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Enhanced vaterite and aragonite crystallization at controlled ethylene glycol concentrations

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ABSTRACT

Calcium carbonate ($CaCO_3$) has three distinct anhydrous polymorphs, namely vaterite, aragonite and calcite. Although there is a high demand for aragonite and vaterite polymorphs for biomedical use, their unstable nature makes it challenging to synthesize them compared to calcite, which is the most stable form of $CaCO_3$. Despite the remarkable effort on stabilizing vaterite and aragonite polymorphs in aqueous solutions, phase-pure vaterite and aragonite polymorphs have not been synthesized yet, without referring to the use of additives, surfactants or elevated temperatures. Herein, the effect of ethylene glycol (EG) concentration and temperature on the formation of vaterite and aragonite particles were investigated at 25 °C and 70 °C. Results showed that 60% EG containing precursor solution -without any other additive- can prevent vaterite/aragonite-to-calcite transformation regardless of the synthesis temperature. Furthermore, the size of $CaCO_3$ particles decreased as EG concentration increased and it reached its minimum average values at 80% EG. The results of this study revealed the potential use of the proposed synthesis route to stabilize vaterite and aragonite polymorphs, tailor their content, morphology and size without using any additives, surfactants and elevated temperatures.

Keywords: Vaterite, Aragonite, Ethylene Glycol, Biomaterial

1. INTRODUCTION

Calcium carbonate ($CaCO_3$) is one of the most abundant minerals on earth [1]. Owing to its availability, non-toxic nature and ideal mechanical properties, $CaCO_3$ is extensively used in cosmetic and healthcare industries [2]. $CaCO_3$ exists in three anhydrous polymorphs, namely vaterite, aragonite and calcite having hexagonal, orthorhombic and rhombohedral crystal structures, respectively [3]. Owing to their biocompatible nature and dispersibility, numerous studies were conducted on the synthesis and transformation of $CaCO_3$ particles to reveal their potential as drug delivery vehicles and additives for orthopedic applications [4,5]. One such example was the use of vaterite particles which are degradable in vivo due to their high solubility in acidic components, i.e. metabolites and hydrolysis enzymes [6]. Thus, utilizing vaterite particles as biodegradable pH-sensitive drug delivery vehicles became prominent [7,8]. Recently, Wang et al. [9] synthesized folic acid modified spherical vaterite particles for doxorubicin (DOX) delivery and observed an

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increased DOX release rate with a decrease in pH in vitro. Similarly, Qiu et al. [10] investigated anti-cancer drug camptothecin loaded vaterite particles and reported an 8-fold increase in the cumulative release of camptothecin at pH 6.0 (simulates cancerous tissue) compared to its release at physiological pH. Along the same line, aragonite also exhibited a great potential as a drug delivery vehicle. Kamba et al. [11] prepared DOX incorporated aragonite particles to fight against breast cancer and observed that DOX treatment was more effective in reduction of cancer cell viability when it was delivered within aragonite particles than that of itself alone. Saidykhan et al. [12] prepared vancomycin loaded aragonite particles for treatment of osteomyelitis (infection in bone) and reported that particles were nontoxic and effective against Staphylococcus aureus growth in vitro. These studies indicated the potential use of vaterite and aragonite particles for drug delivery applications, therefore synthesis methods to obtain these polymorphs at room temperature need to be addressed.

Among various methods to synthesize $CaCO_3$ particles, solution mixing is the most commonly used technique due to its simplicity and ease of control on the process parameters [7]. However, aqueous conditions combined with severe stirring/heating during the solution mixing technique favor the dissolution of unstable $CaCO_3$ polymorphs (vaterite and aragonite) and transform to thermodynamically more stable calcite polymorph. As a matter of fact, this phenomenon was proved to be independent of the synthesis technique where various studies reported vaterite ($\log K_{sp,vaterite} = -7.913$; K_{sp}: solubility product) to crystallize initially from amorphous $CaCO_3$ ($\log K_{sp} = -6.400$), followed by transformation to aragonite ($\log K_{sp,aragonite} = -8.360$) or calcite ($\log K_{sp,calcite} = -8.475$) by dissolution and recrystallization [13,14]. Log K_{sp} values of vaterite and aragonite polymorphs are less negative compared to calcite, indicating the difficulty in stabilizing these polymorphs and preventing calcite transformation in aqueous environments. One potential method to prevent calcite transformation is to provide chemical conditions which inhibit vaterite/aragonite dissolution in the first place. Accordingly, the concentrations of free polymorph ions in the solution become insufficient for calcite supersaturation, and thus vaterite/aragonite polymorphs are stabilized.

In literature, there are various techniques to alter supersaturation by changing the free ionic content of the solution. For instance, additives and surfactants were incorporated into aqueous $CaCO_3$ precursor solutions to control their supersaturation by reacting them with calcium (Ca^{2+}) or carbonate (CO_3^{2-}) ions and determine their influence on $CaCO_3$ crystallization [7,15]. Chen et al. [16] added hexadecyl(trimethyl) azanium bromide (CTAB) and sodium dodecyl sulfate (SDS) surfactants inside the precursor solutions and transformed vaterite into aragonite by altering CTAB/SDS ratio from 0 to 5. Similarly, Yan et al. [17] supplied Pluronic F-127 and SDS into the solution and observed favored vaterite formation upon their simultaneous addition. On the other hand, for a scenario where additives were not incorporated into the solution, a higher energy was required to crystallize the aragonite polymorph, and thus high temperature applications became a potential route for aragonite synthesis. Chen et al. [18] altered synthesis temperature and successfully crystallized vaterite particles at 30-40 °C, while aragonite particles were observed at 80 °C.

