A facile procedure to direct the formation of hydroxyapatite (HAP) in preference to all other calcium phosphate phases and control over nanoparticle size distribution and morphology, with potential application in biomedical implants, is reported. The synthesis is performed in the presence of amine terminated dimer (DAB). Aqueous solutions of calcium, phosphate, and DAB were used at different calcium:DAB molar ratios, viz. 2:1, 1:1, and 1:2 and the resulting suspensions were either kept at room temperature or hydrothermally treated at 80 °C for 16 h, or at 130 °C for 6 h. The resulting nanomaterials were fully characterized in terms of their physicochemical properties by means of XRD, TEM, SEM, and FTIR. It was found that DAB affects nucleation and crystal growth, as, in all cases, nanorods of hexagonal HAP with homogeneous and narrow size distributions (with mean sizes ranging from 10 nm × 5 nm up to 81 nm × 22 nm depending on synthesis conditions) were obtained.

1. Introduction

Hydroxyapatite (HAP), Ca_{10}(PO_4)_{6}(OH)_2, has been extensively used for biomaterials research because of its structural similarities to the inorganic component of bone and teeth as well as its osteoconductive properties and excellent biocompatibility. HAP can be synthesized by a variety of methods such as solid-state reactions, plasma techniques, crystal growth under hydrothermal conditions, layer hydrolysis of other calcium phosphate salts, and sol–gel crystallization. These methods, however, mostly yield HAP crystals of irregular size and/or morphology, which could lead to uncontrolled biological behavior.

Bone is a hierarchical composite consisting of HAP nanoparticles, collagen, and proteins acting as control macromolecules. This structure allows a synergistic function during the mineralization of bone with collagen acting as the framework macromolecule and noncollagenous proteins being involved in the development and fine-tuning, or regeneration, of the skeletal structure. Very recently, it has been recognized that implantable biomaterials for bone grafting should possess characteristics mimicking those of natural bone in order to be successful in their application. Consequently, a lot of research efforts are now being directed toward biomimetic approaches for the synthesis of HAP crystals of various morphologies, using polymers such as monosaccharides, polymers, or more recently dendrimers and block copolymers to modulate crystal nucleation and growth. Further, reverse micelle-mediated synthesis of HAP nanocrystals with controlled morphology using surfactants as templates has been also investigated as this method can yield agglomerate free or soft agglomerated nanopowders.

Dendrimers, compared with linear polymers, present the advantage of a highly precise architecture, as they are highly branched macromolecules composed of a core molecule, a large number of branches regularly extending from the core, and terminal (or surface) groups suitable for further functionalization. Thus, dendrimers have a definite molecular weight and size, in contrast to the usually broad molecular weight distribution of linear polymers, a near spherical shape for generation numbers larger than 4, and are able to encapsulate metal ions, or organic molecules, e.g., drugs. More importantly, the above properties and the presence of multiple functional groups on their surface, render them ideal for the study and modulation of many biological processes. Concerning their potential to mimic crystal growth related to biological systems (such as biomineralization, calcification, silicification), this is also affected by their functional terminal groups as they determine their binding capacity to the crystal surface.

Thus, up to now, poly(propylene imine) dendrimers functionalized with long alkyl chains have been used for the synthesis of dendrimer–HAP composites at room temperature affording micrometer-sized clusters of small crystalinites in the form of thin platelets while carboxylic acid terminated poly(amidoamine) (PAMAM) dendrimers were used as templates for the preparation of amorphous calcium phosphates. On the other hand, the combination of the biomimetic process and the hydrothermal method in the presence of PAMAM dendrimers with surface carboxylate, hydroxyl, or amido groups resulted in HAP nanorods which, however, were not further tested in in vitro experiments. Recently, linear or branched poly(ethyleneimine) (PEI) was also investigated for HAP mineralization to demonstrate the templating efficiency of cationic polymers for HAP nucleation.

It is known that the use of biomolecules containing specific amino acid residues, such as arginine, lysine, or specific peptide sequences, e.g., the RGD sequence, encourages cell attachment,

---

S. Bose—contributing editor
proliferation, and differentiation on HA surfaces.36 In this respect, in the current study, we used a cationic fourth generation

diaminobutane poly(propylene imine) dendrimer bearing 32 amine end groups for the synthesis and characterization of

HAP nanorods. Here, the effect of dendrimer concentration and hydrothermal treatment on HAP crystallinity and nano-
particle morphology was investigated with a number of tech-
niques including XRD, TEM, and FTIR. This method proved a

compact way to synthesize HAP nanorods with tailored properties.

