addition of cationic surfactants to these reactions result in the formation and mineralization of well-ordered arrays of silica with highly ordered cylindrical nanopores \(^11,12,13,14\) that can extend to many microns. Complex micron-sized pore geometry of these mesoporous particles are of interest in the areas of medical and sensor applications \(^15,16\), catalysis \(^17-19\), molecular storage \(^20-22\), separation and filtration technologies \(^23-25\), photonics, \(^26-28\), etc.

The non-trivial properties of these colloidal particles/materials are defined by a combination of the complex micron-scale morphologies and the highly ordered, cylindrical, nano-scale pores. The extended open pore geometry enables a special availability of the pore walls to the particles’ external environment. Specifically, extended pore channels may result in longer diffusion of encapsulated molecules out the particles. Furthermore, open pores allow free diffusion of external molecules into the pore, whereas closed pores do not. Therefore, it is important to understand the formation of the extended pore geometry and their alignment in these complex micron-sized mesoporous colloidal silica particles.

Mechanisms of self-assembly of micron mesoporous silica particles have been studied in the literature quite extensively \(^29-31\). In general, this self-assembly occurs when liquid crystal templates are formed in a solution of amphiphilic molecules when their concentration exceeds the critical micellar concentration. Mineralizable inorganic moieties (for example, silicic acid) condense on the interface of the liquid crystal micelles, which in turn facilitates the growth of the mesoporous phase \(^32\). The initial stage of micellar and liquid crystal formation and transition to mesophase material are also well studied \(^31\). The initial micelle-silica structures evolve into a nematic-like phase and the hexagonal structure of the mesoporous silica develops almost
immediately 33. Scattering methods show the presence of both small and large objects in the intermediate stage of the synthesis 7,8 which eventually evolve into fully developed mesoporous silica particles. Study of SBA-15 5-8, 31 and MCM materials 4 show the presence of small “flocs” or seeds. These 20-50 nm diameter seed particles typically have only a few nanochannels. It was hypothesized that the direct aggregation of the nanoparticles/seeds led to the formation of the final colloidal mesoporous shapes. Condensed silica continues “polymerizing” by decreasing the number of hydroxyl groups embedded in the silica matrix. This leads to two effects: stoichiometric shrinking of the silica and an increase in its elastic modulus 10,34,35. Balancing of the thermal and mechanical free energy drives the formation of the observed shapes. Specific shapes are the integrals of motion of the Euler-Lagrange equation. The shape distribution around these thermodynamic minima conform to the Boltzman distribution 36. Although the particle shape formation mechanisms and pore formation at the nanoscale have been extensively investigated, the pore geometry and its formation mechanism at a larger (extended) scale is virtually unstudied.

Here we report on the study of formation of extended pore geometry, alignment of cylindrical pores, and presence of disrupted open channels in the self-assembled colloidal multi micron-sized silica particles between the stage of formation of mesophase and fully developed mesoporous silica particles. Specifically, we investigate well-known MCM-type syntheses 10,34 which results in a variety of multimicron mesoporous particles rich in complex morphology (see, Fig. 1). It should be noted that the synthesis is rather standard; the only difference is in adding formamide, which is a pH stabilizer. It was added to slow down the synthetic process. Despite their complex micro-scale morphologies, the synthesized particles contain highly ordered, hexagonally packed cylindrical nanochannels that run through the particles. We see that the initial seed particles aggregate in large shapeless multi-micron particles, the process typical for colloidal growth. In contrast to previous works (see, e.g., 7,8), we demonstrate that the pores of the aggregated seed nanoparticles are not aligned right after aggregation. This is in agreement with previous indirect observations of SBA-15 mesoporous silica (synthesized using Pluronic acid template) 6,33.

