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Employing Bicontinuous-to-Micellar Transitions in Nanostructure Morphology for On-Demand Photo-Oxidation Responsive Cytosolic Delivery and off-on Cytotoxicity

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COMMUNICATION

Employing Bicontinuous-to-Micellar Transitions in Nanostructure Morphology for On-Demand Photo-Oxidation Responsive Cytosolic Delivery and off-on Cytotoxicity

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Bicontinuous nanospheres (BCNs) are underutilized self-assembled nanostructures capable of simultaneous delivery of both hydrophilic and hydrophobic payloads. Here, we demonstrate that BCNs assembled from poly(ethylene glycol)-block-poly(propylene sulfide) (PEG-*b*-PPS), an oxidation-sensitive copolymer, are stably retained within cell lysosomes following endocytosis, resisting degradation and payload release for days until externally triggered. The oxygen scavenging properties and enhanced stability of the bicontinuous PEG-*b*-PPS nanoarchitecture significantly protected cells from typically cytotoxic application of pro-apoptotic photo-oxidizer pheophorbide A and chemotherapeutic camptothecin. The photo-oxidation triggered transition from a bicontinuous to micellar morphology overcame this stability, allowing on-demand cytosolic delivery of camptothecin for enhanced control over off-on cytotoxicity. These results indicate that inducible transitions in the nanostructure morphology can influence intracellular stability and toxicity of self-assembled nanotherapeutics.

Introduction

A key rationale for specifying the nanostructure of drug delivery vehicles is to beneficially alter the pharmacodynamics and biodistribution of loaded cargo molecules.¹ By altering the size and morphology, the employed nanostructure may increase on-target delivery and reduce off-target uptake of their bioactive payloads.² Despite these well-documented benefits,

nanostructure-mediated delivery still results in non-specific cellular uptake, particularly by cells of the mononuclear phagocyte system in organs such as the liver and spleen.³ This presents a particular challenge when potent cytotoxic cargoes are delivered. Stimuli-responsive nanostructures may decrease such non-specific cytotoxicity by providing spatiotemporal control over cargo release.⁴

One difficulty in using stimuli-responsive nanostructures is that they are often fabricated via the self-assembly of block copolymers, resulting in sensitive and easily ruptured vehicles. In particular, nanocarriers with aqueous lumens, which are required for facile encapsulation and transport of both hydrophilic and hydrophobic payloads simultaneously, are quickly disrupted under the harsh conditions of intracellular compartments.⁵ For example, despite their known enhanced stability relative to liposomes during *in vivo* transport, polymersomes are equally unlikely to survive intact within the lysosome for more than a few hours regardless of their chemistry. Here, we utilize a highly stable self-assembled polymeric nanocarrier morphology known as a bicontinuous nanosphere (BCN), which is characterized by a cubic lattice of aqueous channels that traverse its hydrophobic interior volume.⁶ BCNs are the polymeric equivalent of lipid cubosomes, and their organized, lyotropic and interconnected internal architecture make them exceptionally robust and stable assemblies, relative to polymersomes, when loading high concentrations of molecular cargo⁷.

BCNs self-assembled from poly(ethylene glycol)-*block*-poly(propylene sulphide) (PEG-*b*-PPS) are capable of loading both hydrophilic and hydrophobic compounds simultaneously, making them a versatile drug delivery vehicle.^{7,8} PEG-*b*-PPS nanocarriers of varying morphology successfully deliver cargo *in vitro* and *in vivo* with a demonstrated lack of toxicity in both murine and non-human primate models.^{7,9-13} The hydrophobic PPS block of the polymer can be oxidized into sulfoxide and sulfone derivatives under physiological conditions, leading to an increase in the hydrophilicity of the block and resulting disassembly of PEG-*b*-PPS nanostructures, including BCN