Although most of the studies reported incorporation of surfactants or high synthesis temperatures to stabilize vaterite and aragonite particles, the use of ethylene glycol (EG) as a precursor solvent to control stability of $CaCO_3$ particles and polymorphic transformations might be an alternative approach [19,20]. EG is a non-aqueous polar solvent and miscible in water at all proportions [20,21]. Since water increases the solubility of metastable polymorphs and favor formation of calcite, using EG instead of water can limit the formation of calcite. In fact, EG based precursors were reported for the synthesis of different $CaCO_3$ morphologies [22,23]. However, systematic changes of precursor EG concentration at multiple temperatures and correlating the associated structural changes of $CaCO_3$ particles were not studied in detail. Herein, a simple processing route to decrease the solubility of vaterite and aragonite through the use of EG solvent was presented. The processing route can serve as a simple alternative to control the formation of $CaCO_3$ particles with various morphologies and sizes, potentially offering ideal candidates for drug delivery and orthopedic applications.

2. EXPERIMENTAL METHODS

2.1. Materials and Methods

Calcium acetate ($Ca(C_2H_3O_2)_2$, Sigma Aldrich), sodium bicarbonate ($NaHCO_3$, Sigma Aldrich), ethylene glycol (EG; $(CH_2OH)_2$, Sigma Aldrich, ≥99%) and ethanol (C_2H_6O, Merck, ≥99.9%) were obtained commercially and used as-received. Ultrapure water (Millipore, Milli-Q purification) was used for each experiment.

Particle synthesis was performed at two temperatures; 25 and 70 °C. For each temperature, 0-90 vol% EG containing aqueous solutions were prepared to investigate the effect of EG concentration on crystallization of $CaCO_3$ polymorphs. To synthesize $CaCO_3$ particles, aqueous calcium acetate (0.3 M) and sodium bicarbonate (0.9 M) precursor solutions were prepared separately, followed by incorporation of EG into each solution to obtain a total volume of 25 ml. Once the temperature of the solutions were stabilized at the desired value (25 or 70 °C), solutions were mixed quickly under magnetic agitation at 500 rpm for 15 minutes to initiate $CaCO_3$ precipitation. Afterwards, the solution was kept at the same temperature for 1 hour without agitation. Then, the precipitate was washed using ethanol and ultrapure water, respectively. Lastly, $CaCO_3$ particles were collected by centrifugation (7200 rpm, 15 minutes, 22 °C) and dried at 50 °C.

2.2. Characterization

The $CaCO_3$ polymorphs were analyzed using Rigaku D/Max-2200 X-ray diffractometer with monochromatic Cu K_α radiation (λ=1.54 Å) at 40 kV accelerating voltage and 30 mA current. $CaCO_3$ particles were scanned between 20-60° (2θ) at a scanning rate of 2°/min. Rietveld refinement analysis was performed using General Structure Analysis System II (GSAS-II) to calculate weight percentages of each $CaCO_3$ polymorph [24]. Morphology and size of particles were investigated using Nova Nano SEM 430 electron microscope at 20 kV accelerating voltage. Prior to SEM imaging, particles were coated with a thin layer of gold to create a conductive path using Quorum SC7640 high resolution sputter coater. ImageJ software was used to calculate average particle size from 30 particles for each sample. For size measurements, longest axis values of the predominant polymorph (vaterite or aragonite) were reported. $CaCO_3$ polymorphs were also characterized using Perkin Elmer Spectrum 100 Fourier Transform Infrared (FTIR) spectrometer using attenuated total reflection (ATR) configuration. Each sample was scanned 4 times in 1000-600 cm^{-1} range with a resolution of 4 cm^{-1}.

3. RESULTS AND DISCUSSION

The XRD patterns of $CaCO_3$ particles synthesized using different concentrations of EG at 25 °C were shown in Figure 1a. In the spectra, all peaks could be indexed to vaterite (ICDD: #033-0268) or calcite (ICDD: #005-0586) polymorphs. In each sample, vaterite peaks at 24.92°, 26.99° and 32.78° corresponding to (110), (112) and (114) crystallographic planes, and/or calcite peaks at 29.40°, 35.90° and 39.50° corresponding to (104), (110) and (113) crystallographic planes were observed at varying intensities [25]. The variation of polymorph content in synthesized $CaCO_3$ particles with respect to the EG concentration is calculated by Rietveld Refinement of XRD patterns given in Figure 1a. As shown in Figure 1b, in the absence of EG in the precursor solution (ultrapure water), $CaCO_3$ particles were composed of 50 wt% calcite and 50 wt% vaterite. As EG concentration of the precursor solutions gradually increased, vaterite fraction of the synthesized