We observe that nanochannels are not aligned and not self-sealed in these initial aggregates, that the channels do not run through the entire particle, and that these intermediate
particles contain many disrupted open channels that are not typically found in the end product of these syntheses. Because the self-sealed channels prevent interaction of the moieties encapsulated in the channels, the presented knowledge allows to control accessibility of the pore content of the described colloidal particles by changing the percentage of disordered versus self-sealed nanochannels. We further show that the observed aligning the channels is in agreement with theory; aligning the channels together is thermodynamically favorable. It decreases the Gibbs free energy of these precursor particles, which drives self-assembly of these particles. Indeed, we observe that such alignment and resulting self-sealing of the channels occur later in the synthetic process and is exhibited in the fully developed particle morphologies exampled in Fig.1. in addition to the ability to control the pore accessibility of the particles, developing a fundamental understanding of the mechanisms of morphogenesis of complex geometry of nanopores will open doors to efficient and controllable synthesis that will, in turn, fuel the practical utilization of these particles. Possible applications of the obtained results are discussed.

RESULTS AND DISCUSSION

Microscopy study
Templated sol-gel self-assembly of particles\textsuperscript{10, 34, 37} brings a large variety of nanoporous silica morphologies. These shapes can range from single microns in size to hundreds of microns and with various aspect ratios, Fig.1. As was shown, these particles have cylindrical pores of \(-\)4nm in diameter packed in the nematic hexagonal order.
Fig. 1. Variety of nanoporous silica morphologies synthesized using a templated sol gel self-assembly approach. 90x90 µm² SEM image is shown. Colors are artificial to highlight different micro-morphologies of particles.

To probe the dynamics of the self-assembly of these shapes at different times of the synthetic process, we periodically extracted and analyzed aliquots of the synthesizing bath which contains both synthesizing chemicals and solid fractions of the mesophase intermediate product. The collected solid fractions were imaged using transmission electron microscopy (TEM), atomic force microscopy (AFM) and scanning electron microscopy (SEM). The first sample collection was done at 0.5 hours after completing the mixing of all chemicals. At this stage, the amount of initially assembled seeds starts to exceed the concentration needed for formation of larger particles. This stage is usually clearly seen by its milky appearance of usually transparent solution. These seed particles were imaged with AFM (the sample preparation is described in the Method section). The particles formed at 12, 16, and 24 hours were too large to be imaged with AFM. Thus, SEM imaging was used.
Fig. 2. Evolution of the morphology of mesophase during self-assembly of silica particles versus synthesis time (the time after completing the mixing of the chemicals used in the synthesis). (a) AFM image at 0.5 h since the synthesis start; SEM images for 12 (b), 16 (c), and 24 h (d) since the synthesis start.
Fig. 3. HRTEM images of the edges of nanoparticles of the particles collected at different synthesis time: (a),(b) – 0.5h, (c),(d) – 12 h, (e),(f) – 16 h and (g), (h) – 24 h showing evolution of pore geometry. Corresponding FT pattern and enlargements are given as insert. White lines indicate the direction of nanochannels/mesopores.
Figure 2 shows representative images of the particle’s morphology obtained with the help of AFM and SEM. (Note that the absence of large particles at 0.5h was verified with the DLS technique.) One can see the evolution of size, morphology and microstructure of silica particles depending on the synthesis time. The synthesis starts from a fast formation (~0.5h) of the initial “seed” particles of the size of approximately 30 nm (see the DLS measurements, Fig.S1 of the Supplementary materials) and are typically spherical in shape (Fig.2 a,b). Over about 12 hours of the synthesis the seed particles fuse into large aggregates of irregular multi-micron shapes. Samples collected after 12 hours demonstrate the presence of large silica particles with particle sizes close to those observed at the synthesis completion (after 24 hours). There is a marked decrease in the degree of irregularity and in the number of “cracks” on the particle surfaces at 16-hours (Fig. 2c). After 24 hours the formation of well-defined, particles having a mixture of complex morphologies are clearly visible in the SEM images (Fig 2d).