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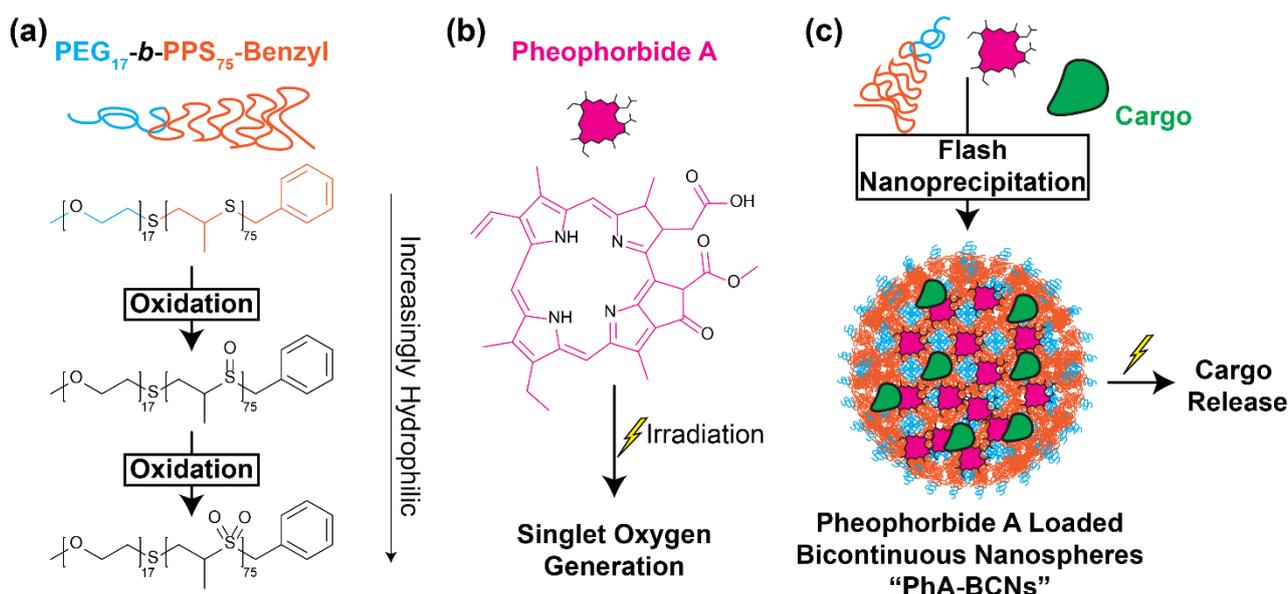
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Electronic Supplementary Information (ESI) available: Experimental details, Figures S1: additional confocal images, S2: low magnification TEM micrograph, S3: Change in turbidity of BCNs upon irradiation, S4: Singlet oxygen generation at high power, S5: Photo-oxidation induced release of hydrophilic cargo, S6: ROS generation upon irradiation of cells, S7: low magnification TEM micrograph of irradiated supernatant, S8: Micelle diameter distribution from TEM micrographs, S9: Confocal images of Pha FITC-BSA BCNs co-localized with lysotracker. See DOI: 10.1039/x0xx00000x



Scheme 1. Formation of PhA-loaded BCNs (PhA-BCNs) from oxidation-sensitive PEG-*b*-PPS polymer. (a) PEG-*b*-PPS polymer structure. Sulfide oxidation into sulfoxide and sulfone groups increases hydrophilicity. (b) Pheophorbide A structure. (c) BCN self-assembly and cargo loading were performed using flash nanoprecipitation (FNP).

(Scheme 1a).¹⁴ This oxidation-based alteration in the hydrophilic mass fraction of the diblock copolymer drives morphological changes in the aggregate nanostructure, and it has previously been demonstrated that this morphological change can result in the release of cargo from PEG-*b*-PPS nanostructures.¹² We have previously demonstrated that PEG-*b*-PPS filomicelles and polymersomes transition into micelles upon photo-oxidation triggered degradation,^{12,14} but how this process impacts intracellular delivery of BCN has not been previously explored.

We hypothesized that PEG-*b*-PPS BCN nanocarriers would stably retain cargo even upon internalization by cells due to their enhanced nanoarchitecture-dependent stability.⁷ Furthermore, we reasoned that their large hydrophobic PPS volume may serve as a dense sponge for enhanced scavenging of reactive oxygen species (ROS),¹⁵ providing BCN with sensitivity to photo-oxidizers for triggered release but without associated ROS-induced cytotoxicity. To test this, we loaded BCN with the cytotoxic photosensitizer pheophorbide A (PhA) and assessed its ability to safely induce intracellular degradation of BCN. Co-loading of BCN with PhA and cytotoxic chemotherapeutics demonstrated an off-on on-demand photosensitive cytotoxicity (Scheme 1b), verifying that the BCN nanoarchitecture could retain cytotoxic payloads intracellularly with extended cell viability. Such capability may allow usage of more sensitive and a wider range of photosensitizers during photo-triggered intracellular delivery as well as decrease off-target side effects associated with photodynamic nanotherapy.

Results and Discussion

To investigate the ability of BCN to stably retain cargo within cell endosomal compartments, we encapsulated fluorescein-tagged