II. Experimental Procedure

(1) Nanorod Synthesis

Aqueous solutions of CaCl2 (0.1 M; Sigma-Aldrich, ACS grade, St. Louis, MO), Na2HPO4 (0.06 M; >99%; Fluka, Buchs, Switzerland), and 1,4-diaminobutane poly(propylene imine) dotriacontaamine dendrimer, [-CH2CH2N[(CH2)3N[(CH2)3CH2N(CH2)3CH2N(NH2)][CH2]3][CH2]3][CH2]3], DAB (0.2 M with respect to primary amino groups; DSM Fine Cheicals, Linz, Austria) were prepared using Millipore water, Ca2+ and dendrimer so-
lutions were combined at different Ca2+:dendrimer molar ratios (2:1, HAPa; 1:1, HAPb; 1:2 HAPc). The resulting solutions had a pH of 10.6, were cooled at 0°C and PO43- solution was slowly added under continuous stirring to a final 10:6 Ca:P molar ratio. The resulting suspensions were allowed to reach room temperature and either used without any further treatment after remaining at room temperature for 1 day or subjected to hydrothermal treatment at 80°C for 16 h, or at 130°C for 6 h (Table I) in an autoclave (Steroclave TKA, Milano, Italy). Finally, the suspensions were centrifuged at 12,000 g for 20 min, washed repeatedly with Millipore water, and allowed to dry under vacuum for characterization. Exactly the same procedure, but in the absence of DAB, was used to prepare control samples in all three different temperature treatments.

(2) Structural and Morphological Characterization Methods

HAP crystallinity was investigated by XRD using CuKα1 radiation from a Rigaku rotating anode X-ray generator (operating at 50 kV, 100 mA) and an R-AXIS IV image plate. Samples were sealed in Lindemann capillaries. FTIR studies were performed using a Nicolet 6700 spectrometer (Thermo Scientific, Waltham, MA) equipped with an attenuated total reflectance accessory with a diamond crystal (Smart Orbit™, Thermo Electron Corporation, Madison, WI). Samples were firmly pressed against the diamond and spectra were recorded at 4 cm−1 res-
olution. A minimum of 64 scans were collected and signal aver-
egaged. Dendrimer content in HAP samples was determined by thermogravimetric analysis (TGA) performed on a TGA 2050 analyzer (TA instruments, New Castle, DE). HAP nanoparticles were heated at a rate of 10°C/min up to 700°C under air flow and then remained at that temperature for 2 h. Morphology and crystalline size of the nanorods were investigated by TEM using an FEI CM20 TEM (Philips, Eindhoven, the Netherlands).

Further, to estimate the dendrimer weight content (%) of different HAP samples prepared, thermogravimetry was used. HAP nanoparticles were dispersed by ultrasound in ethanol and deposited on a copper grid supporting a perforated carbon film. Bright-field micrographs and selected area electron diffraction patterns (SAED) were recorded. It should be noted that HAP particles are reported to be sensitive to the electron beam radiation.37 In the present study, the HAP particles were found to be stable under a focused electron beam for a period longer than 120 s. Consequently, TEM characterization was performed within 60 s with a defocused electron beam to avoid artifacts. Using the TEM pictures, the size distribution (length and width) of HAP particles was further estimated (for this purpose, three pictures for each case were used for the measurements and ana-
yzed using the ImagePro software). Raman spectroscopy was performed to confirm the results of SAED and XRD analyses using a confocal Raman microscope Renishaw RM1000 system (Renishaw, New Mills, UK) consisting of a 632.8 nm laser and a Leica DM/LM microscope (magnification used was ×50). The entrance slit to the spectrometer was set to 50 μm while the grating of the spectrometer has 1800 lines/mm. Spectra were acquired with a Peltier-cooled CCD detector (Renishaw) using either the continuous or static scan mode (no rotation of the grating) of the WIRE system interface, which is controlled through the GRAMS32 software.

