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Hydrolytic Degradation of Poly(ethylene oxide)-*block*-Polycaprolactone Worm Micelles

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Degradable polymers are foundational to a number of fields from environmental chemistry to biomedical devices.[1] While degradable homopolymers and random copolymers are commonly used in bulk materials, micro/nanoparticles, and films/ monolayers,[2] degradable self-assemblies of block copolymer amphiphiles are also emerging.[3,4] Attention has thus far been limited to spherical micelles assembled from copolymers of hydrophobic degradable polyesters, typically polylactide or polycaprolactone, plus a hydrophilic, biocompatible block such as poly(ethylene oxide).[4] However, degradation has subtle if detectable effects on spherical morphologies, and degradation mechanisms and kinetics in such assemblies are not clearly distinguished in time scales or pathway(s) from degradation in bulk or film preparations.[4,5] Here, we report novel giant and flexible worm micelles prepared from degradable poly(ethylene oxide)-*b*-poly(ϵ-caprolactone) copolymers (PEO−PCL, denoted OCL). The OCL worm micelles spontaneously shorten to generate spherical micelles due, we show, to chain-end hydrolysis of the PCL. Kinetics as well as mechanism are elucidated via Arrhenius fits to key activating conditions of temperature, pH, and copolymer molecular weight, providing novel insight into this microphase transition.

The dominant morphology of amphiphilic copolymer aggregates in water is generally dictated by average block proportions.[6] Giant and flexible worm micelles were prepared from two OCL copolymers with weight fractions of PEO ($f_{EO} = 0.42$) that favor worm micelle formation,[6] OCL1 ($M_n = 4770$) and OCL3 ($M_n = 11\,500$), using a cosolvent/evaporation method (see Supporting Information). Fluorescence microscopy (FM) was used to then visualize dye-labeled worm micelles (Figure 1) and track how the contour lengths change with time.[7] The mean contour length (L) of freshly made OCL worm micelles is more than 10 μm (Figure S1), and their flexibility, expressed as the persistence length, l_P, is 0.5 μm for OCL1 micelles and 5 μm for the larger diameter OCL3 micelles (Figure S2). Core diameters of $d = 11$ and 29 nm, respectively, were measured from cryo-TEM images.[8] Values of l_p and d prove to be very similar to those for PEO−polybutadiene worm micelles of similar copolymer M_n and, likewise, fit well to the scaling relation $l_p \sim d^{2.8}$ that indicates a fluid rather than glassy or crystalline aggregate.[7]

On time scales of days, these giant OCL worm micelles shorten spontaneously to spherical micelles, as seen in FM and cryo-TEM (Figure 1), as well as DLS (not shown). The predominant new species generated was found by GPC to be 6-hydroxycaproic acid (6-HPA), that is, the monomer product of PCL hydrolysis (Figure 2a). For both block copolymers, accumulation of 6-HPA parallels in form and time scales with the decays in mean contour length L of OCL worm micelles (Figure 2b). No other significant degradation products were detected, and the polydispersity of the OCL copolymer remained essentially the same (Figure S3). Loss of caprolactone units from the OCL copolymer was confirmed by

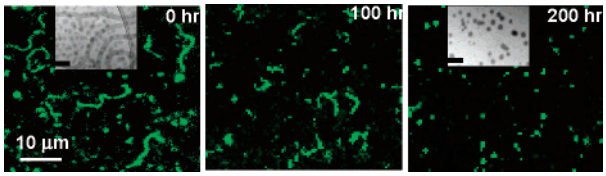

Figure 1. OCL worm micelles spontaneously shorten with time to spherical micelles in water, visualized by FM and cryo-TEM (inset, scale = 100 nm). Shown are 0.2 mg/mL OCL3 worm micelles at 37 °C.

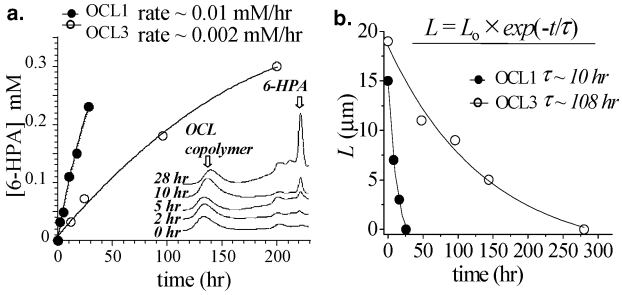

Figure 2. (a) Cumulative production of PCL hydrolysis monomer, 6-hydroxycaproic acid, 6-HPA (inset: GPC chromatograms) with (b) the decay of OCL worm micelle mean contour length L (37 °C, pH 5 buffer).

NMR (Figure S4). The analytical results thus demonstrate that PCL in these copolymers hydrolyzes from the end by "chain-end cleavage" rather than by a process of "random scission"[9] that would yield various degradation products and broaden the polydispersity of the polymer far more than found here.