bovine serum albumin (FITC-BSA) within BCNs using flash nanoprecipitation (FNP). FNP is currently the only scalable technique capable of forming uniform spherical BCNs and loading them with hydrophilic and hydrophobic compounds (Scheme 1c).^{8,11,16} Free FITC-BSA was rapidly endocytosed by phagocytic RAW 264.7 cells, forming small punctae that colocalized with a lysosomal stain (Figure 1a). These FITC-BSA punctae decreased in intensity over the course of 48 h, likely due to pH-dependent degradation of the fluorophore and the BSA, which is known to occur rapidly.^{17,18} In contrast, BCN-encapsulated FITC-BSA retained fluorescence intensity within endosomes (Figure S1) well beyond 48 h and was still visible within cells 168 h (7 days) after internalization (Figure 1b). Overall, the intracellular retention at the 168 h timepoint for BCN-loaded FITC-BSA was comparable to free FITC-BSA after 48 h. The persistence of fluorescent punctae confirmed using lysotracker red and lack of FITC-BSA cytosolic signal strongly suggests that BCNs aid in the stable retention of cargo within the lysosomes of cells. Indeed, quantification of fluorescent signal from cells demonstrates that while free FITC-BSA signal was higher at the early 4 h time point compared to FITC-BSA BCNs, it was significantly lower than the FITC-BSA BCN signal at 48 h (Figure 1c). This precipitous decline in free FITC-BSA intracellular signal is not seen with FITC-BSA encapsulated within BCNs between 4 h and 48 h (Figure 1c). To further confirm this phenomenon, BCNs were loaded with a BODIPY-BSA and intracellular degradation was quantified over a 24 h period. BODIPY-BSA utilizes a quenched fluorophore, which increases in fluorescence as the BSA is degraded until the BODIPY itself degrades. Free BODIPY-BSA treated cells demonstrated a rapid increase in fluorescence that peaked within 2 h, indicating the nearly immediate degradation of the BSA and resulting dequenching of the BODIPY fluorophores