Figure 3 shows the results of high resolution TEM (HRTEM) study. As reported in previous work, and as shown in the TEM images in Figure 3, the mesophase is present at all time intervals. TEM imaging of the edges of the particles assembled by hour 12 show multiple directions of the nanoporous channels within the same particles (highlighted with white lines in Fig. 3c,d). The samples collected at 16 hours demonstrate almost fully developed unidirectional mesoporous channels for the majority of the particle surface. For example, the TEM image of Fig.3e clearly shows the hexagonal packing arrangement (FT pattern insert also shows strong and sharp spots corresponding to the hexagonal symmetry). However, Figure 3f shows that a good portion of the particle surface is not aligned with the unidirectional bulk. This is seen in both SEM and TEM images. Finally, the images of the particles taken after 24 hours (Figures 2d and 3g,h) show a rather smooth surface having predominantly uniform parallel pores.

**Study of presence of disrupted open channels**

To further support our self-assembly hypothesis—that seed particles first aggregate, then re-arrange so that the pores align in order to minimize their free energy—we employed confocal microscopy with a fluorophore to probe the degree of openness of the pores at the different stages of synthesis. Perfect extended alignment of cylindrical nanopores should result in self-
sealing of such nanoporous in the round geometry. Misalignment of cylindrical pores in the initial aggregates of seed particles can leave disrupted open channels in the silica structures. This can be verified using small fluorescent molecules by observing diffusion of these molecules inside the synthesized particles. The dye cannot diffuse inside perfectly aligned self-sealed channels (or it takes long time to diffuse along an extended nanochannel). This would leave fluorescently dark regions inside the particles. And oppositely, the dye can diffuse inside disrupted channels open to the dye molecules. These would create bright fluorescent regions inside of the particles. Thus, the presence of perfectly aligned or extended and disrupted channels can be visualized by using fluorescent microscopy of these particles. Here we use rhodamine 6G (\(\sim 1.1 \times 1.6\)nm) molecule to verify this hypothesis.

The particle samples for this study were extracted as previously at 10, 12, 16, and 24 hours after completing the mixing of all chemicals. The samples were now not washed as in the previous preparation but used as is. A droplet of aliquots was deposited on a glass slide, and a small amount of aqueous solution of rhodamine 6G was added. The imaging started in about five minutes after adding the dye. To demonstrate penetration of the fluorescent dye molecules inside silica particles, confocal scanning laser microscopy was utilized.

Figure 4 demonstrate diffusion of rhodamine 6G fluorescent dye inside silica particles assembled at different times. The confocal images shown in figure 4, were collected within 20 minutes after adding the dye. It should be noted that the fluorescent images become rather stable after that time. One can see that the dye penetrates quickly inside of all particles assembled by 10 hours of the synthesis, Fig. 4a. It is interesting to note some dark regions inside of particular large aggregates. These are presumably already developed regions of aligned self-sealed nanochannels. The particles assembled by 12 hours demonstrate still a substantial amount of disrupted open channels, which allow fluorescent dye to diffuse, Fig. 4b. The particles assembled by 16 hours show already noticeable resistance for the dye to penetrate in quickly, Fig. 4c. One can see that the dye penetrates inside the particles but not deep enough. This indicates the presence of perfectly aligned self-sealed cylindrical nanochannels inside the particles. Figure 4d show rather low dye penetration in the fully developed silica particles. The dye was able to penetrate only a thin surface layer of these particles.
It is worth noting that, in principle, the decrease of fluorescence can come from creation of H-dimers of the dye molecules inside the silica pores. However, as we recently demonstrated \(^{41}\), such dimers are not formed in noticeable quantities inside of mesoporous silica particles. Therefore, the possibility of the appearance of the dark spots due to dimerization of the fluorescent dye should be excluded.

Fig. 4. Diffusion of R6G fluorescent dye inside nanoporous silica particles extracted at different times of self-assembly. Confocal optical images are shown at (a) 10h, (b) 12h, (c) 15-17h, (d) 24h since the synthesis beginning.

Another concern to address is related to the imaging of transparent silica particles with optical microscopy. Multi-micron size particles can work as good optical lenses showing a dark spot artifact in the middle. To confirm that we are not dealing with this type of artifacts, images
of the particles presented in figure 4 were collected at various focal depths. Examples of such imaging are presented in the Supplementary materials, Fig.S2-S4.