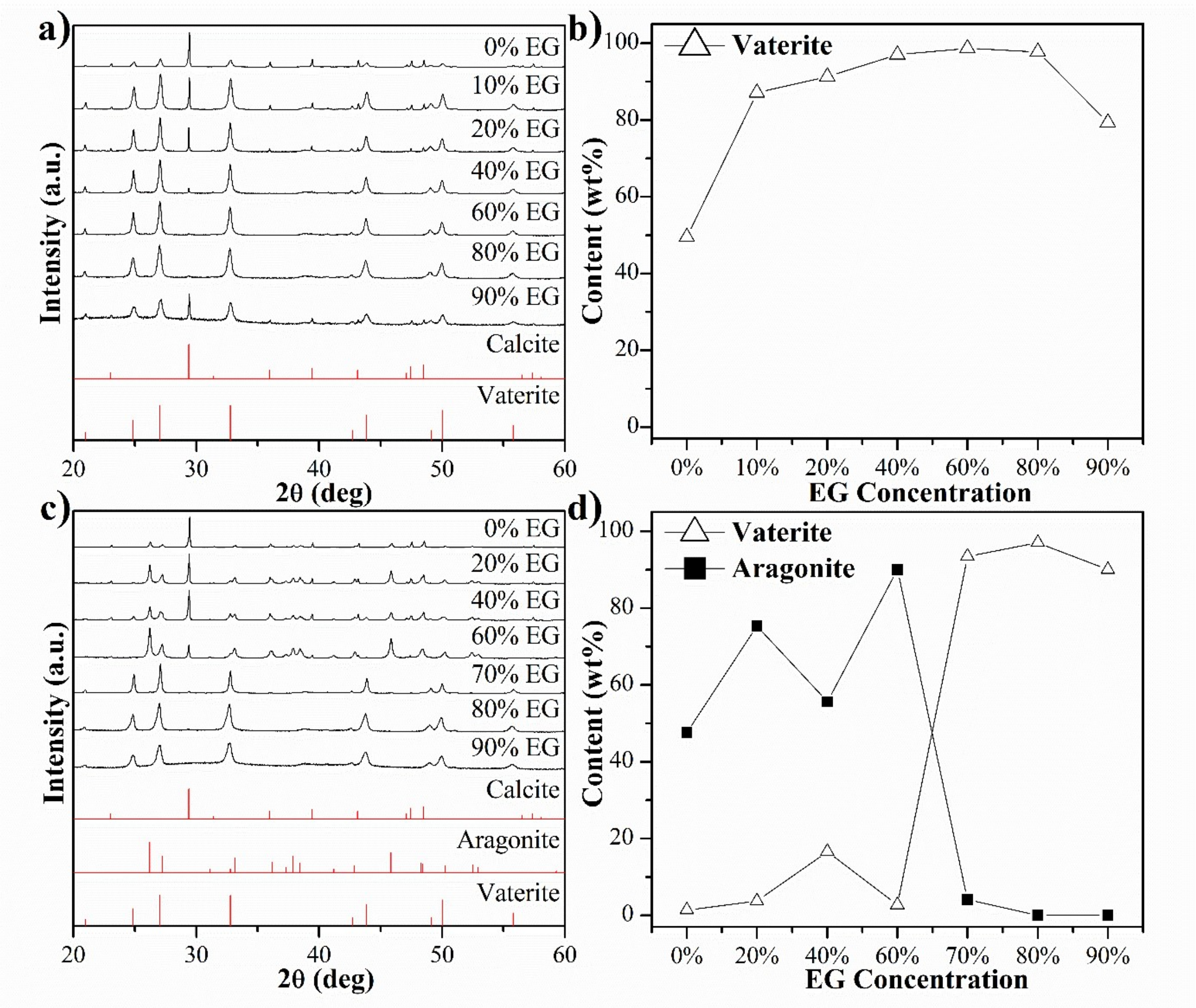

Figure 1. XRD spectra of $CaCO_3$ particles synthesized at a) 25 °C and c) 70 °C. Weight percentages of vaterite and aragonite at b) 25 °C and d) 70 °C in different EG concentrations. Red lines are ICDD references for calcite, aragonite and vaterite.

powder increased, reaching a maximum value at 60% EG. As mentioned previously, calcite is the stable form of $CaCO_3$ at ambient conditions and synthesis of vaterite or aragonite without forming calcite is challenging [26]. However, almost phase-pure vaterite (98 wt%) was obtained in this study by incorporating 60% EG into the aqueous precursor solutions. Having this said, when the $CaCO_3$ synthesis temperature increased to 70 °C, aragonite particles (ICDD: #005-0453) along with calcite and vaterite particles formed. XRD patterns of $CaCO_3$ particles synthesized using different concentrations of EG at 70 °C were displayed in Figure 1c. Characteristic peaks of aragonite at 26.3° and 45.9° corresponding to (111) and (221) crystallographic planes and calcite peaks at 29.40°, 35.90° and 39.50° corresponding to (104), (110) and (113) crystallographic planes were observed, respectively [27]. As shown in the Rietveld refinement results (Figure 1d) of the XRD patterns given in Figure 1c, incorporation of EG into the precursor solutions favored formation of metastable $CaCO_3$ polymorphs (vaterite and aragonite) rather than calcite polymorph. In the absence of EG in the precursor solution, $CaCO_3$ particles consisted of 51 wt% calcite and 48 wt% aragonite. Upon the incorporation of 20% EG into the precursor solutions, vaterite/aragonite-to-calcite transformation was suppressed. At this EG concentration, particles contained 4 wt% vaterite, 75 wt% aragonite and 21 wt% calcite. As EG concentration increased further, the aragonite and vaterite content of the synthesized particles significantly altered. Up to 60% EG, aragonite formation dominated over vaterite, whereas after 70% EG, vaterite content maximized. These results indicated 60-70% EG concentration range to be critical in determining the degree of vaterite-

