Formation of Lithium Phthalocyanine Nanotubes by Size Reduction Using Low- and High-Frequency Ultrasound

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High-power ultrasound is commonly used at relatively low frequencies (e.g., 20 kHz) to disperse and modify micrometer and nanosized particles in liquids. However, our a priori hypothesis was that relatively high frequency ultrasound is capable of modifying solid particles in aqueous solutions, because cavitation bubbles have a smaller resonance size at high frequency and would be more likely to violently collapse on the interface of submicrometer-sized particles. A combination of low-frequency (22.5 kHz) followed by high-frequency (70, 354, 803, and 1024 kHz) ultrasound was used to disperse and miniaturize a microcrystalline powder of lithium phthalocyanine (LiPc), an electron paramagnetic resonance oxygen-sensitive probe, in aqueous solution. In the absence of a stabilizing agent, high-frequency sonolysis produced nanosized particles that tended to agglomerate into clusters that were larger in size than the original particles. Furthermore, all of the particles sonicated exhibited some degree of sonochemical degradation, as evidenced by color changes of the sonicated solutions. The addition of sodium dodecyl sulfate (SDS) prior to high-frequency sonolysis of LiPc suspensions had a profound effect on stabilizing individual particles in solution, thereby creating relatively monodispersed, nanosized particles in water. These particles retained their EPR activity; however, unlike the micrometer-sized LiPc particles, the nanosized LiPc particles were almost insensitive to oxygen. High-frequency ultrasound creates interesting modifications to the LiPc particles, resulting in extremely thin, rod-shaped nanotubes that are not observed following high-power, low-frequency ultrasound exposure.

Introduction

Microcrystalline particles of lithium phthalocyanine (LiPc)[1] have been shown to be useful for in vivo applications of electron paramagnetic resonance (EPR) oximetry in brain[2], tumor,[3] and heart[4] tissues. Although all three known forms (α, β, and x) of crystalline LiPc are neutral radicals and therefore paramagnetic, only the x form exhibits sensitivity to the partial pressure of oxygen (pO_2). The EPR spectrum of the x form of LiPc under anoxic conditions consists of a single, sharp line (peak-to-peak line width, LW = 0.03 G)[5] which broadens as the partial pressure of oxygen is increased. Broadening of the EPR line of LiPc by oxygen is believed to arise from the Heisenberg spin exchange between the neutral radical and molecular oxygen, resulting in shortening of the spin−spin relaxation time (T_2).[6,7]

Current synthetic methods result in LiPc crystals with particle sizes on the order of micrometers.[8] The excellent stability and paramagnetic properties of these particles makes them good candidates for internalization into cells and tissues for studies involving the visualization of cell proliferation and/or migration.[9,10] The current method of internalization includes treatment of the raw LiPc particles by 22.5 kHz ultrasound to disperse them in aqueous solution, resulting in particle size distributions with average particle diameters of as low as 200 nm. Decreasing the size of the particles even further would improve their internalization into cells. Furthermore, nanosized LiPc particles could be conjugated with specific antibodies, etc., to allow them to reach a specific site following vascular infusion.

The chemical and physical effects of high-power ultrasound are attributed to the phenomenon of acoustic cavitation.[11,12] Depending on the conditions of sonolysis of water, microbubbles of varying sizes are formed and may grow to a resonance size, at which point the sudden growth and inertially driven, almost adiabatic collapse of bubbles occurs

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to create high-temperature and -pressure regions in water known as "hot spots".[13] These nanosized hot spots are the source of sonochemistry[14] and sonoluminescence.[15] Furthermore, the growth and collapse of bubbles can create shear stress on an interface in close vicinity to the bubble, whereas the symmetrical collapse of a free bubble in solution can lead to the formation of a shock wave.[16] Bubbles that collapse on a solid surface do so asymmetrically, resulting in the formation of a liquid jet that passes through the bubble with a force so powerful that it can erode the solid surface.[16]

Given this, it is not surprising that high-power ultrasound has been shown to degrade solid polymers,[17−19] damage carbon nanotubes[20] and MoO_3 particles,[21] destroy nanoclay clusters,[22] and accelerate particles through solution so rapidly that metal particles have been shown to fuse following ultrasound exposure in a liquid.[23] However, the majority of studies in the literature have focused attention on the sonomechanical effects of low-frequency ultrasound on the physical degradation of solid materials (see above). The reason for this is that it is known that low-frequency, high-power ultrasound creates greater physical effects throughout the bulk liquid being irradiated.[24] This results in efficient mixing of the liquid and dispersion of solid particles. However, we hypothesized that because the resonance radius of the bubble is inversely proportional to the ultrasound frequency,[24] employing a higher frequency of power ultrasound (following the low-frequency treatment) would create smaller bubbles that would be more likely to collapse asymmetrically on the surface of submicrometer-sized particles and more efficiently create cracks in the carbon crystal. Low-frequency ultrasound creates bubbles that may be too large to asymmetrically collapse on the interface of submicrometer-sized LiPc particles. Therefore, physical destruction of particles would occur primarily through shear stress and shock wave effects brought on by the motion and symmetrical collapse of free bubbles in the liquid and would not be as efficient at physically degrading submicrometer-sized particles, compared to the asymmetrical collapse of bubbles on the surface of such particles.

