A hierarchical self-similar structure of oriented calcite with association of an agar gel matrix: inheritance of crystal habit from nanoscale†

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Calcite crystals grown with association of an organic gel matrix led to the generation of a hierarchical, self-similar architecture possessing rhombohedral habit under ambient conditions.

Self-organization, a typical bottom-up approach, is a promising strategy to integrate nanomaterials under ambient conditions without using special equipment.[1–6] We found that carbonate biominerals can be made by the oriented architectures of nanocrystals less than 100 nm in size with incorporation of biological macromolecules.[6,7] The facts indicate that living organisms construct hierarchically organized structures from nanoscopic to macroscopic scales by means of nanocrystals as building blocks. Various ideas have been demonstrated for the development of biomimetic pathways to make up the hierarchical structures under ambient conditions. Therefore, complex morphologies and hierarchical structures emerged from nanocrystals with association of organic molecules in synthetic systems. On the other hand, morphological control combined with hierarchy and self-similarity was achieved in a limited number of systems.[8–14] Self-similarity is found on a broad range in nature and understanding of the origin is a significant challenge. An exquisite control of growth and inhibition in tailored spatiotemporal scales is required for the emergence of hierarchical self-similarity through crystal growth. In the previous studies, the construction of hierarchical structures was mainly associated with a specific adsorption of organic molecules. Herein, we show another pathway for the fabrication of hierarchical self-similar architectures using a gel matrix.

Nucleation and morphology of crystals has been controlled in a hydrogel medium.[15–21] It is well known that the reduction of nucleation rate and the suppression of convection enable the growth of defect-free single crystals.[15–17] In the other cases, diffusion-controlled growth led to the emergence of complex morphologies and periodic precipitation under nonequilibrium conditions.[18–21] Mineralization of calcium carbonate ($CaCO_3$) was performed in various types of organic gel matrices consisting of biological and synthetic polymers.[22–30] Ellipsoidal shapes of calcite and porous hexagonal columns of vaterite were grown in gelatin matrices under controlled growth conditions.[22,23] Star-shaped calcite particles were obtained in agarose gels.[24] In a silica gel, specific adsorption of silicate anions resulted in morphological evolution including sheet and helical shapes.[25,26] Synthetic gels having carboxy and sulfate groups and acrylamide moieties led to habit modification of calcite and miniaturization of the crystallites.[27,28] The combination of a soft gel matrix on a substrate and soluble polymeric species directed the formation of characteristic thin films.[29,30] While many researchers demonstrated morphological control of carbonates by means of polymers and molecules, the mineralization in organic matrices has not been fully studied in recent years.

Here we found that hierarchical self-similar structures of calcite formed in an organic gel matrix under ambient conditions. An agar matrix contributed to the morphogenesis at different levels from nanoscopic to macroscopic scales. The isotropic growth of calcite resistant to the gel matrix, determined the macroscopic shapes, and the nanocrystal engineering, as observed in biominerals, induced the microscopic structures. Therefore, we successfully prepared a hierarchical self-similar architecture of calcite possessing a rhombohedral habit in three different levels. Along with habit modification by polyelectrolytes, the results suggest a novel pathway for crystal growth leading to both hierarchy and self-similarity at ambient conditions.

The precipitation of $CaCO_3$ crystals took place in agar hydrogel containing 0.3 M of calcium chloride ($CaCl_2$) aqueous solution with introduction of CO_2 at ambient conditions. The agar concentration (C_{ag}) was varied in a range between 0.1 and 8.0 wt%. After 3 days, the agar matrix was removed by washing with hot water and then the calcite precipitates were dried at room temperature. The morphologies were observed by field-emission scanning electron microscopy (FESEM) and transmission electron microscopy (FETEM) with selected area electron diffraction (SAED). The crystal structure was characterized using X-ray diffraction (XRD). The remaining agar molecules were studied from thermogravimetry–differential thermal analysis (TG–DTA). Detailed procedures are provided as ESI.†

Fig. 1(a)–(g) show the overview of the hierarchical self-similar calcite in three levels as denoted by tiers 1–3. The formation of calcite was confirmed by XRD analysis at C_{ag} = 0.1–8.0 wt%, while a small amount of vaterite was contained in the precipitates without addition of agar molecules. When the initial C_{ag} was set within a range of 0 to 0.2 wt%, a smooth surface of rhombohedral calcite was observed at any magnification in FESEM images (Fig. S1 in ESI†). A hierarchical self-similar architecture at three levels was generated at C_{ag} = 2.0 wt% (Fig. 1(a)–(c)). The macroscopic rhombohedra, *ca.* 20 μm in size, consisted of interconnected units in submicrometer scale (tiers 1 and 2 in Fig. 1(a), (b)). The self-similarity of the rhombohedral habits in tiers 1 and 2 was recognized from the highly ordered arrangement of the submicrometric units (Fig. 1(d)). Furthermore, the units were composed of