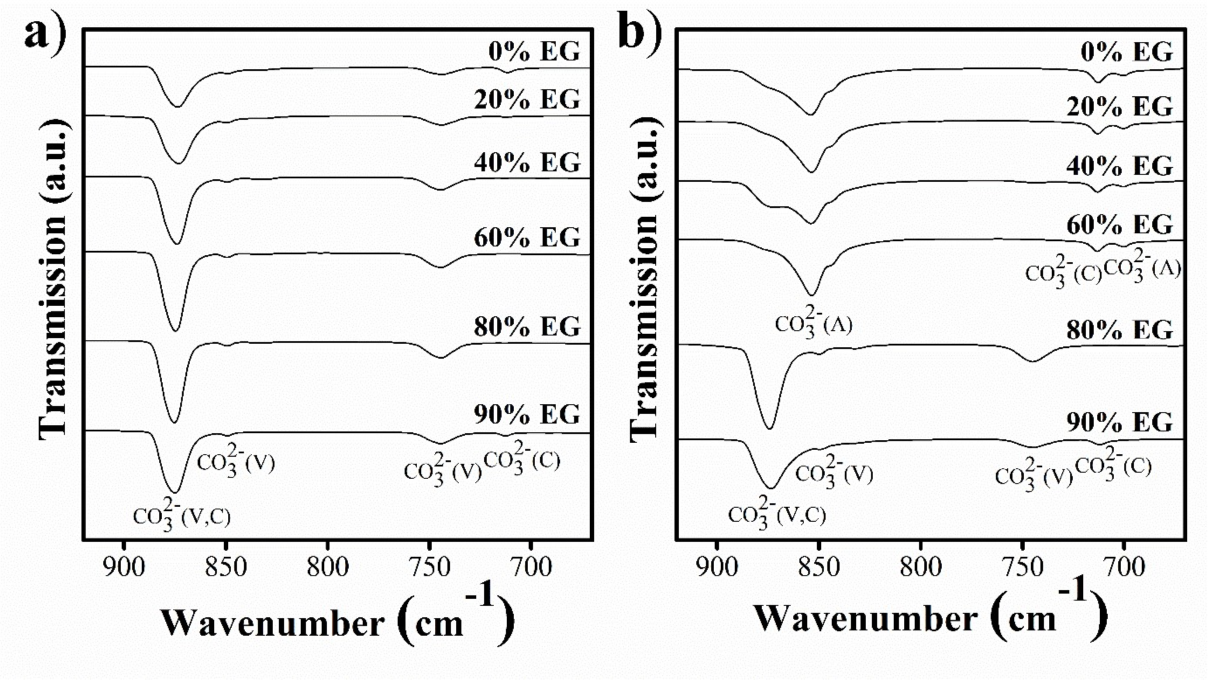

Figure 2. FTIR spectra of $CaCO_3$ particles synthesized using 0%, 20%, 40%, 60%, 80% and 90% EG at a) 25 °C and b) 70 °C. ‘‘V’’, ‘‘A’’ and ‘‘C’’ denote peaks corresponding to vaterite, aragonite and calcite polymorphs, respectively.

to-aragonite transformation at moderate synthesis temperature.

XRD findings were in agreement with previously published literature on $CaCO_3$ synthesis [20,28]. As previous literature suggested, incorporation of EG into the precursor solutions provided hydroxyl (OH^-) groups for the system. These (OH^-) groups possessed high cohesive energy and attracted oppositely charged free Ca^{+2} cations [20,28]. The adsorption of (OH^-) ions changed the surface free energy of vaterite leading to higher thermodynamic stability compared to calcite, and thus, vaterite dissolution was prevented [28,29]. In fact, the efficiency for suppression of calcite crystallization depended on the amount of OH^- groups supplied by EG incorporation into the precursor solutions.

Aragonite particles, on the other hand, required extra energy to crystallize, which was typically provided via increasing the synthesis temperature. For instance, Flaten et al. [28] were able to synthesize aragonite particles (up to 76 wt%) at 80 °C, whereas repeating the same procedure at room temperature failed to obtain aragonite. In another study, Chen et al. [30] increased solution temperature during nucleation stage by microwave power and synthesized bundle-shaped aragonite particles at 90 °C. Similarly, in this study, almost phase pure vaterite particles synthesized at 25 °C using high EG concentrations, yet aragonite particles could not be formed at this temperature independent of the presence or absence of EG. Along the same line, it can be speculated that incorporation of EG limited dissolution of vaterite to form aragonite and stabilized this polymorph despite higher synthesis temperature, indicating its dominating efficacy at higher concentrations. It is clear that changing the amount of (OH^-) ion supply to the precursor solution via altering the precursor solvent type can provide potential stabilization routes for each $CaCO_3$ polymorph at temperatures $\leq$70 °C.

In addition to XRD, FTIR was also used to characterize the $CaCO_3$ polymorphs. Particles synthesized at 25 °C (Figure 2a) showed absorption bands of vaterite at 877 cm^{-1} (v_2), 849 cm^{-1} (v_2), 744 cm^{-1} (v_4) and calcite at 877 cm^{-1} (v_2), 712 cm^{-1} (v_4), whereas particles synthesized at 70 °C (Figure 2b) showed additional aragonite absorption bands at 854 cm^{-1} (v_2) and 700 cm^{-1} (v_4) [31,32]. These results were consistent with XRD results given in Figure 1, where vaterite and aragonite polymorphs were observed at different amounts depending on the EG concentration and synthesis temperature. FTIR confirmed formation of almost phase-pure vaterite using 60% EG at 25