III. Results and Discussion

FTIR spectroscopy was used to confirm the chemical structure of resulting nanorods. In all spectra of samples prepared in the presence of DAB, a rather sharp band at 3571 cm−1 assigned to the symmetric stretching of OH groups together with the band at 631 cm−1 attributed to the stretching mode of the hydroxyl group, and the bands at 1088, 1046 (shoulder), 1022, and 1012 cm−1 all assigned to the asymmetric and symmetric stretching of P–O bonds of the phosphate groups together with the bands at 599, 574 (shoulder), 560, 472, and 462 cm−1 assigned to the bending modes of O–P–O bonds clearly suggest the formation of HAP.4 The characteristic bands of DAB at 2952, 2814, 1456, and 1419 cm−1 (CH2 stretching and bending modes) are also present in the spectra, their intensity varying according to the Ca2+:dendrimer molar ratios used during their synthesis (Fig. 1). A peak at 2654 cm−1 clearly suggests that the NH2 end groups of the dendrimer are protonated as expected due to their interaction with the phosphate groups of HAP. Finally, additional peaks at 1410 (shoulder) and 875 cm−1 suggest the presence of carbonates in the samples due to traces of dissolved CO2 in the starting solutions. Because carbonated HAP is more close to biological apatites, which contain several foreign ions, mainly carbonate that plays a vital role in bone metabolism, no attempt was made to produce noncarbonated HAP in this study.

Further, to estimate the dendrimer weight content (%) of different HAP samples prepared, thermogravimetry was used. HAP nanoparticles were heated at a rate of 10°C/min up to 700°C under airflow and then allowed to remain at that...
temperature for 2 h, to ensure that constant weight is reached. Under these conditions, DAB is quantitatively decomposed (99.6%). All thermal decomposition profiles are alike, differing only in the amount of weight loss that reflects the organic fraction in the materials and the loss of apatite hydroxyl groups. Taken that the initial weight loss (up to 120°C) is due to moisture loss from the samples, the weight loss for each preparation is calculated and presented in Table I. In addition, the weight loss of calcium phosphate prepared under the same conditions and hydrothermally treated at 130°C but in the absence of DAB (control sample) is also registered. The weight loss found in this case (3.28%) corresponds to the loss of the OH groups of Ca10(PO4)6(OH)2 (theoretical value: 3.38%). From the results shown in Table I, it is evident that the hydrothermal treatment temperature and initial Ca2⁺:dendrimer molar ratio have a clear impact on the final DAB content. Higher crystallization temperatures lead to significantly lower polymer content as temperature increase reduces the equilibrium binding constant of the ionic interaction between the dendrimer and the charged HAP surface. On the other hand, increase of the used Ca2⁺:dendrimer molar ratio, as expected, leads to a decrease of the final DAB content in HAP nanoparticles, which, however, seems to reach a plateau value when the ratio value exceeds the 1:1 for all the three different treatment temperatures examined.

XRD experiments were used to examine phase purity and the structure of HAP samples prepared with different treatment procedures and DAB content. The phase purity and structure of different powders prepared were investigated by XRD analysis. As shown in Fig. 1(a), the powders, obtained in the absence of DAB, consist of a mixture of phases depending on treatment temperature. Thus, at room temperature, the main phase detected is dibasic calcium phosphate dehydrate (CaPO4(OH)2H2O, DCPD or brushite, JCPDS 9-77), which coexists with an amorphous phase as well as with small amounts of its dehydrated form, dibasic calcium phosphate anhydrous (CaPO4(OH), DCPA or monetite), (JCPDS 9-80), and poorly crystalline HAP (JCPDS 9-432) or possibly octacalcium phosphate (Ca8H2(PO4)6 5H2O, OCP, JCPDS 26-1056). When the sample is subjected to hydrothermal treatment at 80°C, then the main phase becomes monetite, which coexists with larger amounts (compared with what was observed in the previous case) of HAP. The powder becomes single phase consisting of HAP only after hydrothermal treatment at 130°C.

In contrast, as shown in Fig. 2(b), the powders obtained in the presence of DAB exhibit in all cases the characteristic diffraction patterns of hexagonal HAP (JCPDS 9-432). This is also true even for the samples obtained at room temperature without any hydrothermal treatment, although they present very broad peaks denoting a very fine nanostructure. The hydrothermal treatment improves significantly the crystallinity especially in the case where hydrothermal treatment is performed at 130°C. In this case, the diffraction peaks are clearly resolved and are in excellent agreement with those of pure HAP (JCPDS 9-432).