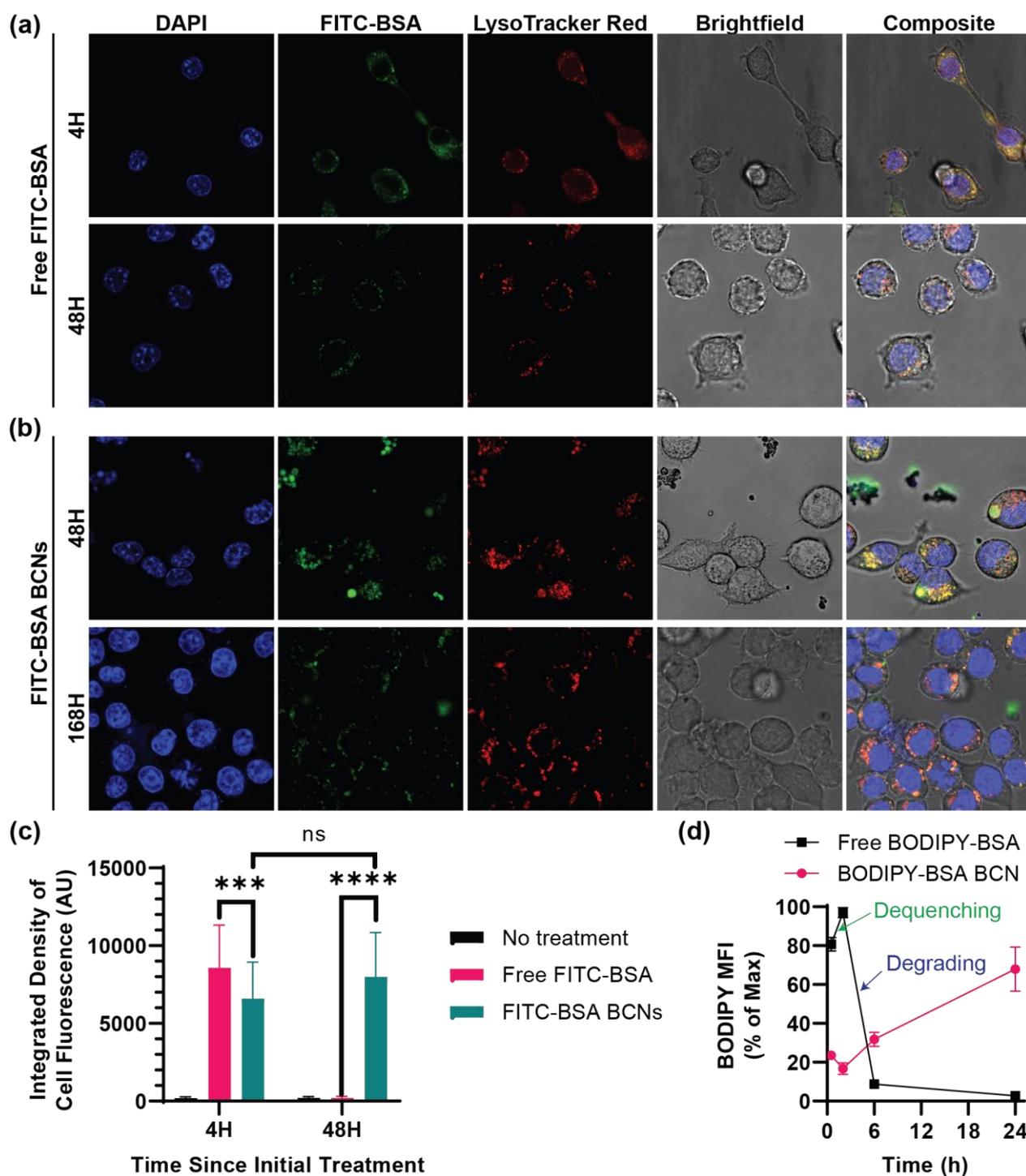


Figure 1. Subcellular localization and retention of free or encapsulated FITC-BSA. (a) Representative confocal micrographs of RAW 264.7 cells immediately after a 4-h incubation with FITC-BSA or 48 h after a 4-h incubation with FITC-BSA. (b) Representative confocal micrographs of RAW 264.7 cells after 48 or 168 h after a 4-h incubation with BCNs loaded with FITC-BSA. All cells were stained with NucBlue for nuclei and LysoTracker Red for lysosomes, magnification 40x objective lens. (c) Integrated density of fluorescent signal for randomly selected cells that underwent no treatment or treatment with either free FITC-BSA or FITC-BSA BCNs. Data was obtained for 30 cells per condition and 3 separate experiments using ImageJ Software. Cells were analyzed immediately after a 4 h incubation with treatment or 44 h after a 4 h incubation with treatment (48 h total). Statistical analysis was performed using 2-way ANOVA, *** $p < 0.001$ and **** $p < 0.0001$. (d) Flow cytometric analysis of BODIPY fluorescence in RAW 264.7 cells at various timepoints after a 30-minute BODIPY-BSA BCN incubation ($n = 4$; error bars = s.d.).

(Figure 1d). This was followed by a gradual decline in fluorescence as the fluorophore is processed and cleared by the cells. In contrast, BODIPY-BSA encapsulated in BCNs

demonstrated a very gradual increase in fluorescence and never peaked over the 24 h period of the study, indicating retarded degradation of the BSA.