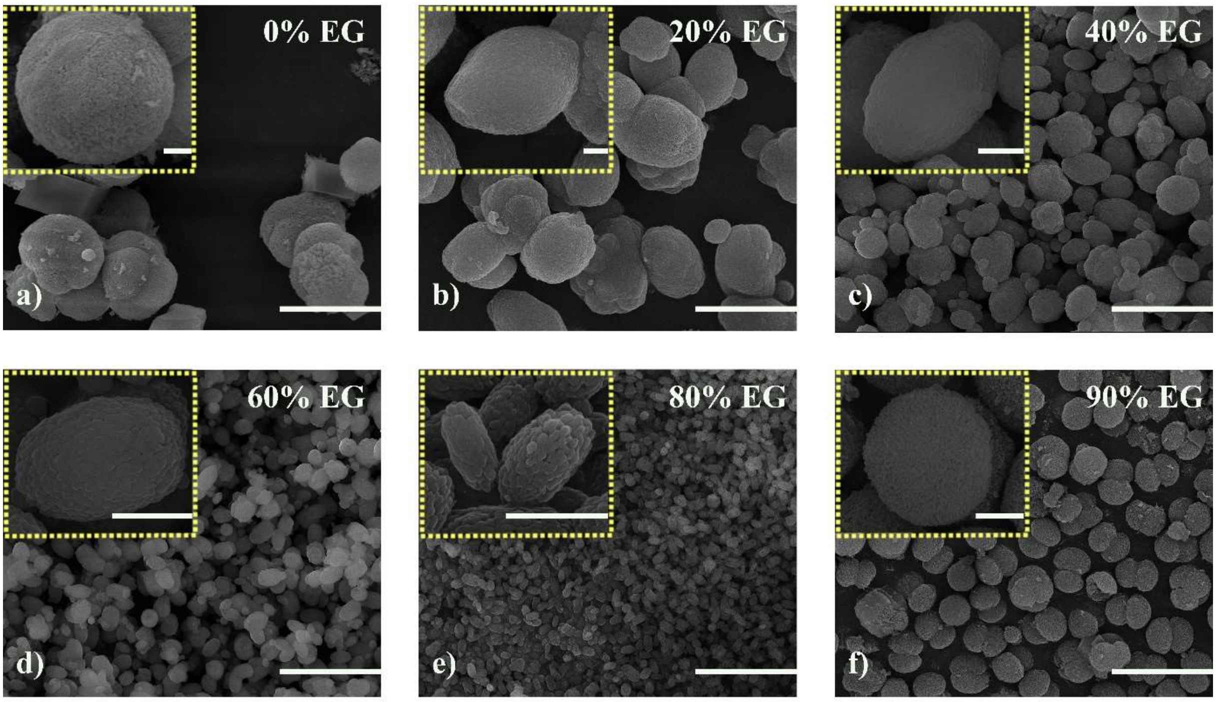

Figure 3. SEM micrographs of $CaCO_3$ particles synthesized at 25 °C using a) 0%, b) 20%, c) 40%, d) 60%, e) 80% and f) 90% EG concentrations. Images were taken at 10.000x and the scale bars are 10 µm. Insets show higher magnification micrographs of $CaCO_3$ particles and their scale bars are 1 µm.

°C and predominant formation of aragonite at EG concentration below 70% at 70 °C. Additionally, absorption bands of EG at 725 cm^{-1} (C-H) and 885 cm^{-1} (C-C) were not evident in any of the FTIR spectra, indicating complete removal of adsorbed EG from the precipitated $CaCO_3$ particles [33].

In order to investigate the effect of EG incorporation on the particle size and morphology, synthesized $CaCO_3$ particles were visualized under SEM. Figure 3 showed SEM micrographs of the particles obtained at 25 °C using various EG concentrations (0-90%). In literature, vaterite polymorph typically obtained in ellipsoidal or spherical, calcite polymorph typically obtained in rhombohedral and aragonite polymorph typically obtained in needle-like morphologies [7]. SEM micrographs of vaterite particles shown in Figure 3 were in line with these findings. At the two extreme EG concentrations (0% and 90% EG), vaterite crystals exhibited spherical growth, whereas at intermediate concentrations, they grew in ellipsoidal and spheroidal morphologies. Considering that the sphere morphology is the lowest (surface) energy form of particles synthesized in solutions, ellipsoidal vaterite particles possessed higher surface free energy at intermediate EG concentrations. It can be speculated that as vaterite dissolution was inhibited by OH^- groups of EG, highly viscous nature of EG limited migration of anions from the solution towards the surface, delaying growth of vaterite nuclei, and thus forming individual vaterite particles formed by attachment of much smaller vaterite crystals (i.e. Figure 3e inset) [33]. Along the same line, these results also suggest that there might be a competition between the oriented attachment of small crystals on their highest energy planes leading to growth into ellipsoidal particles (i.e. Figure 3e inset) and dissolution and attachment of small nucleates uniformly across the particle to obtain a more spheroidal morphology (i.e. Figure 3 b, c, d inset) [34]. In addition, particles synthesized at 0% EG had the highest average particle size (8.09 ± 1.64 µm) at 25 °C as shown in Figure 5. Incorporation of more EG into the precursor solutions decreased average particle size, reaching its minimum (0.92 ± 0.17 µm) at 80% EG. Parakhonskiy et al. [35] observed a similar trend in their study and obtained minimum vaterite particle sizes using 83% EG compared to 0% (pure water), 17% and 50% EG concentrations.