One can see that the results of the dye diffusion shown in Fig.4 are in agreement with the TEM images (Fig. 3). Based on these observations, we can suggest a schematic of the nanoporous channels inside these particles. It is presented in Figure 5. The initial formation of mesophase, hexagonally packed cylindrical pores, is already seen in the seed particles (0.5h). The irregular shape multi-micron aggregates/intermediate particles, which were observed at 12 hours of the assembly, have the cylindrical pores and oriented in multiple directions (though some of the cylinders may become already fully aligned and fused together forming spherical regions of self-sealed pores). The amount of disrupted open pores is very high. By 16 hours of the synthesis, the core of the particles has mostly an aligned self-sealed pore structure. The amount of disrupted open channels is substantially decreased. The latter are mostly localized near the particle surface. By 24 hours of the assembly, the particles reach their fully developed morphology with virtually all cylindrical pores of aligned / self-sealed. The amount of disrupted open channels reached its minimum and localized and the particle’s surface.

![Fig.5. Schematics of the internal orientation of cylindrical pores inside the initial seed nanoparticles (formed at 0.5 hours) and self-assembled mesoporous silica micron size particles (formed by 12, 16, and 24 hours). Cylindrical channels are schematically indicated by lines. One can see the development of fully aligned self-sealed cylindrical pores with the increase of the assembly time. The number of disrupted open channels decreases with increase of assembly time as well.](image-url)
**Theoretical reasoning for the nanochannel alignment**

The observed behavior of cylindrical nanochannels makes sense within the model suggested in previous work \(^\text{10, 34, 35, 42}\). This behavior becomes more apparent once the free energy of the particle at its various stages of self-assembly are calculated. The Gibbs free energy \(F_d\) of silica particles having hexagonal nematic symmetry of the channels and overall cylindrical symmetry can be described as follows \(^\text{35}\)

\[
F_d = C \left[ \int_0^{2\pi} d\varphi \left( 1 - \alpha \cos 2\varphi \right) (1 + \psi'^2) + \Delta \int_0^{2\pi} d\varphi \left| \nabla_\varphi r \right|^2 \right].
\]  

(1)

Here the notations are shown in figure 6a and given by the following formulas:

\[
\alpha = \frac{K_b - K_s}{K_b + K_s}, \quad \psi' = \frac{\partial \psi}{\partial \phi}, \quad \left| \nabla_\varphi r \right| = \frac{1}{R} \frac{\partial R}{\partial \phi}, \quad \Delta = \sigma \frac{2}{K_b + K_s} \frac{R}{\ln R / R_0},
\]

(2)

where \(K_b\) (\(K_s\)) are the bend (splay) elastic modulus, \(R\) is the particle radius, \(R_0\) is the smallest radius of the discoid, i.e., the diameter of the pore, \(C\) is a geometrical constant independent on the internal pore geometry, \(\sigma\) is the surface tension of silica particle during its formation (\(\Delta\) is a dimensionless parameter given the relative contribution of the surface tension; as was estimated, \(\Delta \sim 100\) during the assembly of the silica shapes \(^{35}\)).

Fig.6. (a) Geometrical notations used to define free energy of mesoporous silica particles which have hexagonal nematic symmetry of nanochannels. Here \(\vec{n}\) is a unit vector of the director-field of the nematic phase, \(r\) and \(\phi\) is the polar coordinates, the angle \(\psi\) of between \(\vec{n}\) and the radius vector \(\vec{r}\). (b) The schematics of particles having misaligned channels facing each other at some angle.
Although the free energy described by equation (1) is given for the particles having overall cylindrical symmetry, it is also valid if even a small part of the particle has such symmetry; in this case the integral with respect to $\varphi$ in equation 1 should be taken between angles $\varphi$ describing the beginning and the end of this particular particle. The particles which have cylindrical pores misaligned (running towards each other at some angle) and aligned (the angle between these two cylindrical domains is equal to zero) are shown in figure 6b. The case of $\psi_0=\pi/2$ response to the aligned (fused channels). The energy penalty for having misaligned channels can now be found.