End hydrolysis of PCL increases f_{EO} and consequently shifts the preferred morphology toward a higher curvature structure, namely, from a cylinder to a sphere.[6] By the time worms have disappeared, PCL chains have, on average, lost ~30% of their length by hydrolysis (Figure 2), which corresponds to increases in f_{EO} from 0.42 to 0.55. Such slight asymmetry, with f_{EO} above 0.5, favors spherical micelle formation.[6] This simple estimation highlights the reason worm micelles are so susceptible to morphological transformation: only an extremely narrow range of f_{EO} favors the worm micelle structure, whereas spherical micelles are found with a much broader range of f_{EO} and are thus less sensitive to hydrolysis.[6]

The worm-to-sphere transition occurs with bulb formation at the end of the worm, consistent with release of spherical micelles from the end[10] (Figure S5). Conservation of mass allows one to show that the hydrolysis kinetics is the rate-limiting step in worm shortening kinetics. The amount of monomer generated initially from OCL1 and OCL3 worm micelles, ~0.01 and 0.002 mM/h, respectively (Figure 2a), gives the volume of spherical micelles generated from the worm micelles, based on the above changes in f_{EO} and PCL's volume density.[11] The estimations yield respective shortening rates of ~1.0 and 0.1 μm/h, as observed in FM (Figure 2b). Such estimations apply equally well to the two copolymers

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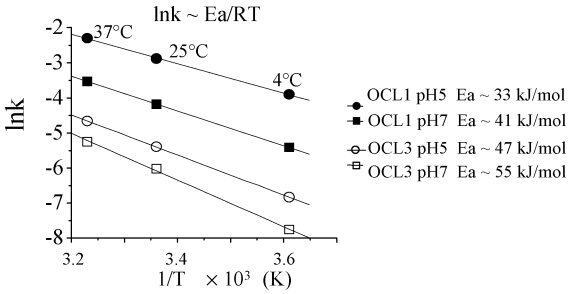

Figure 3. Arrhenius plots of OCL worm micelle shortening rate constants, *k*, with temperature (4, 25, and 37 °C), $R = 8.314$ kJ/mol.

that differ in molecular weight and thus differ in molecular mobility within worms by far more than 2-fold.[12] This suggests that the rate-limiting process is indeed hydrolysis rather than chain diffusion and segregation post-hydrolysis.

While the end-cleavage of PCL within worm micelles appears to be consistent with both the chemical and the nanoscale physical changes, it is also considerably faster than the slow hydrolysis reported for PCL homo/copolymer bulk, particle, or films,[2] that is, on the time scale of months−years under the same condition. The distinction arises with the specific effect of OCL worm micelles on PCL hydrolysis. As speculated from studies on spherical micelles,[13] the terminal −OH of the hydrophobic PCL block is not strictly sequestered in the "dry", hydrophobic core but will tend to be drawn into the hydrated corona. A "micellar catalysis" effect[14] involving interfacial water plus this likely participation of the terminal hydroxyl group[15] collectively fosters the attack by H_2O of the end-ester group nearest the chain terminus. Following this ester hydrolysis, a new −OH is generated to restart the process of PCL end-degradation. To provide direct evidence for the crucial role of the terminal −OH, −OH was modified in OCL1 to an acetate group by esterification. Worm micelles still formed with OCL1−acetate, but they showed no significant morphological change after more than 24 h at 37 °C (Figure S6), by which time OCL1 worm micelles are completely degraded (Figure 2b).

For both OCL1 and OCL3 worm micelles, shortening rate constants measured from FM (Table S1) increase exponentially with temperature, with minimal degradation at 4 °C, but considerable hydrolysis at the physiological temperature of 37 °C. The temperature dependences fit classic Arrhenius behavior and yield activation energies, E_a, for the morphological transformations (Figure 3). Consistent with acid-catalyzed ester hydrolysis, acidic pH 5 (physiological HEPES buffer) enhances the shortening rate by 2−4-fold systematically and also lowers E_a by 7−8 kJ/mol, compared to that of neutral pH 7 (PBS buffer). At either pH, the higher M_n OCL3 decreases the shortening rate by 3−4-fold and raises E_a by 10 kJ/mol compared to that of OCL1. This higher E_a is consistent with a larger entropic penalty for an activated reptation,[12] that is, entanglement release, of the terminal hydroxyl group of the longer OCL3 chain to the micellar interface. Moreover, the values for E_a (33−55 kJ/mol) of OCL worm micelle shortening are in good agreement with E_a of homogeneous hydrolysis of water-soluble polyester oligomers reported in the literature.[16] This adds to the proof that PCL hydrolysis is the driving force for worm micelle shortening, and that such hydrolysis is surprisingly homogeneous rather than heterogeneous and limited—as seen in polyester degradation of bulk and particles—by the infiltration of water.

Degradable OCL worm micelles with such unique degradation mechanism and kinetics are potentially useful for numerous applications, in particular, for drug delivery. While polymeric spherical micelles have already proven to be extremely useful for therapeutic applications,[17] worm micelles are just now emerging as novel alternatives that provide larger core volume for drug loading and an ability to flow readily through capillaries and pores due to their cylindrical shape and flexibility.[18] One novel strategy for drug delivery would be to start with worm micelles and then progressively degrade into spherical micelles as desired. Furthermore, strong effect of temperature, pH, and M_n on degradation rate could also be used for controlled drug release.[19]

In summary, we show that worm micelles self-assemble from degradable PEO-*b*-PCL block copolymers and spontaneously shorten to spherical micelles. Such morphological transition is triggered by hydrolytic degradation of PCL, governed by an end-cleavage mechanism that is faster than that in bulk/film. Degradation rate can be tuned by temperature, pH, and M_n, and quantitative assessment appears to be consistent with the molecular explanation, whereby the hydroxyl end of the PCL chain localizes to the hydrated interface of the micelle.

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Supporting Information Available: Materials and Methods, OCL worm micelle contour length distribution and flexibility, GPC, NMR, transition intermediate, OCL−acetate worm micelles, and data of shortening rate constants (PDF). This material is available free of charge via the Internet at http://pubs.acs.org.

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