The current study investigates the effect of high-power ultrasound on the miniaturization of micrometer-sized lithiated phthalocyanine particles. The effect of SDS on stabilizing the particles formed to prevent their agglomeration is also investigated.

Materials and Methods

Chemicals. The particles used in the current study (i.e., microcrystalline lithium phthalocyanine, LiPc) were synthesized according to the method of Ilangovan et al.[8] A full description of the physical nature of these particles has been characterized by a number of studies.[4,6,10] Micrometer-sized, solid crystals can be readily dispersed in water by 22.5 kHz ultrasound. The initial concentration of LiPc particles used in all experiments was 5 mg/mL. Sodium dodecyl sulfate (SDS, >99%) was supplied by Fluka. All solutions were made using Milli-Q filtered water (conductivity $< 1 \times 10^{-6}$ S cm^{-1}).

EPR Measurements. EPR measurements were performed using a Bruker ER-300 spectrometer with a TM_{110} cavity operating at X-band (9.78 GHz). The spectral acquisitions were carried out using software developed in house. Unless mentioned otherwise, the EPR line widths (LW) reported are peak-to-peak widths of the first derivative spectra. The EPR line width versus partial pressure of oxygen was recorded from X-band EPR measurements on LiPc equilibrated with an oxygen/nitrogen gas mixture, as reported previously.[25]

Sonication Procedure. Prior to sonication, the microcrystals of LiPc were ground into a fine powder. LiPc powder (15 mg) was suspended in 3 mL of water either in the presence or absence of SDS. All samples were then sonicated at 22.5 kHz using a horn with a tip diameter of 3 mm (Sonic Dismembrator model 100, supplied by Fisher Scientific). The ultrasound power was set to a value of 2 (out of a possible value of 9) on the generator. In the absence of temperature control, sonolysis of water (3 mL) for 30 s under these conditions resulted in a temperature rise of 6.5 °C. Therefore, the calorimetrically determined ultrasonic intensity using this power setting was calculated[26] to be 4 W cm^{-2}. LiPc particles were placed in a glass vial, to which 3 mL of water was added. Sonolysis was conduced for 5 min, creating a "homogeneous" dispersion of particles in water. The temperature rise in the sample during sonolysis in a surrounding ice bath was negligible.

Following sonolysis at 22.5 kHz, the resulting suspensions of LiPc particles were filtered to remove particles larger than 450 nm in diameter, because it was of interest to study the effect of high-frequency ultrasound on submicrometer-sized LiPc particles. These samples were then treated at ultrasound frequencies of 1 MHz, 803, 354, or 70 kHz using a flat plate transducer, as described elsewhere.[27,28] The flat plate transducers and power generator were supplied by ELAC−Nautik, GmbH, and their type and model numbers were 1 MHz, USW 51-52; 803 kHz, USW 51-104; 354 kHz, USW 51-052; and 70 kHz, USW 51-106. The generator (CESAR RF power generator) was set to an electrical power output of 60 W.

This experimental setup has been previously characterized and described.[27] In essence, 1 mL samples of microparticle solutions treated using the 22.5 kHz system were filtered to remove particles above a size of 0.45 μm and placed in an exposure vessel (Kimble borosilicate glass culture tubes, with a diameter and length of 13 mm × 100 mm) that was placed reproducibly in the center of a jacketed bath of water (300 mL) in contact with the stainless steel flat plate. The exposure vessel was positioned in such a way that the gas/solution interface of the sample was approximately level with the gas/solution interface of the bath water. In this way, the position of the sample tube could be reproduced from run to run,

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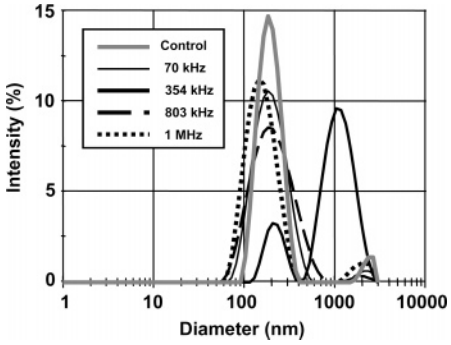