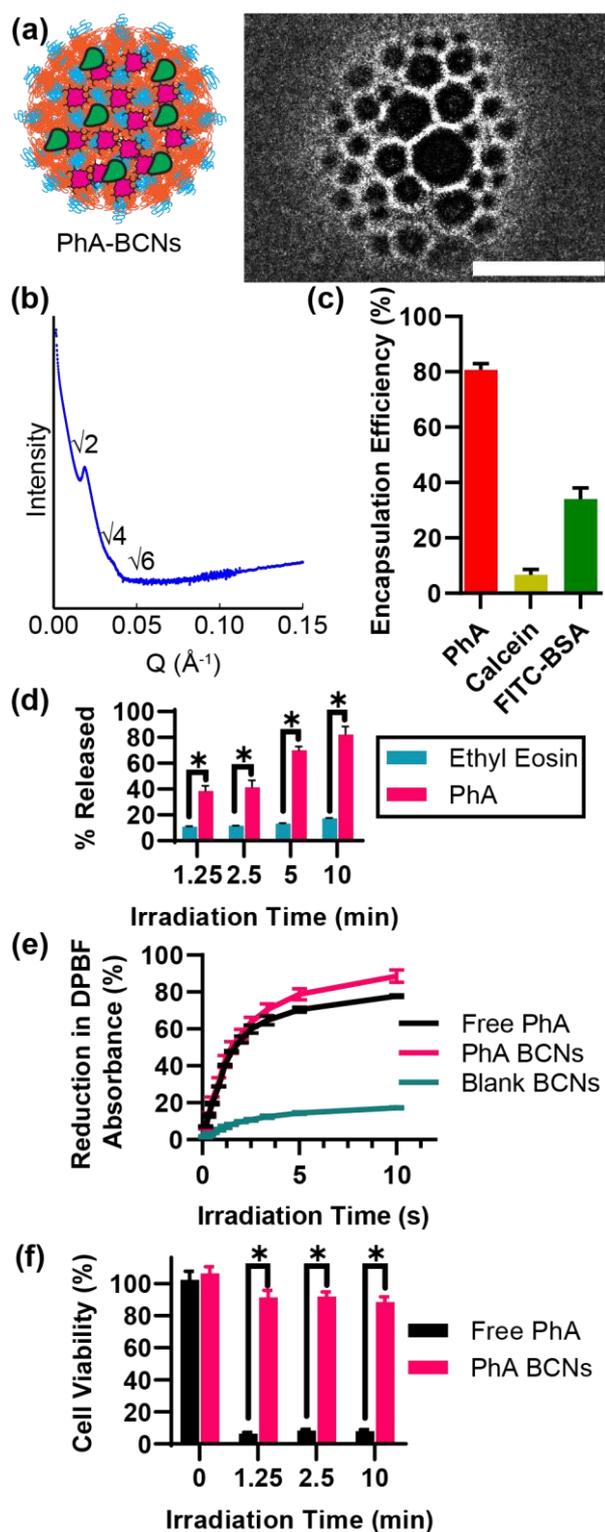


Figure 2. Physical characterization of PhA-BCNs. (a) TEM micrograph of PhA-BCN. Scale bar = 100 nm. (b) SAXS curve of PhA-BCN, with labelled Bragg peaks. (c) Encapsulation efficiency of PhA and hydrophilic cargoes calcein and FITC-BSA (n = 3, error bars = s.d.). (d) Percentage of fluorescence signal in the supernatant after irradiation and centrifugation of BCNs loaded with PhA and ethyl eosin, n = 4, error bars = s.d. (e) Generation of singlet oxygen assayed by percent decrease in DPBF absorbance for free solubilized PhA, blank BCNs, and PhA-BCNs (n = 4, error bars = s.d.). (f) MTT assay of cell viability with or without irradiation for cells treated with free PhA or PhA-BCNs, 12 h after irradiation. For all subfigures * p < 0.0001.

We next sought to probe the stimuli-responsiveness of BCNs by co-encapsulating a photo-oxidizer. We chose pheophorbide A (PhA), known to produce singlet oxygen and ROS at such high levels that it is typically employed to induce cell death for cancer applications.^{19,20} PhA is a relatively hydrophobic molecule (logP = 6.93), which correlates well with high encapsulation efficiency in PEG-*b*-PPS nanostructure systems.^{8,11,13,21} However, encapsulation of hydrophobic cargoes can at times disrupt the aggregate morphology of nanostructures, so we assessed the PhA-loaded BCN (PhA-BCN) formulations for their morphological characteristics.⁷ Negative-stained transmission electron micrographs (TEM) showed that PhA-BCNs retained their characteristic morphology (**Figure 2a and S2**). DLS analysis of the formulations found a diameter of 245.8 ± 11.6 nm and a PDI of 0.15 ± 0.02 , which was in good agreement with previous studies on the characteristics of PEG-*b*-PPS BCNs.^{7,8} Formulations were also analysed by small-angle x-ray scattering for Bragg peaks at the $\sqrt{2}$, $\sqrt{4}$, and $\sqrt{6}$ ratios, indicating that their primitive cubic type (*Im3m*) internal organization was preserved (**Figure 2b**).⁶ As expected for its hydrophobicity, PhA was encapsulated at a high efficiency of $80.7 \pm 2.2\%$. Hydrophilic cargo was simultaneously encapsulated with PhA into BCNs for release studies (**Figure 2c**). Agreeing with previously published results, loading of hydrophilic payloads into BCNs is size dependent, with FITC-BSA outperforming calcein encapsulation, $34 \pm 4\%$ vs $6.7 \pm 1.9\%$, respectively.⁸

Photo-oxidation of PEG-*b*-PPS nanostructures had previously been explored using the photo-oxidative fluorophore ethyl eosin.^{8, 14} Other nanostructures, such as polymersomes and filomicelles, appeared to readily release cargo upon photo-oxidation with ethyl eosin, but BCNs contain considerably more PPS in their interior volume and may therefore require a stronger photo-oxidizer. We therefore compared the ability of ethyl eosin and PhA to induce photo-oxidation-triggered degradation of BCNs. After irradiation of a solution of BCNs loaded with photo-oxidizer, the samples were centrifuged to pellet the intact BCNs for sampling of the supernatant. PhA was released into the supernatant in significantly greater amounts than the comparably ineffective ethyl eosin (**Figure 2d and S3**), verifying the superior capacity of PhA to generate the oxidation required for degradation of the dense BCN nanoarchitecture. These results also demonstrated the high stability of BCN and their ability to resist degradation under conditions that would normally rupture polymersomes¹⁴. Upon irradiation, free PhA significantly decreased the absorbance of DPBF, a probe for singlet oxygen generation (**Figures 2e and S4**). Blank BCNs, i.e. BCNs which were not loaded with PhA, showed no effect on DPBF absorbance. Encapsulation of PhA in BCNs did not inhibit the generation of singlet oxygen, with PhA-BCNs demonstrating significantly more singlet oxygen generation than blank BCNs (p < 0.0001). This generation of singlet oxygen was able to trigger the release of cargo from BCNs loaded with both PhA and a hydrophilic molecule – either calcein or FITC-BSA. After irradiation, BCNs were centrifuged into a pellet and the supernatant containing released cargo was removed. The pellet was resuspended and assayed for the presence of cargo. Both hydrophilic cargoes were rapidly released from BCNs upon