Raman spectroscopy was also performed to confirm the results of SEM and XRD on HA formation. The Raman spectrum of HAP has a prominent PO4 symmetric stretching mode (ν1) at 962 cm⁻¹, which is clearly evident in the spectra of all calcium phosphate compounds prepared in the presence of DAB and hydrothermally treated either at 80°C or 130°C (Fig. 3, spectra a, b, and c). In addition, in these spectra, it is possible to resolve three peaks attributed to asymmetric stretching mode (ν3) of the PO4 group at 1073, 1048, and 1030 cm⁻¹ as well as three peaks attributed to bending mode (ν3) of the PO4 group at 608, 591, and 579 cm⁻¹. The OH stretching frequency appears at 3572 cm⁻¹, while it is also possible to discern peaks at 2934 cm⁻¹.
cm\(^{-1}\) assigned to CH\(_2\) asymmetric stretching mode and at 2875 and 2825 cm\(^{-1}\) assigned to CH\(_2\) symmetric stretching modes.\(^{40,41}\) The same CH\(_2\) peaks are also observed in the spectra of calcium phosphate prepared at room temperature in the presence of DAB (Fig. 3, spectrum d), which, however, does not exhibit the characteristic HAP peak at 962 cm\(^{-1}\) but a rather broad peak at 953 cm\(^{-1}\), together with a low-intensity broad peak at 1073 cm\(^{-1}\). The \(v_1\) PO\(_4\) mode of ACP has been observed at 950 cm\(^{-1}\), the band shifting to higher wavelengths (viz. 962 cm\(^{-1}\)) upon transformation of ACP to crystalline HAP.\(^{42}\) Therefore, the position of the band at 953 cm\(^{-1}\) could suggest the existence of very fine nanostructured HAP in accordance with the X-ray and TEM (see below) analyses. Finally, the spectra of control samples, prepared in the absence of DAB after hydrothermal treatment, confirm the presence of a mixture of crystalline calcium phosphates. In fact, in the spectrum of the sample hydrothermally treated at 80°C (Fig. 3, spectrum e), the high-intensity Raman peak is observed at 988 cm\(^{-1}\) and can be attributed to monetite or brushite.\(^{43,44}\) While a lower intensity band at 962 cm\(^{-1}\) is also observed indicating the presence of HAP. Additional bands at 1132 and 1092 cm\(^{-1}\) can be assigned to \(v_3\) PO\(_4\) stretching of monetite, the band at 590 cm\(^{-1}\) to HAP, monetite, and brushite, while the bands at 575 and 569 cm\(^{-1}\) can be assigned to \(v_3\) PO\(_4\) of brushite. On the whole, in this case, a mixture of calcium phosphate crystalline phases is present, with brushite being the main component in full agreement with the XRD data.

Overall, the results clearly show that the presence of DAB during crystallization affects the crystal growth and nucleation procedures determining the phase content of the final powders obtained. Another important parameter is the hydrothermal treatment, which has also a clear effect on powders’ diffraction patterns. In contrast, not any marked differences among the patterns of samples obtained at different Ca\(^{2+}\):dendrimer molar ratios could be noticed.

To investigate particle size, morphology, and homogeneity of the different samples prepared, TEM analysis was used. Figure 4 shows that when the synthesis is performed in the presence of DAB elongated rod-like nanoparticles are developed, the dimensions of which depend greatly on the conditions used. As expected, the temperature of treatment procedure (treatment at room temperature or hydrothermal treatment at 80°C or 130°C) is an important parameter affecting considerably the particle size.

In fact, by increasing the temperature of hydrothermal treatment from 80°C to 130°C, a considerable rise of mean particle size is observed followed by a small decrease of particle aspect (length to width) ratio (Table II), whereas the samples kept at room temperature present a very fine nanostructure with the mean length and width reaching down to 11 and 5 nm, respectively (Fig. 4(a)). SAED patterns, obtained for all three temperature treatments, confirmed the crystal structure of the samples showing the diffraction rings from the characteristic hexagonal HAP planes in agreement with the XRD results. A very interesting observation is that the dendrimer concentration during the synthesis reaction, i.e., the Ca\(^{2+}\):dendrimer molar ratio, has also a great effect on particle size. The presence of dendrimer hinders the HA grain growth and as a consequence the lesser the dendrimer during reaction, the larger is the mean grain size of HAP obtained. This is observed irrespective of hydrothermal treatment temperature. Thus, the coarsest structure, with elongated grains of a mean length and width at around 81 and 22 nm, respectively, was obtained in the case of the sample prepared using the highest Ca\(^{2+}\):dendrimer molar ratio, 2:1, after hydrothermal treatment at 130°C (Fig. 4(h)).