Figure 1. Control; LiPc particle size distribution (initial concentration = 5 mg/mL) following sonolysis (22.5 kHz, 5 min, $I = 4$ W cm^{-2}). The control sample was filtered above 450 nm and diluted 10 times with water. LiPc, pretreated with 22.5 kHz irradiated (i.e., control samples) irradiated with one of four higher frequencies of ultrasound: 1 MHz, 803 kHz, 354 kHz, or 70 kHz. High-frequency ultrasound exposure conditions were sonolysis time = 60 min, $p = 60$ W, air-exposed solutions.

and we also found that a greater amount of sonochemical activity occurred in this region. Once in position, the samples were exposed to ultrasound for a given length of time. The temperature was kept constant in the bath during sonolysis by setting the cooling jacket around the bath to a predetermined temperature so that the bath water remained at a constant temperature of 20 °C.

Particle Size Analyzer. The particle size analyzer was from Malvern Instruments (model Zetasizer, nano-s) with the capability to characterize particle sizes ranging from 0.6 nm to 10 μm. Following sonolysis, the particle suspensions were immediately transferred to a cuvette (Plastibrand, 1.5 mL volume, catalog no. 7591 50), and the particle size distribution was determined.

Transmission Electron Microscopy (TEM). TEM images were acquired using a Philips CM12 microscope operating at 80 kV. Sonicated LiPc particles were adsorbed to a standard Formvar-coated grid by applying a drop of aqueous LiPc suspension to the grid. After 3 min, the drop was carefully adsorbed onto a piece of filter paper and the grid was treated with a drop of uranyl acetate. Two minutes later, the drop of uranyl acetate was adsorbed onto a piece of filter paper. The grid was allowed to dry in air, and the images were then acquired.

Results and Discussion

The particle size distribution for LiPc particles (5 mg/mL) sonicated at 22.5 kHz for 5 min followed by filtration (to remove particles of $d > 450$ nm) and a 10× dilution with water is shown in Figure 1. The particles were filtered and diluted in this way in order to prevent larger or concentrated particle suspensions from influencing the passage of higher-frequency ultrasonic waves, as described below. The resulting particle suspension had a distribution with a maximum number of particles at approximately 190 nm and a size ranging from 100 to 400 nm. Simply mixing 5 mg/mL of LiPc particles with water in the absence of ultrasound and then filtering and diluting them as described above produced no such curve, indicating that the particles formed within this distribution are solely due to the ultrasound-induced miniaturization of larger-sized LiPc particles by 22.5 kHz ultrasound. These suspensions of 22.5 kHz irradiated, filtered, and diluted particles are termed the "control" samples of LiPc throughout the text.

The control LiPc samples were then irradiated for 60 min at one of four different frequencies of ultrasound: 1 MHz, 803 kHz, 354 kHz, or 70 kHz, as shown in Figure 1. The 803 kHz and 70 kHz ultrasound treatments resulted in the formation of a slightly broader size distribution of particles compared to that of the control sample. However, the maximum in the size distribution was approximately equal to the control sample. Ultrasound exposure at a 1 MHz frequency shifted the whole size distribution to slightly smaller values. Given the conditions of sonolysis at higher frequencies (Figure 1), the extent of miniaturization was clearly not an efficient process. Furthermore, when the control sample was treated with 354 kHz ultrasound, a new distribution of particles was formed with a maximum centered at approximately 1 μm and a size distribution range from a minimum particle size of 400 nm through to a maximum particle size of 3 μm.

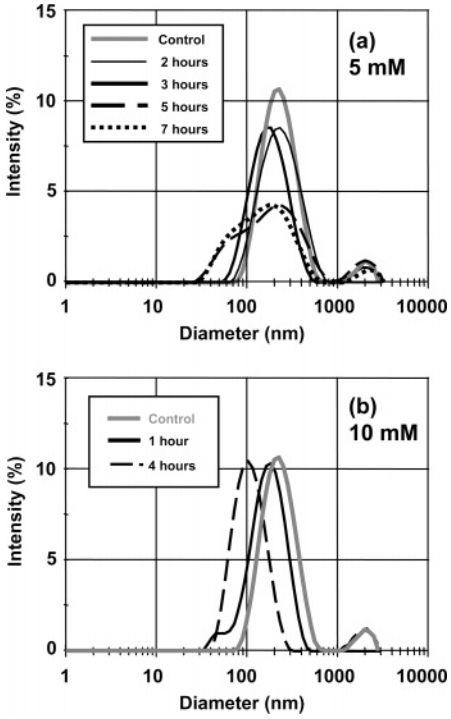

Figure 2. Effect of sonolysis time on the ultrasound-induced miniaturization of LiPc particles in the presence of SDS at concentrations of (a) 5 and (b) 10 mM. Conditions of sonolysis were $f = 354$ kHz, $p = 60$ W, air-exposed.