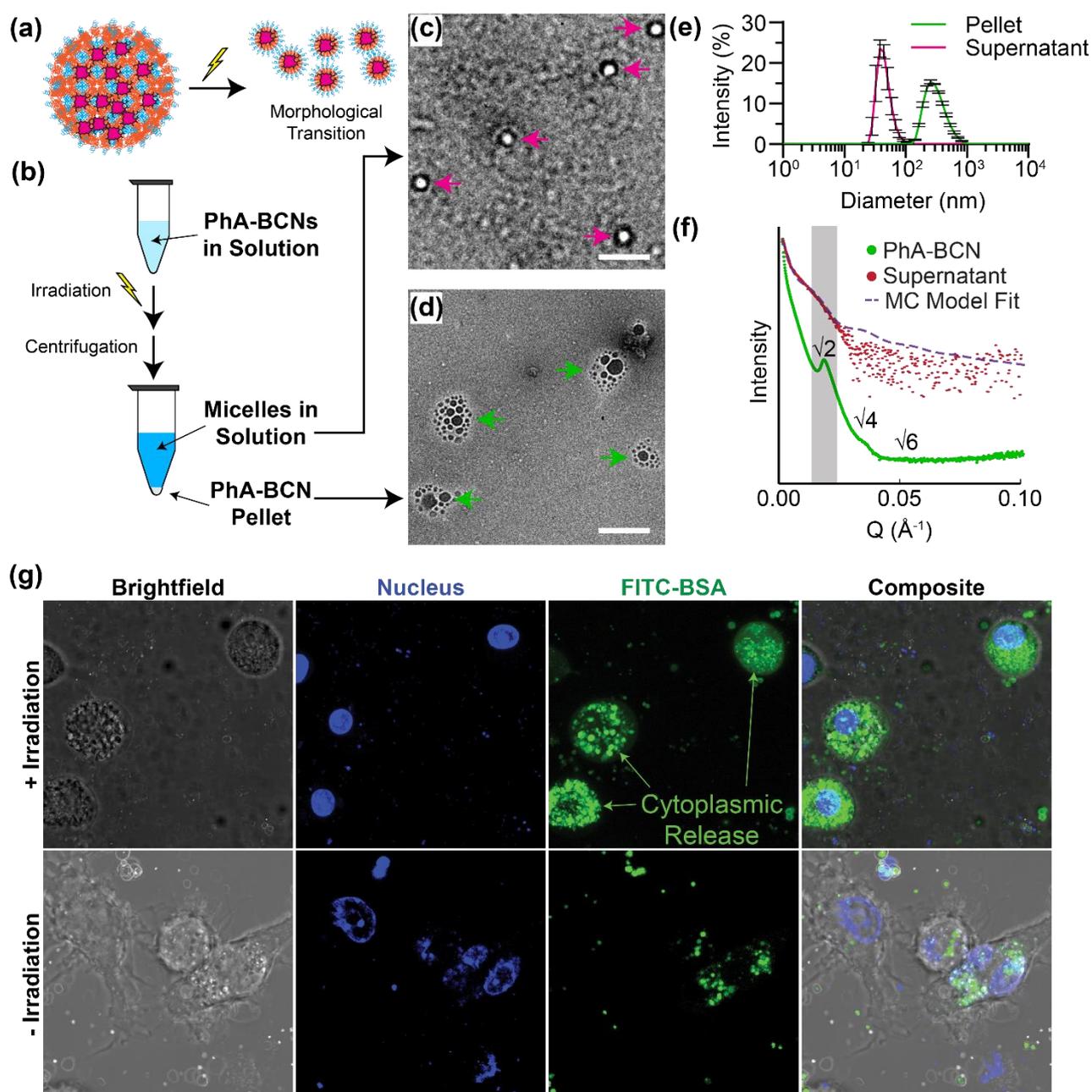


Figure 3. Morphological transition of BCNs into micelles by photo-oxidation. (a) Diagram of PhA-BCNs degrading into micelles after irradiation. (b) Schematic of BCN isolation from micelles by centrifugation at 10,000 rcf. (c) TEM micrograph of micelle nanostructures isolated from the supernatant of centrifuged samples after irradiation. (d) TEM micrograph of BCNs isolated from the pellet of centrifuged samples after irradiation. Scale bar of TEM micrographs = 200 nm. (e) DLS size distributions of samples shown in (c) and (d) ($n = 3$, error bars = s.d.). (f) SAXS curve of the supernatant shown in (c), shown with the SAXS curve of non-irradiated PhA-BCNs (same curve as shown in Figure 2b). Supernatant SAXS data was fitted with a micelle model fit using SASView. SAXS data was arbitrarily offset between BCN and supernatant data. (g) Confocal images of PhA-BCNs loaded with FITC-BSA, internalized by RAW 264.7 cells, with or without irradiation, magnification 63x objective lens.

irradiation, most likely due to the destabilization of the BCN morphology allowing for release from the internal aqueous channels (Figure S5a). This release of cargo, determined by measuring the decrease in fluorescence of the sample, was corrected for any photobleaching effects (Figure S5b).