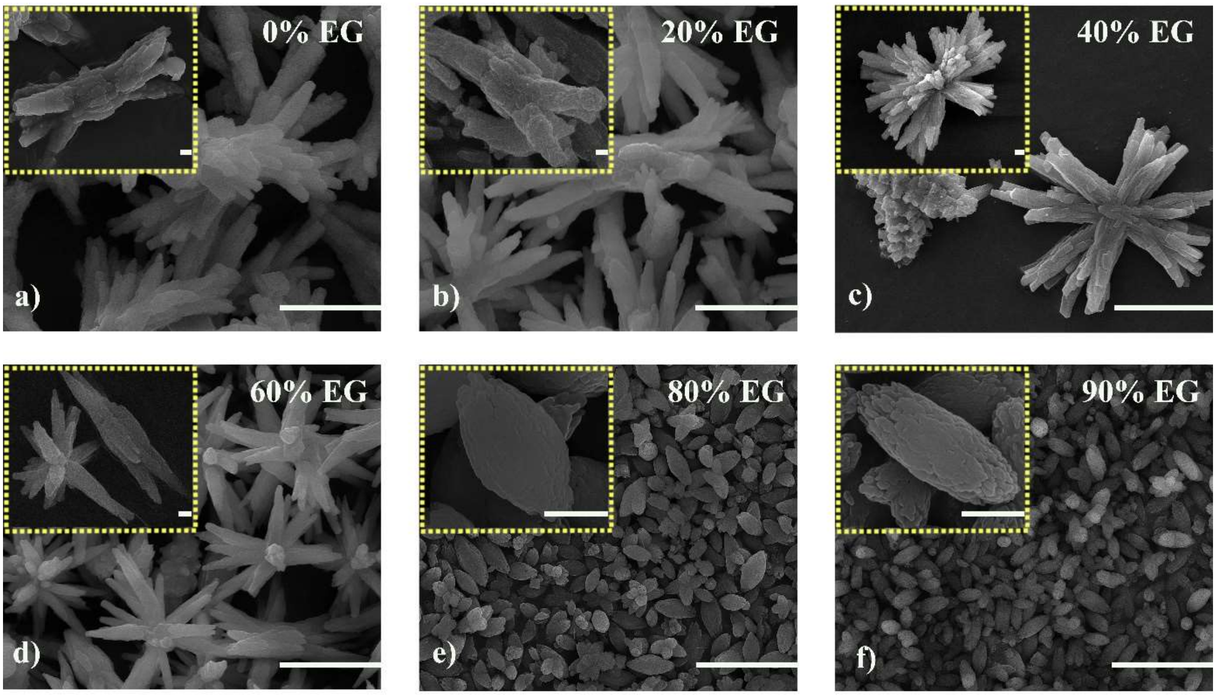

Figure 4. SEM micrographs of $CaCO_3$ particles synthesized at 70 °C using a) 0%, b) 20%, c) 40%, d) 60%, e) 80% and f) 90% EG concentrations. Images were taken at 10.000x and the scale bars are 10 µm. Insets show higher magnification micrographs of $CaCO_3$ particles and their scale bars are 1 µm.

At 70 °C (Figure 4), urchin-like aragonite particles grew at concentrations below 60% EG. The attachment of aragonite needles to each other most probably on the same crystallographic plane leading to urchin-like morphology was repeatedly observed at each EG concentration below 60%. Similar to vaterite growth, the underlying reason for attachment of needles to obtain urchin-like particles was to decrease the surface energy via growing around their highest energy crystallographic planes. For $CaCO_3$ particles synthesized at 70 °C, the effect of EG concentration on average particle size was more remarkable due to polymorphic transformation and morphological change. Figure 5 showed highest average particle size (20.60 ± 2.11 µm) at 0% EG for particles synthesized at 70 °C, while this value decreased with increasing EG concentrations, reaching its minimum (1.92 ± 0.52 µm) at 80% EG.

In this study, the effects of EG incorporation and temperature on the polymorph, morphology and size of $CaCO_3$ particles were investigated. Conclusions can be summarized under two main titles:

1 – Stabilization (dissolution inhibition) of the polymorphs under certain conditions:

To stabilize a $CaCO_3$ polymorph, one of the key parameters is decreasing its solubility in the mixed precursor solution. Higher the solubility of a polymorph -proportional to the number of the free polymorph ions in solution (i.e. Ca^{2+} and CO_3^{2-})- higher the probability of dissolution. Among four main $CaCO_3$ polymorphs, amorphous $CaCO_3$ has the highest solubility leading to its rapid formation and dissolution during the initial stages of precursor solution mixing. Once amorphous $CaCO_3$ dissolves, ions recrystallize into vaterite, which is the least stable among anhydrous forms of $CaCO_3$ (vaterite, aragonite, calcite) [36]. The inhibition of dissolution and calcite crystallization (lowest solubility and free energy) by changing the number of available ions in solution is the key approach to stabilize vaterite and/or aragonite. This study suggested that OH^- groups in EG can bind polymorph ions, therefore, alter supersaturation and lead to stabilization of unstable $CaCO_3$ polymorphs [29].

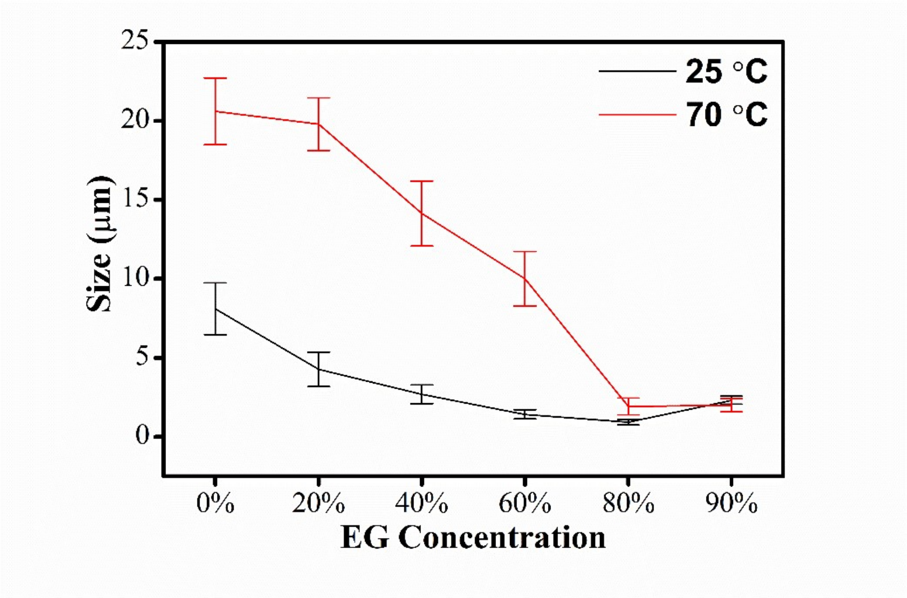

Figure 5. Average size of $CaCO_3$ particles synthesized at 25 °C and 70 °C using different EG concentrations. Values are mean ± SEM; n=3.