The formation of larger-sized particles at 354 kHz indicated one of two possibilities. First, ultrasound was forcing particles to agglomerate by kinetically overcoming any repulsive forces between the particles. Second, high-frequency treatment at 803 and 354 kHz and to some extent at 70 kHz (Figure 1) created smaller-sized particles that were thermodynamically unstable and would be more likely to agglomerate to form larger-sized particles.[29] A suitable stabilizing agent can be added to the suspension prior to sonolysis at these higher frequencies in an attempt to prevent agglomeration of particles.

The effect of SDS on preventing ultrasound-induced agglomeration of LiPc particles was studied at an ultrasound frequency of 354 kHz, because the formation of larger-sized LiPc particles in the absence of a dispersant (Figure 1) was most prominent at this frequency, compared to the other

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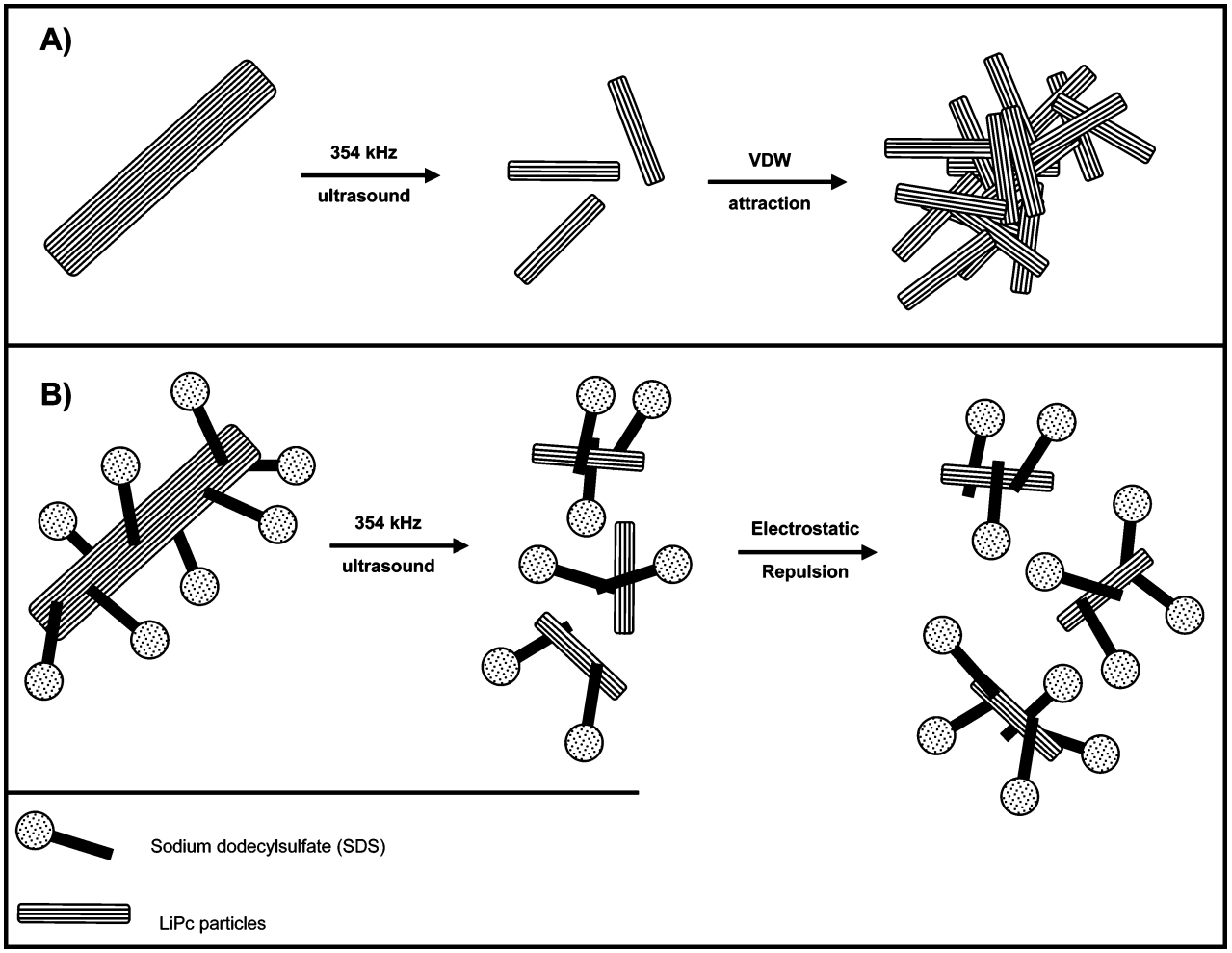

Figure 3. Depiction of the effect of ultrasound on LiPc particles in (A) the presence and (B) absence of SDS during sonolysis. In the absence of SDS, freshly formed smaller particles tend to agglomerate to form larger-sized particles. When sonolysis is conducted in the presence of SDS, the freshly formed smaller particles are stabilized through electrostatic repulsion.

frequencies studied. SDS was chosen as the dispersing agent because it would be expected to have a strong affinity for the hydrocarbon surface of the particles and would therefore result in LiPc particles with a negatively charged interface.