The most common application of PhA is as a photodynamic therapy agent for generating cytotoxic ROS in anti-cancer applications.¹⁹ Indeed, free PhA with very brief irradiation was able to promote significant cell death in RAW 264.7 cells (Figure

2f). In contrast, PhA-BCNs demonstrated little, though statistically significant ($p < 0.001$), decreases in cell viability after the same duration of irradiation compared to non-irradiated cells, despite forming similar levels of singlet oxygen (Figure 2e). This difference in ROS generation (Figure S6) is likely due to the rapid consumption of singlet oxygen species by PPS,¹⁵ which effectively scavenged ROS in the immediate area where they are being formed.

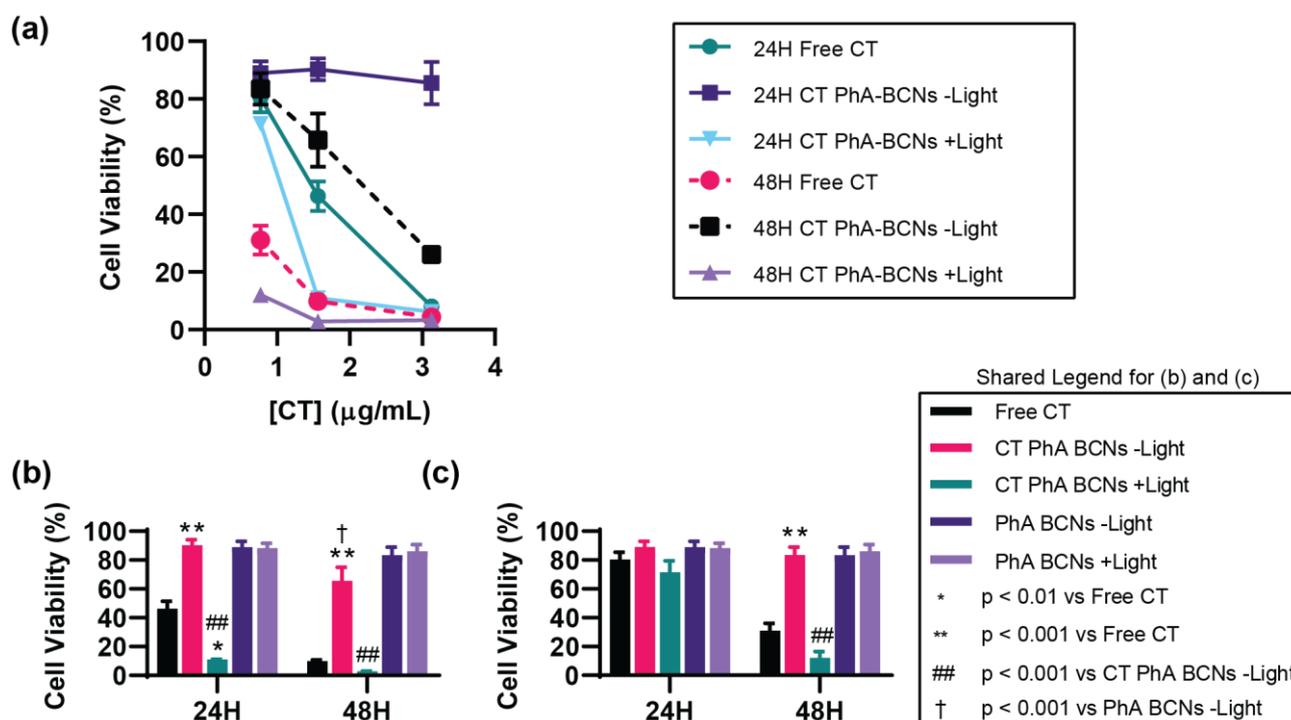


Figure 4. Cytotoxic delivery of cytotoxic cargo. (a) MTT viability assay of RAW 264.7 cells. Cells were treated with free camptothecin (CT) or PhA-BCNs loaded with CT at three concentrations (0.63, 1.56, and 3.1 µg/mL) for 8 h before a group was irradiated. Cells were irradiated for 2.5 minutes and incubated further to allow for the induction of cell death, either 24 or 48 h total. Statistical evaluation of the treatment groups at the (b) 1.56 µg/mL and (c) 0.63 µg/mL concentrations. PhA BCN -Light and +Light data at 24 and 48 h are duplicated between (b) and (c), displayed as such for ease of comparison. For all subfigures $n=4$, error bars are s.d., statistical analysis by 2-way ANOVA.

In PEG-*b*-PPS polymersome and filomicelle systems, photo-oxidation triggers morphological changes, ultimately resulting in the formation of micelle structures.^{12,14} We hypothesized that a similar morphological transition would occur for photo-oxidized BCNs (Figure 3a). To investigate, we sought to enrich for the morphological transition products by exploiting the propensity of the highly dense BCNs to pellet upon centrifugation (Figure 3b). Examination of the supernatant and pellet using TEM revealed that the supernatant contained micellar structures while the pellet contained BCNs (Figure 3c, d and S7). Dynamic light scattering (DLS) size distributions showed a reduction in the diameter of nanostructures after irradiation, representing a shift from larger BCNs to smaller micelles (Figure 3e), which was also seen after quantification of nanostructure diameters via TEM (Figure S8). This change in aggregate structure was corroborated by SAXS data, which revealed changes in scattering upon irradiation, particularly the loss of the high intensity crystalline ν_2 Bragg peak in the supernatant (Figure 3f). The supernatant SAXS scattering profile was best fit ($\chi^2 = 0.0005$) by a micelle model with core radius of 16.8 nm. Since micellar structures contain a hydrophobic core and are unable to load hydrophilic molecules, this transition from BCN to micelle presents an explanation for the rapid release of hydrophilic cargo after irradiation of PhA-BCNs.