2 – Morphology and size of the precipitated polymorphs:

Results revealed that 60% EG as the optimum concentration to synthesize phase-pure vaterite at 25 °C and aragonite at 70 °C. At 25 °C, spheroidal (20-60% EG), ellipsoidal (80% EG) and spherical (0% and 90% EG) vaterite particles were observed. As temperature increased to 70 °C, urchin-like aragonite particles (0-60% EG) in addition to spheroidal (80% EG) and ellipsoidal (90% EG) vaterite particles were obtained. At both temperatures, average particle size was highest at 0% EG, i.e. 8.09 ± 1.64 μm at 25 °C and 20.60 ± 2.11 μm at 70 °C, whereas these values decreased with increasing EG concentration and minimum values were obtained at 80% EG, 0.92 ± 0.17 μm at 25 °C and 1.92 ± 0.52 μm at 70 °C. Though the determination of growth behavior necessitated detailed HRTEM analysis, it could be speculated that with increasing EG concentration, particles nucleated and stabilized at smaller sizes, and then grew by oriented attachment depending on the limits imposed by the presence (dissolution inhibition) and the motion (increased viscosity) of the free ions in the solution [34].

This study presents a road-map to limit and control extend of vaterite/aragonite-to-calcite transformation at lower temperatures without using additives, surfactants or complex synthesis procedures as reported in literature. According to the results, EG concentration and temperature of precursor solutions may be used as experimental variables for precisely controlling polymorph, morphology and size of synthesized $CaCO_3$ particles to maximize the efficacy of these particles in biomedical applications.

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REFERENCES

[1] M. Ni and B. D. Ratner, “Differentiating calcium carbonate polymorphs by surface analysis techniques - An XPS and TOF-SIMS study,” *Surf. Interface Anal.*, vol. 40, no. 10, pp. 1356–1361, 2008.

[2] T. Schüler and W. Tremel, “Versatile wet-chemical synthesis of non-agglomerated CaCO3 vaterite nanoparticles,” *Chem. Commun.*, vol. 47, no. 18, pp. 5208–5210, 2011.

[3] A. C. Tas, “Aragonite coating solutions (ACS) based on artificial seawater,” *Appl. Surf. Sci.*, vol. 330, pp. 262–269, 2015.

[4] J. R. Lakkakula, R. Kurapati, I. Tynga, H.

Abrahamse, A. M. Raichur, and R. W. Maçedo Krause, “Cyclodextrin grafted calcium carbonate vaterite particles: efficient system for tailored release of hydrophobic anticancer or hormone drugs,” *RSC Adv.*, vol. 6, no. 106, pp. 104537–104548, 2016.

[5] K. Sariibrahimoglu, S. C. G. Leeuwenburgh, J. G. C. Wolke, L. Yubao, and J. A. Jansen, “Effect of calcium carbonate on hardening, physicochemical properties, and in vitro degradation of injectable calcium phosphate cements,” *J. Biomed. Mater. Res. Part A*, vol. 100A, no. 3, pp. 712–719, 2012.

[6] D. B. Trushina, T. V Bukreeva, M. V Kovalchuk, and M. N. Antipina, “CaCO3 vaterite microparticles for biomedical and personal care applications,” *Mater. Sci. Eng. C*, vol. 45, pp. 644–658, 2014.

[7] Y. Boyjoo, V. K. Pareek, and J. Liu, “Synthesis of micro and nano-sized calcium carbonate particles and their applications,” *J. Mater. Chem. A*, vol. 2, no. 35, pp. 14270–14288, 2014.

[8] S. Maleki Dizaj, M. Barzegar-Jalali, M. H. Zarrintan, K. Adibkia, and F. Lotfipour, “Calcium carbonate nanoparticles as cancer drug delivery system,” *Expert Opin. Drug Deliv.*, vol. 12, no. 10, pp. 1649–1660, 2015.

[9] A. Wang, Y. Yang, X. Zhang, X. Liu, W. Cui, and J. Li, “Gelatin-Assisted Synthesis of Vaterite Nanoparticles with Higher Surface Area and Porosity as Anticancer Drug Containers in Vitro,” *Chempluschem*, vol. 81, no. 2, pp. 194–201, 2016.

[10] N. Qiu *et al.*, “Calcium carbonate microspheres as carriers for the anticancer drug camptothecin,” *Mater. Sci. Eng. C*, vol. 32, no. 8, pp. 2634–2640, 2012.

[11] A. Shafiu Kamba, M. Ismail, T. A. Tengku Ibrahim, and Z. A. B. Zakaria, “A pH-sensitive, biobased calcium carbonate aragonite nanocrystal as a novel anticancer delivery system,” *Biomed Res. Int.*, vol. 2013, 2013.

[12] L. Saidykhan, M. Z. B. A. Bakar, Y. Rukayadi, A. U. Kura, and S. Y. Latifah, “Development of nanoantibiotic delivery system using cockle shell-derived aragonite nanoparticles for treatment of osteomyelitis,” *Int. J. Nanomedicine*, vol. 11, pp. 661–673, 2016.

[13] Y. Wang, Y. X. Moo, C. Chen, P. Gunawan, and R. Xu, “Fast precipitation of uniform CaCO3 nanospheres and their transformation to hollow hydroxyapatite nanospheres,” *J. Colloid Interface Sci.*, vol. 352, no. 2, pp. 393–400, 2010.

[14] A. Sarkar and S. Mahapatra, “Synthesis of All Crystalline Phases of Anhydrous Calcium Carbonate,” *Cryst. Growth Des.*, vol. 10, no. 5, pp. 2129–2135, 2010.