The size distributions of LiPc particles sonicated for various times at a frequency of 354 kHz and in the presence of 5 mM or 10 mM SDS is shown in Figure 2. On this occasion, the maximum in the size distribution of particles in the freshly prepared control suspension (22.5 kHz, 5 min) was approximately 213 nm (control; panels a and b of Figure 2). The distribution of micrometer-sized particles that were observed following sonolysis (354 kHz) of LiPc in the absence of SDS (Figure 1) was not observed following sonolysis in the presence of SDS (5 mM; Figure 2). Furthermore, it is clear that particles of a size less than 100 nm are formed and stabilized in solution following sonolysis (354 kHz) in the presence of SDS (Figure 2). Longer times of sonolysis had a progressively larger effect in shifting the size distribution to smaller values. Therefore, it can be concluded that newly formed, smaller particles can be stabilized by SDS through long-range electrostatic repulsion to prevent their agglomeration, as depicted in Figure 3.

In addition to a smaller particle size, we also observed a progressive color change of the particle suspension during sonolysis from a dull green to a bright blue. From UV−visible absorbance studies, we concluded that the blue color was a soluble byproduct of LiPc sonolysis (Figure 4), most probably aqueous Pc^{2-} ions, because a pure solution of aqueous dilithium phthalocyanine (Li_2Pc) possesses a similar blue color. The UV−visible spectra of sonicated LiPc suspensions and pure, aqueous solutions of Li_2Pc possessed some similarities (Figure 4). We found that the LiPc suspensions following sonolysis had a pH of between 4.0

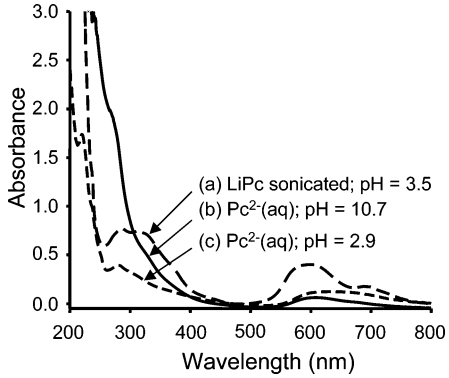

Figure 4. UV−visible spectra of (a) LiPc suspension sonicated in the presence of SDS (5 mM) at 22.5 kHz, 5 min, $I = 4$ W cm^{-2}, followed by sonolysis at 354 kHz, sonolysis time = 60 min, $p = 60$ W, air-exposed solutions; pH 3.5; (b) pure solution of dilithium phthalocyanine salt (Pc^{2-}), pH 10.7; (c) solution of dilithium phthalocyanine salt (Li_2Pc) acidified to pH 2.9 using H_2SO_4.

Table 1. Effect of Ultrasound on the Oxygen Sensitivity of LiPc Particles[a]

	sonolysis under air			sonolysis under argon		
354 kHz sonolysis time (h)	peak to peak LW[b] (G)			peak to peak LW[b] (G)		
	N_2	air	ΔLW (mG)	N_2	air	ΔLW (mG)
0[c]	0.07	1.0	930	0.12[c]	0.99[c]	870
2	0.17	1.25	1080	0.12	0.21	90
3	0.13	0.19	60	0.12	0.19	70
5	0.14	0.22	80	0.13	0.15	20
7	0.14	0.16	20	0.12	0.21	90

[a]Oxygen sensitivity of LiPc particles (initial concentration = 5 mg/mL) dispersed in 10 mL of an aqueous SDS (5 mM) solution by 22.5 kHz ultrasound for 5 min in an atmosphere of air. The 22.5 kHz sonicated samples were then filtered above 0.45 μm to produce the "control" samples (i.e., 0 h in the table). One milliliter of these control samples was then treated at 354 kHz under either air or argon gas. [b] It should be noted that 22.5 kHz sonolysis is conducted only in the presence of air, and not argon gas. The variation in the value of LW for the two control samples that are treated with the 22.5 kHz ultrasound horn only is due to difficulty in positioning of the ultrasonic horn accurately in the sample solution, i.e., the 22.5 kHz ultrasound energy deposited in our control samples cannot be accurately controlled. However, this has no bearing on the value of LW observed during exposure of the control samples to high frequency ultrasound, where positioning of the sample tube can be accurately reproduced. [c] The 0 h time point was always sonicated at 22.5 kHz in the presence of air and not in argon gas.

and 3.4 and acidification of pure Li_2Pc solutions with H_2SO_4 produced a spectrum that more closely resembled that of sonicated LiPc particles (Figure 4). This supports the idea that Pc^{2-} ions leached from the LiPc particles during sonolysis.