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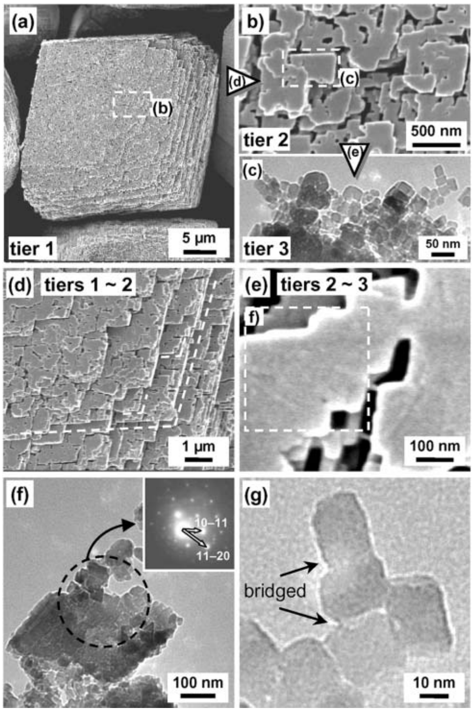

Fig. 1 Typical FESEM (a, b, d and e) and FETEM (c, f and g) images of the hierarchical self-similar architecture formed at C_{ag} = 2.0 wt%. (a) the macroscopic morphology in tier 1, (b) the submicrometric units in tier 2, (c) the nanocrystals in tier 3, (d) the rhombohedral habit inherited in tiers 1 and 2, (e) the submicrometric unit consisting of nanocrystals, (f) the SAED pattern (inset) suggesting the formation of the oriented architecture in tiers 2 and 3, (g) the nanocrystals connected by bridges.

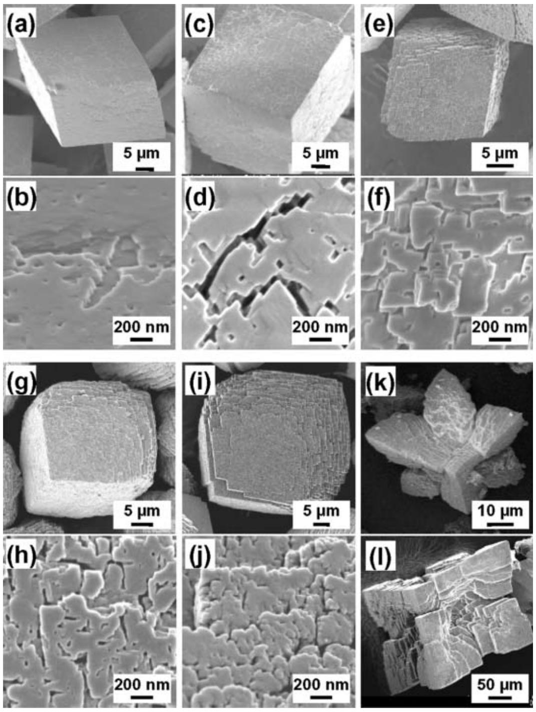

Fig. 2 FESEM images of the macroscopic morphological variation with a change of the initial agar concentration (a–j) and comparative experiments (k and l): (a and b) C_{ag} = 0.4, (c and d) C_{ag} = 0.6, (e and f) C_{ag} = 1.0, (g and h) C_{ag} = 3.0, (i and j) C_{ag} = 5.0, (k) skeletal shape of calcite obtained in agar gel matrix with introduction of carbonate ions from Na_2CO_3 aqueous solution, (l) the similar skeletal morphology formed in a pectin matrix.

smaller subunits approximately 20 nm in size (tiers 2 and 3 in Fig. 1(b), (c)). The magnified FESEM image also suggests the ordered alignment of nanocrystals in the submicrometric units and the spotted SAED pattern supports the formation of an oriented architecture (Fig. 1(e), (f)). As observed in biominerals,[7b] it seems that the each nanocrystal is connected by several bridges (Fig. 1(g)). However, a magnified image could not be acquired because of irradiation damage. The macroscopic shape, submicrometric units, and nanocrystals, each exhibiting rhombohedral habits were hierarchically organized in three different scales from nanoscopic to macroscopic. Therefore, the resultant calcite can be regarded as a hierarchical self-similar architecture.

In order to study the formation process, the morphological variation with a change of initial C_{ag} values is displayed in Fig. 2. The morphology at the submicrometer scale became porous with increasing C_{ag} whereas the size and rhombohedral habit in macroscopic shapes were not so changed (Fig. 2(a)–(j)). The results suggest that the morphogenesis was associated with the agar gel matrix. However, in accordance with the following experiments, agar molecules did not direct habit modification with strong adsorption on the crystal face of calcite. When the carbonate ions were rapidly supplied from a 0.3 M Na_2CO_3 aqueous solution loaded upon agar gel, skeletal shapes were obtained (Fig. 2(k)). Similar skeletal morphologies were also fabricated in a pectin gel that is not solidified (Fig. 2(l)). In addition, similar star-shaped calcite was synthesized in earlier work.[24] Our previous studies suggested that the morphological variation from polyhedra to skeletal shapes in the gel matrix resulted from diffusion-controlled growth.[17] Consequently, agar molecules did not induce habit modification with a specific interaction but served as a growth matrix. In addition, moderate reaction and growth conditions would be achieved by introduction of the carbonate source from the gas phase into the densified gel matrix. Thus, the skeletal shapes originating from diffusion-controlled growth are not formed in this study. It is inferred that the network structure of the gel matrix and the moderate reaction conditions lead to the hierarchical growth of calcite. The presence of agar gel networks leads to resistance for isotropic crystal growth of calcite and leads to the submicrometric units with rhombohedral habits. More segmentalized units were actually observed with an increase in the initial agar concentration (Fig. 2(a)–(j)). The segmentalization of calcitic units may be similar to the branching at the interface of two fluids with different viscosities, so called viscous fingering.[31,32] In consequence, the hierarchy and self-similarity in tiers 1 and 2 emerged from the isotropic growth of calcite resistant to the agar gel network under moderate reaction conditions.