[15] I. Udrea *et al.*, “Vaterite synthesis via gas-liquid route under controlled pH conditions,” *Ind. Eng. Chem. Res.*, vol. 51, no. 24, pp. 8185–8193, 2012.

[16] Z. Chen and Z. Nan, “Controlling the polymorph and morphology of CaCO3 crystals using surfactant mixtures,” *J. Colloid Interface Sci.*, vol. 358, no. 2, pp. 416–422, 2011.

[17] G. Yan, L. Wang, and J. Huang, “The crystallization behavior of calcium carbonate in ethanol / water solution containing mixed nonionic / anionic surfactants,” *Powder Technol.*, vol. 192, no. 1, pp. 58–64, 2009.

[18] J. Chen and L. Xiang, “Controllable synthesis of calcium carbonate polymorphs at different temperatures,” *Powder Technol.*, vol. 189, no. 1, pp. 64–69, 2009.

[19] R.-J. Qi and Y.-J. Zhu, “Microwave-assisted synthesis of calcium carbonate (vaterite) of various morphologies in water-ethylene glycol mixed solvents.,” *J. Phys. Chem. B*, vol. 110, no. 16, pp. 8302–8306, 2006.

[20] D. B. Trushina, T. V. Bukreeva, and M. N. Antipina, “Size-Controlled Synthesis of

Vaterite Calcium Carbonate by the Mixing Method: Aiming for Nanosized Particles," *Cryst. Growth Des.*, vol. 16, no. 3, pp. 1311–1319, 2016.

[21] F. Chen, Y.-J. Zhu, K.-W. Wang, and K.-L. Zhao, "Surfactant-free solvothermal synthesis of hydroxyapatite nanowire/nanotube ordered arrays with biomimetic structures," *CrystEngComm*, vol. 13, no. 6, pp. 1858–1863, 2011.

[22] H. Cölfen and M. Antonietti, "Crystal Design of Calcium Carbonate Microparticles Using Double-Hydrophilic Block Copolymers," *Langmuir*, vol. 14, no. 3, pp. 582–589, 1998.

[23] E. M. Flaten, M. Seiersten, and J. P. Andreassen, "Induction time studies of calcium carbonate in ethylene glycol and water," *Chem. Eng. Res. Des.*, vol. 88, no. 12, pp. 1659–1668, 2010.

[24] B. H. Toby and R. B. Von Dreele, "GSAS-II: The genesis of a modern open-source all purpose crystallography software package," *J. Appl. Crystallogr.*, vol. 46, no. 2, pp. 544–549, 2013.

[25] J. A. Juhasz-Bortuzzo, B. Myszka, R. Silva, and A. R. Boccaccini, "Sonosynthesis of Vaterite-Type Calcium Carbonate," *Cryst. Growth Des.*, vol. 17, no. 5, pp. 2351–2356, 2017.

[26] G.-T. Zhou, Q.-Z. Yao, S.-Q. Fu, and Y.-B. Guan, "Controlled crystallization of unstable vaterite with distinct morphologies and their polymorphic transition to stable calcite," *Eur. J. Mineral.*, vol. 22, no. 2, pp. 259–269, 2010.

[27] R. M. Santos, P. Ceulemans, and T. Van Gerven, "Synthesis of pure aragonite by sonochemical mineral carbonation," *Chem. Eng. Res. Des.*, vol. 90, no. 6, pp. 715–725, 2012.

[28] E. M. Flaten, M. Seiersten, and J. P. Andreassen, "Polymorphism and morphology of calcium carbonate precipitated in mixed solvents of ethylene glycol and water," *J. Cryst. Growth*, vol. 311, no. 13, pp. 3533–3538, 2009.

[29] Q. Li, Y. Ding, F. Li, B. Xie, and Y. Qian, "Solvothermal growth of vaterite in the presence of ethylene glycol, 1,2-propanediol and glycerin," *J. Cryst. Growth*, vol. 236, no. 1–3, pp. 357–362, 2002.

[30] Y. Chen, X. Ji, and X. Wang, "Microwave-assisted synthesis of spheroidal vaterite CaCO3in ethylene glycol-water mixed solvents without surfactants," *J. Cryst. Growth*, vol. 312, no. 21, pp. 3191–3197, 2010.

[31] R. Ševčík, M. Pérez-Estébanez, A. Viani, P. Šašek, and P. Mácová, "Characterization of vaterite synthesized at various temperatures and stirring velocities without use of additives," *Powder Technol.*, vol. 284, pp. 265–271, 2015.

[32] D. Chakrabarty and S. Mahapatra, "Aragonite crystals with unconventional morphologies," *J. Mater. Chem.*, vol. 9, no. 11, pp. 2953–2957, 1999.

[33] Y. I. Svenskaya *et al.*, "Key Parameters for Size- and Shape-Controlled Synthesis of Vaterite Particles," *Cryst. Growth Des.*, vol. 18, no. 1, pp. 331–337, 2018.

[34] J. Andreassen, "Formation mechanism and morphology in precipitation of vaterite — nano-aggregation or crystal growth ?," *J. Cryst. Growth*, vol. 274, pp. 256–264, 2005.

[35] B. V. Parakhonskiy, A. Haase, and R. Antolini, "Sub-micrometer vaterite containers: Synthesis, substance loading, and release," *Angew. Chemie - Int. Ed.*, vol. 51, no. 5, pp. 1195–1197, 2012.

[36] R. Beck and J. P. Andreassen, "The onset of spherulitic growth in crystallization of calcium carbonate," *J. Cryst. Growth*, vol. 312, no. 15, pp. 2226–2238, 2010.