Having obtained a method for ultrasonically miniaturizing micrometer-sized LiPc particles, it was of interest to test the EPR sensitivity of particles exposed to air and under anoxic conditions (i.e., 100% N_2) before and after 354 kHz sonolysis, as shown in Table 1. We investigated the effect of the oxygen sensitivity of LiPc particles treated with 354 kHz ultrasound in the presence of either air or argon gas, because the gas type is known to influence sonochemical reactions.[12] For example, sonolysis in the presence of air and SDS (5 mM) would be expected to produce an oxidizing environment, whereas argon and SDS would produce a reductive environment.[30] Therefore, this might give some insight to the effect of sonochemistry on the EPR sensitivity of sonicated LiPc particles.

In Table 1, the sonolysis under air data correspond to the particle size distributions shown in Figure 2a. It is clear that following the treatment of LiPc suspensions for 5 min at 22.5 kHz (Table 1, 0 h; sonolysis under air), the LiPc particles are EPR active and oxygen sensitive, as evidenced by the effect of oxygen on the EPR line width. The same can be said following the subsequent exposure of particles for 2 h with ultrasound at a frequency of 354 kHz (Table 1; 2 h; sonolysis under air). It is interesting to note that the maximum particle size distribution following treatment of LiPc particles for 2 h at 354 kHz (Figure 2a) is similar to that observed following sonolysis for only 5 min at 22.5 kHz (Figure 2a, control). However, sonolysis of LiPc particles at 354 kHz and for more than 2 h results in a smaller particle size distribution (Figure 2a) and a substantial loss of oxygen sensitivity, as evidenced by the very small change in line width when EPR measurements are conducted in either air or 100% N_2 (Table 1; sonolysis under air, ΔLW column). In addition to this, the EPR line became relatively sharp in the presence of air, because of the substantial loss of oxygen sensitivity.

When sonolysis was conducted under an atmosphere of argon gas, a similar trend was observed compared to that of sonolysis in air (Table 1). Again, the LiPc particles had a substantial loss of oxygen sensitivity, and the EPR line became relatively narrow in the presence of air compared to the control sample (Table 1; sonolysis in argon, air column). In an attempt to understand these effects, we studied the effect of ultrasound on LiPc particles using transmission electron microscopy (TEM).

It is known that the x form of LiPc consists of a tetragonal crystal structure in which the one-dimensional stacking of LiPc molecules in a face-to-face configuration occurs.[31] Strong π interaction occurs between the molecules in the stack, whereas relatively weak intermolecular forces exist between molecules in adjacent stack columns in the crystal. The stacking of LiPc molecules in columns in a tetragonal fashion results in the formation of channels that are large enough to allow the free diffusion of oxygen. TEM images of the LiPc particles during exposure to ultrasound in the current study gave interesting insight as to how larger-sized LiPc particles are miniaturized by ultrasound. TEM images of LiPc particles were taken following 22.5 kHz ultrasound exposure for 5 min (images A and B of Figure 5) and subsequent exposure to 354 kHz ultrasound for 3 h in the presence of air (images C and D of Figure 5). Following exposure to 22.5 kHz ultrasound, rod-shaped particles were observed with a length of less than a micrometer and a variable width, generally from about 100 to 50 nm in diameter (images A and B of Figure 5). However, following exposure to 354 kHz ultrasound for 3 h, (images C and D of Figure 5), the original rod-shaped particles began to splice apart parallel to their stack axis to create much thinner rod-shaped particles (from 10 to 25 nm diameter) with a length up to 700 nm (images C and D of Figure 5). These images indicate that the stacked LiPc units in the particles have a relatively powerful attraction within any given stack, but not between neighboring stacks. The observation that 354 kHz ultrasound created long and thin LiPc particles cannot explain the results shown in Table 1 on the basis of current knowledge of why the x form of LiPc possesses a narrow anoxic EPR line.