While it may appear obvious that bending or kinks in the channels results in an increase of free energy, this is not necessarily correct, since excessive bending energy can be shared with splay, and it can be redistributed through various geometric relaxations. One of the most well-known geometric relaxations of excessive mechanical energy is buckling \(^{43}\). Finally, the actual geometry of the particle is also defined by the presence of nonzero thermal energy. As a result, the distribution of free energies of particles is described by the Boltzmann distribution in the equilibrium thermodynamics limit \(^{36}\). Finally, the actual geometry of the particle is defined by the presence of nonzero thermal energy, which results in the Boltzmann distribution of free energies amongst the particles which channels are facing each other at some angle $\psi_0$ as shown in figure 6b.

The minima of energy are given by the Euler–Lagrange equation \(^{44}\), which is written as follows for the functional of eq.(1):

\[
(1 - \alpha \cos 2\psi)\psi'' = \alpha \sin 2\psi \ (1 - \psi'^2) - \Delta \frac{\cos \psi}{\sin^3 \psi} .
\]  

\(3\)

The solutions of this equation (the integrals of motion which give the local energy minima) are presented by the following equation:

\[
(1 - \alpha \cos 2\psi) \ (\psi'^2 - 1) - \frac{\Delta}{\tan^2 \psi} = \text{const}(\varphi) \equiv Q .
\]  

\(4\)

The constant $Q$ is determined from the following boundary condition by the equation
\[
\frac{2\pi}{3} = \int_{\psi_0}^{\pi-\psi_0} d\psi \sqrt{\frac{1-\alpha \cos 2\psi}{1+Q-\alpha \cos 2\psi + \Delta / \tan^2 \psi}}.
\]  

(5)

Note that \(\psi(r, \varphi) = \psi_0 = \pi/2\) is also a solution of equation 3 (in this case \(Q = -(1+\alpha)\)). The solutions of this equation define the geometry of the particles and the alignment of their channels, which correspond to the local minima of free energy. Finding numerically these solutions of equation (4) with the definition given by equation (5), one can calculate the integral of free energy given by substituting the solutions in equation (1). To find the relative change of the free energy of misaligned versus aligned channels, we only need to know the surface tension parameter \(\Delta\) and relative modulus \(\alpha\). Both parameters were estimated previously \(^{35, 36}\). Self-assembling mesoporous silica micron particles have \(\Delta \sim 100\) and \(K_s < K_b\). Because of the uncertainty in \(K_s\) and \(K_b\), we considered several possible values of \(\alpha\). Table 1 shows the results of numerical calculations of the ratios of free energies of misaligned and aligned channels for two examples of the values of the angle of misalignment \(\psi_0 = \pi/4\), and \(\psi_0 = \pi/40\). One can see that the values are noticeably larger than 1, which means that the free energy of misaligned channels is higher than the energy of the aligned channels.

<table>
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<tr>
<th>(\alpha)</th>
<th>(\psi_0 = \pi/4)</th>
<th>(\psi_0 = \pi/40)</th>
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<td>0.1</td>
<td>4.1</td>
<td>22</td>
</tr>
<tr>
<td>0.5</td>
<td>3.4</td>
<td>14</td>
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<tr>
<td>1.0</td>
<td>2.9</td>
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Table 1. Ratio of the free energies of misaligned to aligned channels.