More specifically, observing the particle size distributions obtained for the different cases examined (Fig. 5) as well as the mean particle size values in Table II, it can be easily seen that when HAP nanorods are formed at 80°C, a behavior is observed at higher crystallization temperatures, i.e., 130°C, although both particles’ length and width are higher (almost double compared with the respective values measured after treatment at 80°C). Again, an increase of the Ca\(^{2+}\):dendrimer molar ratio from 1:2 to 1:1 during reaction decreases HAP particle’s length by some 50%, whereas a further increase of this ratio to 2:1 (HAPc) does not decrease the length any further, suggesting that a saturation level is reached. It should also be noted that DAB content has in this case no effect on nanorods’ width. It can be derived, thus, that crystal size decreases in length, but not in width, by increasing DAB content up to a certain value. This result is also confirmed by the TGA results, which again showed the presence of a saturation level for DAB content during reaction above which the DAB content of crystals does not increase or increases marginally.

At this stage, in an effort to better clarify the role of dendrimer concentration in reaction solution during synthesis, very dilute solutions exhibiting, however, the same molar ratio of precursors reactants but concentrations that are the 1/10 of those discussed up to now were used to synthesize HAP particles. The analysis showed very similar results (data not shown) with the particle size distributions obtained differing only slightly compared with the respective curves obtained from the more concentrated solutions, suggesting, thus, the Ca\(^{2+}\):dendrimer molar ratio and not the dendrimer concentration itself (at least in the concentration range examined) as the determining parameter for the final particle size.

The significant effect of dendrimer on particle morphology is also evidenced observing the images obtained for samples prepared in the absence of DAB (Figs. 4(c), (f), and (i)). In all three different temperature treatments examined, particles of varying sizes and/or morphology were found coexisting in the samples. This is in accordance with the XRD analysis, which detected a mixture of phases especially in the samples prepared at room temperature or hydrothermally treated at 80°C. Thus, at room temperature, very fine, poorly crystalline nanoparticles along with nanoparticles of an amorphous calcium phosphate phase, according to SAED analysis at various areas, were detected. Particles of two different morphologies were also detected in the case of hydrothermal treatment either at 80°C or 130°C. In both cases, very long needle-like crystals coexist with nanorods. This
is also reflected on particle size distributions obtained for these samples, which are very broad compared with the respective curves plotted for the samples prepared in the presence of DAB. It can be easily concluded, in this way, that DAB affects the whole crystallization process leading to HAP nanorods of homogeneous particle size distributions with mean values depending on dendrimer content during the synthesis reaction and treatment temperature, in contrast with the material received in the absence of dendrimer that is polyphasic and inhomogeneous.

Literature data on particle size and morphology obtained in the presence of dendrimers or dendritic polymers are limited up

### Table II. Particle Size Characteristics of Hydroxyapatite (HAP) Nanoparticles Obtained under the Different Conditions Used