The very narrow EPR signal of the x-form of LiPc under anoxic conditions (LW = 30 mG) is known to be due to delocalization and/or diffusion of spins along the stack axis of the crystal. Additionally, calculations of the line width anisotropy also suggest that narrowing is attributed to the length of these stacks, assuming that the spin excitations are extremely mobile and delocalized over distances of several micrometers.[31] Therefore, it follows that decreasing the crystal size will result in line-broadening. Indeed, this effect was observed in the current study following 22.5 kHz

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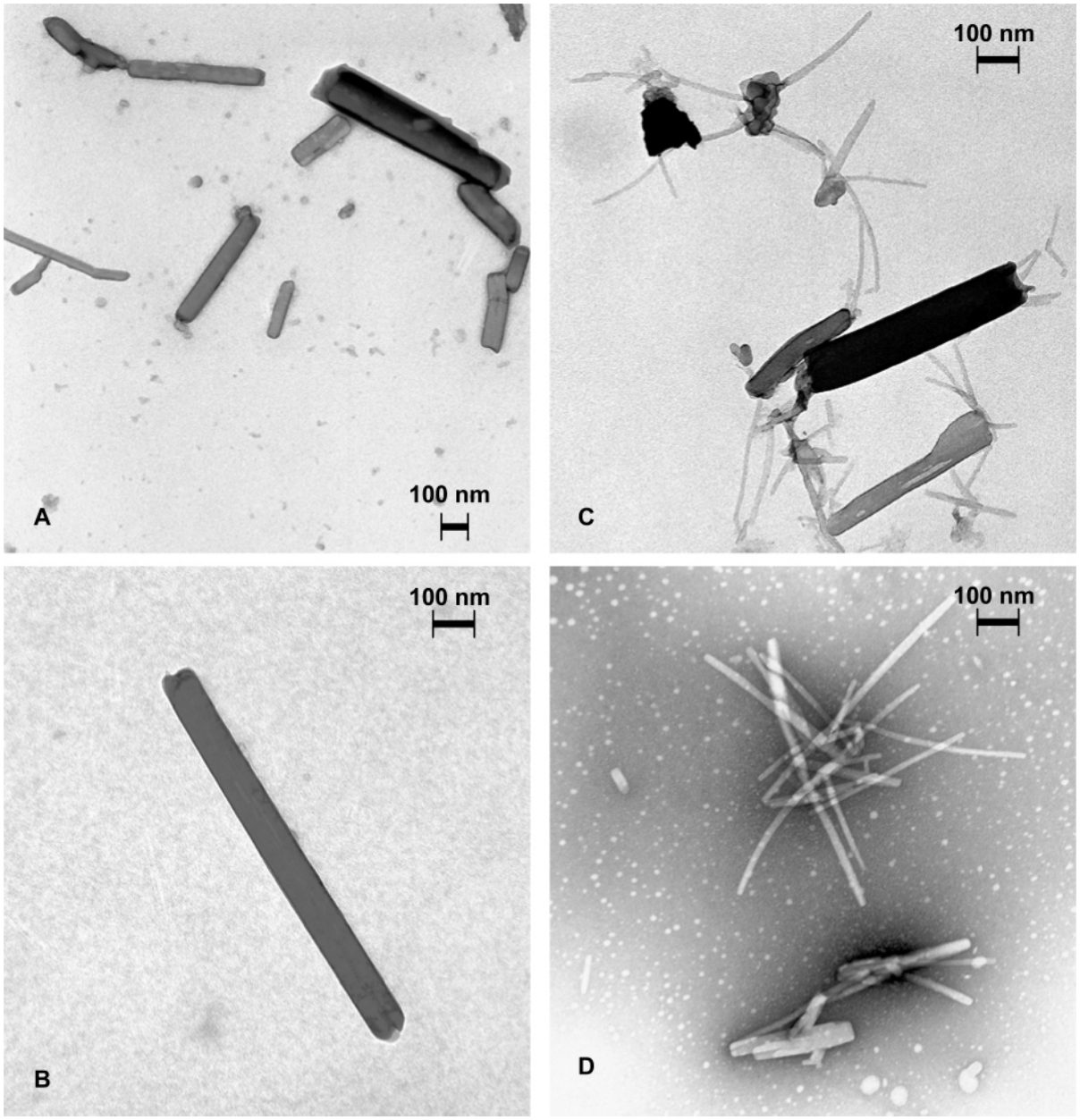

Figure 5. TEM micrographs (magnification: A, 22 000×; B−D, 35 000×) of LiPc particles. (A, B) Control samples (i.e., sonicated at 22.5 kHz for 5 min only); (C, D) Control samples that were then sonicated at 354 kHz for 3 h.

treatment of the raw LiPc particles, because the EPR line width increased from a value of 30 mG for the untreated particles to a value of 70 or 120 mG for the two samples treated at 22.5 kHz only[1] (Table 1; 0 h, N_2 columns). This suggests that 22.5 kHz ultrasound treatment results in a substantial decrease in the length of LiPc particles. However, it is also clear from the data in the current study that increasing times of sonolysis at 354 kHz under argon gas resulted in no change of the anoxic line width (Table 1; sonolysis under argon, N_2 column). This suggests that there is no significant reduction in the average length of LiPc particles during exposure to 354 kHz ultrasound, a conclusion that is supported by the TEM results (Figure 5). The reasons for an increasing anoxic line width for LiPc particles treated with 354 kHz ultrasound in the presence of air (Table 1; sonolysis in air, N_2 column) is not related to the length of the LiPc particles, as will be discussed in more detail below.