**CONCLUSION**

We studied formation of extended (from submicron to micron-scale) geometry of nanoscale channels inside colloidal multi-micron mesoporous silica particles. We demonstrated that these large particles are forming from the initial seed nanoparticles that have developed nanochannels, which is in agreement with the existing literature. However, we found that in the early stages of
self-assembly, multi-micron mesoporous particles form particles of irregular shapes which had virtually no extended aligned nanochannels. Our work indicates that these extended channels start aligning after the seed particles form large aggregates. These large aggregates initially have a lot of open disrupted channels which re-arrange and align over time. Further, we demonstrated that the extended alignment of these channels occurs because it is energetically favorable and results in a minimization of free energy within the particles. In agreement with the theoretical prediction, we observed that the number of open and misaligned channels decreases with the increase of the synthesis times. By reaction completion, the extended full alignment of cylindrical self-sealed nanochannels running across the entire particles is created. These results shed light on the mechanisms of self-assembly of complex geometries of nanopores. Additionally, these results will aid in development of new materials for various applications e.g. timed drug release, filtration, separation, enzyme stabilization, etc.

**EXPERIMENTAL SECTION**

**Materials:**

Tetraethyl orthosilicate (TEOS, 98%, Aldrich), cetyltrimethylammonium chloride (CTAC, 25% aqueous, Aldrich), hydrochloric acid (HCl, 36% aqueous, J T Baker), Formamide (HCONH$_2$, 98%, Aldrich) and Rhodamine 6G (R6G, Exciton Inc.), ultrapure water (18 MΩ-cm, MilliQ Ultrapure) were used without further purification.

**Synthesis:**

The initial synthesis procedure was adapted from the previously reported works $^{10,34}$. The following molar ratio of the reactants 100H$_2$O:8HCl:0.11CTACl:0.13TEOS: 9.5HCONH$_2$ was used. Fluorescent dye (R6G) can be added as a contrast agent for fluorescent microscopy (~1x10$^{-4}$ relative molar concentration was used) At first, water, formamide and dye were mixed in a plastic bottle, and stirred using a magnetic stirrer (Hotplate stirrer, Lab Depot, Inc.) at 550 RPM for 5 min at room temperature. In the next step, hydrochloric acid was added to the reaction mixture. As a result, the temperature of the reaction mixture rose to 40°C. The reaction mixture was then put in an ice bath and stirred until it reached room temperature. Further, CTAC
was added to the reaction mixture and stirred for 45 min. As a last step, TEOS was added and stirred for 2 hours. The reaction mixture was then kept in quiescent condition (without stirring) up to 24 hours. The time of reference for sample extraction is the time when TEOS was mixed with rest of the reaction mixture. Particles were then extracted after 0.5 h, 10 h, 12 h, 16 h, and 24 h.

Characterization:

The synthesized particles were characterized with confocal scanning laser microscopy (CSLM) (Eclipse C1, Nikon Inc.). The particles were extracted from the synthesizing bath and imaged after adding aqueous solution of rhodamine 6G fluorescent dye. For characterization with other types of microscopy, such as scanning electron microscopy imaging (SEM, by Phenom, FEI), transmission electron microscopy (TEM, Tecnai G2 30 UT), and atomic force microscopy (Bruker Icon), the particles were prepared as follows. For the study of the initial seed particles, a droplet of the synthesizing bath solution was deposited on a layer of graphite for AFM measurements, and gently washed with water after 2 minutes of waiting. Then the sample was dried and imaged with AFM. The procedure of collection of solid phase was repeated at 12, 16, and 24 hours after completing the mixing of all chemicals. For SEM imaging, samples extracted at those particular times were first dried in an oven (Isotemp 500 series, 60° C) for 24 hours and then placed on a double sided carbon tape attached to the SEM stub, followed by a light gold coating (Hummer 6.2 spattering system) for 1 minute. TEM study was done on crushed calcined samples prepared by using ultrasonic bath, dissolving in ethanol and depositing on holey carbon grid. TEM was operated at 300 kV with 0.17 nm point resolution and equipped with an EDAX EDX detector. A low intensity electron beam and medium magnification were used in order to avoid electron beam damage of the structure inside the microscope.

ASSOCIATED CONTENT

Supporting Information
The Supporting Information is available free of charge on the Dynamic light scattering to define the size of the initial seed particles; Diffusion of fluorescent dye inside particles: verification of the absence of lensing artifacts. (PDF)

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Notes

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