In tiers 2 and 3, the oriented architecture of nanocrystals formed with incorporation of agar molecules. Hydroxy groups in the agar molecules would interact with the surface of calcite crystals although strong electrostatic interaction is not expected between calcite surfaces and agar molecules because of the molecular structure of the polysaccharide. The interaction of hydroxy groups with $CaCO_3$ crystals has been demonstrated in previous reports.[33,34] While the carboxy groups selectively adsorbed on the specific crystal faces leading to habit modification, the weak interaction of agar molecules would allow the isotropic growth of

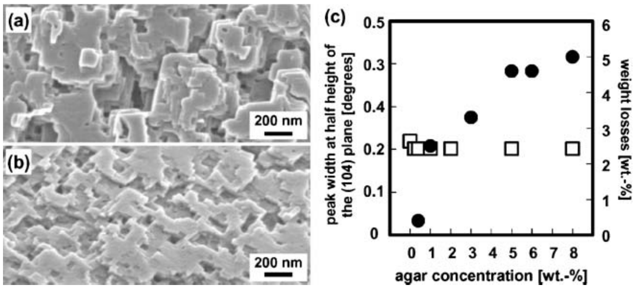

Fig. 3 The oriented structure of nanocrystals with incorporation of agar molecules: (a and b) FESEM images before and after NaClO treatment for the removal of agar molecules, respectively, (c) weight losses in TG–DTA analysis (right vertical axis with filled circles) and peak with at half height of (104) planes in XRD patterns (left vertical axis with open squares) with a change of the initial agar concentration.

calcite in this study. Fig. 3(a), (b) show the FESEM images of the surfaces before and after sodium hypochlorate (NaClO) treatment. The rhombohedral facets clearly appeared after the treatment because the remaining organic molecules were removed. The presence of incorporated agar molecules at the nanoscopic scale was also reflected to the weight loss in TG–DTA analysis (Fig. 3(c)): the weight losses increased in response to the initial C_{ag} values. Peak broadening was not observed in the XRD pattern at any C_{ag} values, suggesting the formation of oriented architectures (Fig. 3(c)). In this way, the nanocrystals induced the oriented architecture with incorporation of agar molecules in tiers 2 and 3. Since the adjacent nanocrystals seemed to be partially connected with each other (Fig. 1(g)), the formation process would be related to that of biominerals and biomimetic architectures as studied in our previous reports.[6,7,12] Similar nanocrystal engineering has attracted much interest in recent years, such as oriented attachment and mesocrystals. On the basis of these concepts, the colloidal aggregates of the primary nanoparticles mediated the oriented assembly and their fusion resulted in the single-crystalline structures.[3–5] The ions as sources of $CaCO_3$ can be transported in the hydrogel matrix. In contrast, the transfer and assembly of the pre-formed nanoparticles would be difficult in a gel medium. It is inferred that the stepwise growth including inhibition and restarting would take place in an agar gel medium. Löbmann *et al.* reported crystallization of calcite in polyacrylamide gel as a noninteractive matrix, but the rhombohedral shape was not retained in their system.[28a] The morphological change would be ascribed to habit modification with a specific interaction, even though the interaction may be weak. In contrast, it is inferred that the nonspecific and weak interaction of agar gel matrix to calcite contributed to the inheritance of rhombohedral habit in the present case.

In summary, the hierarchical self-similar architecture of calcite was generated in an agar gel matrix under ambient conditions. The macroscopic morphogenesis from tiers 1 to 2 was induced by the isotropic growth of rhombohedral calcite resistant to agar gel matrix. The oriented architecture of nanocrystals, in tiers 2 and 3, was generated from stepwise crystal growth with incorporation of agar molecules, as demonstrated in biominerals and biomimetic architectures. Agar molecules acted in dual roles in different scales from nanoscopic to macroscopic. Although porous and/or hierarchical structures of $CaCO_3$ and the crystal growth in a gel medium were reported in earlier work, the strong interaction of organic molecules mainly contributed to the morphogenesis and, to our best knowledge, rhombohedral calcite combined hierarchy and self-similarity was not presented. Moreover, crystal growth was controlled in multiple spatiotemporal scales by using a hydrogel medium in this study. Reproduction of the morphogenesis in other materials is a current challenge for the further development of this approach. Tailoring the combination of crystals and organic matrices and the growth conditions would facilitate hierarchical, self-similar superstructures at ambient conditions.

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