<table>
<thead>
<tr>
<th>Treatment temperature (°C)</th>
<th>Ca$^{2+}$ : dendrimer molar ratio</th>
<th>Length (nm)</th>
<th>Width (nm)</th>
<th>Aspect ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td>RT</td>
<td>Without DAB</td>
<td>11.6 ± 7.3</td>
<td>5.9 ± 1.9</td>
<td>2.0</td>
</tr>
<tr>
<td></td>
<td>2:1</td>
<td>11.3 ± 2.1</td>
<td>7.1 ± 1.7</td>
<td>1.6</td>
</tr>
<tr>
<td></td>
<td>1:2</td>
<td>9.6 ± 3.1</td>
<td>5.0 ± 2.0</td>
<td>1.9</td>
</tr>
<tr>
<td>80</td>
<td>Without DAB</td>
<td>46.9 ± 37.5</td>
<td>9.7 ± 5.0</td>
<td>4.8</td>
</tr>
<tr>
<td></td>
<td>2:1</td>
<td>43.4 ± 18.7</td>
<td>10.8 ± 3.1</td>
<td>4.0</td>
</tr>
<tr>
<td></td>
<td>1:1</td>
<td>23.0 ± 8.7</td>
<td>8.8 ± 2.0</td>
<td>2.6</td>
</tr>
<tr>
<td></td>
<td>1:2</td>
<td>22.5 ± 7.4</td>
<td>7.1 ± 1.7</td>
<td>3.2</td>
</tr>
<tr>
<td></td>
<td>1:3</td>
<td>18.4 ± 6.5</td>
<td>6.0 ± 1.4</td>
<td>3.2</td>
</tr>
<tr>
<td>130</td>
<td>Without DAB</td>
<td>77.0 ± 83.5</td>
<td>24.1 ± 14.3</td>
<td>3.2</td>
</tr>
<tr>
<td></td>
<td>2:1</td>
<td>81.4 ± 36.6</td>
<td>21.6 ± 6.3</td>
<td>3.8</td>
</tr>
<tr>
<td></td>
<td>1:1</td>
<td>41.1 ± 19.5</td>
<td>19.6 ± 5.0</td>
<td>2.1</td>
</tr>
<tr>
<td></td>
<td>1:2</td>
<td>41.1 ± 17.6</td>
<td>20.6 ± 5.0</td>
<td>2.0</td>
</tr>
</tbody>
</table>

RT, room temperature.
to now. Donners et al. were the first who studied the morphology of HAP obtained at room temperature in the presence of amphiphilic cationic PEI dendrimers functionalized with long alkyl chains but in the presence of surfactants such as octa-decylamine, hexadecyltrimethylammonium bromide, or sodium dodecylsulfate. Plate like nanoparticles in size could be obtained when CTAB was used, whereas in the presence of the other two amphiphiles mixtures of plates with needles or clusters were received. In following studies, the ionic starburst PAMAM dendrimer was investigated as a nucleation agent for HAP. It was shown that the kind of surface groups in dendrimer, its generation, and its concentration in solution, all affect the size and/or morphology of obtained HAP. Using PAMAM of generation 5.5 with surface carboxylate groups, fibers (200 nm/15 nm) were obtained at room temperature, while nanorods (80 nm × 10 nm) were formed after hydrothermal treatment at 150°C. Similar nanorods were obtained with PAMAM of various generations with amido terminal groups after hydrothermal treatment at 150°C. However, mixtures of elliptical particles and nanorods were received in the presence of lower generation carboxylic or polyhydroxyl terminated PAMAM.

More recently, cationic linear or branched PEI was investigated for calcium phosphate mineralization at room temperature. PEI, in all cases, proved (after a 7 days mineralization procedure) an efficient template for the fabrication of sub-100 nm spherical HAP/polymer hybrid particles at pH values above 8. In contrast, needle-like brushite was observed at initial pH 5. A nucleation and growth mechanism was suggested based on a mineralization-trapping pathway, where at the beginning of the precipitation small CaP particles form. At pH values of ca. 8, particle growth is prevented by PEI absorption onto initial particles, which are further stabilized by adsorption and reproto-

nation of PEI during the rest of mineralization process. At low pH, however, much larger particles form, which most likely grow via heterogeneous nucleation and growth on existing, polymer modified CaP surfaces. DAB, which has a similar chemical structure to that of PEI, could act in a similar pathway affecting HAP nucleation and inhibiting the growth process, by its absorption on crystal nuclei, providing thus a flexible and rapid way to synthesize nanorods of tailored size.

IV. Conclusions

Nanostructured HAP was synthesized in the presence of amine terminated diaminobutane poly(propylene imine) dendrimer using various calcium:dendrimer molar ratios at room temperature and at two different hydrothermal treatment temperatures. In all cases, hexagonal hydroxyapatite nanorods of narrow particle size distributions were obtained. In contrast, in control samples, prepared in the absence of DAB, pure HAP phase was only detected after hydrothermal treatment at 130°C, whereas the particle size distributions were very broad and the morphology inhomogeneous. The calcium to dendrimer molar ratio used proved to be an important parameter controlling the crystallization procedure and affecting the final crystal size and the final dendrimer content into the HAP nanocrystals. These results can provide the basis for facile method of HAP formation, which can be utilized in biomedical applications. Further work toward this end is under way.

Acknowledgment

The authors are most grateful to Dr. Elias Chatzitheodoridis for operating the Raman instrumentation.