The substantial loss of oxygen sensitivity following high-frequency treatment of LiPc particles (Table 1; ΔLW columns) also cannot be explained by current knowledge of LiPc oxygen sensitivity. Oxygen sensitivity for the x form of LiPc has been attributed to channels formed through aligned packing of the tetragonal units that make up the crystal structure[8] of the rod-shaped LiPc particles. Oxygen (size 2.8 × 3.9 Å^2) can diffuse through these channels (diameter = 6 Å) to perturb spin diffusion via the Heisenberg spin exchange.[32] This exchange shortens the relaxation time of the spin probe, causing an increase in the width of the EPR line.[32] Again, it is clear from the TEM images that high-frequency ultrasound resulted in the formation of very thin, rod-shaped particles, or more accurately, nanotubes of a length similar to the 22.5 kHz control treated samples, i.e.,

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the loss of oxygen sensitivity cannot be due to a decreasing length of the nanotubes.

From a sonochemical perspective, there are two other possibilities that could explain a loss of oxygen sensitivity (Table 1; ΔLW columns) and relatively narrow EPR lines observed for air-exposed LiPc particles that had been treated with 354 kHz ultrasound (Table 1; air columns). The sonochemical effects of acoustic cavitation could result in chemical reactions on the surface of the particles, resulting in blockage of the opening of these channels, thereby preventing oxygen from diffusing in and out of the particles. Blockage of the channels on the surface of the particle could hinder the free diffusion of oxygen through the particle and might result in the loss of oxygen sensitivity and the relatively narrow EPR line for air exposed LiPc particles. Similar effects could also be brought about if cavitation effects created new defects in the crystal structure, which prevented oxygen from freely diffusing through these channels. Sonochemical reactions and/or the introduction of defects in the crystal structure may also result in permanent trapping of any oxygen already in the particle when sonolysis is conducted in an atmosphere of air. There is some evidence for this when 354 kHz sonolysis is conducted in the presence of air, because the anoxic line width increases from the original value of 0.07 G for the 22.5 kHz treated control (Table 1; sonolysis under air, N_2 column). As mentioned earlier, when 354 kHz sonolysis is conducted in the presence of argon gas, the anoxic line width does not change (Table 1; sonolysis under argon, N_2 column). Therefore, our data suggest that a relatively small amount of oxygen was trapped inside the particles during sonolysis under air, resulting in a slight increase in the anoxic line width from 0.07 G when 354 kHz sonolysis was conducted in the presence of air. However, almost no oxygen can penetrate the LiPc particles after 354 kHz sonolysis (in either air or argon gas), resulting in a relatively narrow EPR line when the particles are exposed to air (Table 1; sonolysis in air, air column), compared to the control sample (i.e., 0.07 G).

From a mechanistic perspective, it has been shown previously that ultrasound can result in the formation of smaller particle sizes through sonochemical reduction processes.[26,30] However, the mechanical effects of ultrasound have been shown to be powerful enough to result in the physical destruction of particles[33] and polymers.[17−19,34] Therefore, particle size reduction in the current study may be due to either sonochemical and/or sonomechanical effects. It can be concluded that the method of ultrasound-induced miniaturization of micrometer-sized LiPc particles is a promising new technique that may find applications for size reduction of other microcrystalline substances.

Conclusions

The treatment of micrometer-sized LiPc particles with 22.5 kHz ultrasound is sufficient to reduce particles to a maximum size distribution of approximately 200 nm. We have shown that various combinations of high frequency ultrasound and SDS concentrations can be used following 22.5 kHz ultrasound to further reduce the size of these particles to diameters of tens of nanometers and maximum size distributions of as low as 100 nm. In the current study, relatively long sonolysis times up to 4 h were required to create the greatest size-reduction effect during exposure of particles to high-frequency ultrasound. It could be possible to further optimize the particle size effect by finding more suitable dispersing agents and by varying the initial concentration of particles and the ultrasound exposures conditions (gas type, frequency, intensity, exposure geometry and temperature, solution viscosity, and so on). We have not considered all of these variables in our system, but it is expected that the results of the current study should create interest in optimizing ultrasound-induced particle size reduction for other systems. The ultrasound miniaturization technique creates LiPc nanotubes that effectively lose their oxygen sensitivity. Hence, this technique would be of interest for producing nanosized and EPR-active particles for cell tagging or vascular infusion as an imaging agent, because the particles would not be substantially affected by oxygen concentration.

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