Although often difficult to achieve without cytotoxicity, cytosolic delivery of endosomal payloads is particularly useful for a variety of therapeutic applications, including vaccination.^{9,22} A number of mechanisms have been employed

to traverse endosomal membranes, the majority of which induce lysosomal rupture and release of previously sequestered toxic enzymes and ions.²³ In contrast, PEG-*b*-PPS nanostructures have been found to deliver payloads to the cytosol via temporarily increasing endosomal membrane permeability and without toxicity.^{9,14} For polymersomes, this process involves the oxidation and resulting increased hydrophilicity of the PPS block (Scheme 1a), which triggers a thermodynamically driven conversion from stable vesicles to unstable micelles capable of fusing with and permeabilizing neighbouring membranes for milliseconds.¹⁴ We hypothesized that the morphological transition of BCNs and contemporaneous release of hydrophilic cargo could similarly destabilize endosomal compartments to also allow release of hydrophilic cargo into the cytosol. As a qualitative assessment of this hypothesis, PhA FITC-BSA BCNs, i.e. BCNs co-encapsulating both PhA and FITC-BSA, were incubated with RAW 264.7 cells. Cells were repeatedly washed to remove BCNs that were not internalized and subsequently irradiated prior to confocal imaging. Both irradiated cells and non-irradiated controls contained green fluorescent punctae representing BCNs and cargo localized within the endolysosomal pathway (Figure 3g and S9). Non-irradiated cells only featured fluorescence within these punctae, reminiscent of FITC-BSA BCNs imaged in Figure 1. In contrast, irradiated cells featured cytosolic release of the FITC-BSA, visible as a diffuse fluorescence throughout the cytosol.

Having demonstrated that PhA-BCNs are both minimally toxic and capable of triggered cytosolic delivery, we sought to

assess their ability to protect cells from toxic cargo until on-demand release was triggered for off-on cytotoxicity. Camptothecin is an anti-cancer drug that functions by inhibiting DNA topoisomerase I, a protein present in the nucleus of cells.²⁴ Thus, camptothecin activity is dependent upon cytoplasmic and ultimately nuclear localization, which results in apoptosis and a rapid decrease in cell viability. Reasoning that the release of hydrophilic camptothecin from BCNs would occur upon oxidation of PEG-*b*-PPS, we loaded BCNs with camptothecin, with or without co-loading of PhA. Camptothecin-loaded PhA-BCNs (CT-PhA-BCNs) containing three concentrations of loaded drug were evaluated for cytotoxicity at 24 and 48 h after an 8 h incubation. Samples were either irradiated or non-irradiated and were also compared to free camptothecin. Non-irradiated CT-PhA-BCNs demonstrated no change in cytotoxicity at 24 h across the three camptothecin concentrations (**Figure 4a**). This was statistically very similar to the non-toxic effects of PhA-BCNs as seen in **Figure 2f** ($p = 0.091$). Non-irradiated CT-PhA-BCNs did show cytotoxicity at 48 h in a dose dependent manner, suggesting that by that timepoint there is enough release of drug to have a cytotoxic effect at the higher concentrations (**Figure 4a**, dagger marker). In contrast to the non-irradiated encapsulated drug, free camptothecin demonstrated dose-dependent cytotoxicity at 24 h and high cytotoxicity regardless of concentration at 48 h. The irradiation of CT-PhA-BCNs, which resulted in cytosolic release of camptothecin, significantly reduced the viability of RAW 264.7 cells. At a camptothecin concentration of 1.56 $\mu\text{g/mL}$, irradiated CT-PhA-BCNs were significantly more cytotoxic than non-irradiated samples and at 24 h were even significantly more cytotoxic than free drug (**Figure 4b**). At the lower 0.63 $\mu\text{g/mL}$ concentration, significant cytotoxic effects were not seen for any treatment at 24 h (**Figure 4c**). At 48 h however, both irradiated CT-PhA-BCNs and free CT were significantly more cytotoxic than non-irradiated CT-PhA-BCNs. These results demonstrate that at relevant drug concentrations, BCNs can safely retain drug within endosomal compartments until release is triggered, enhancing both the efficacy and selectivity of on-demand cytotoxicity.

Conclusions

In this work, we have demonstrated the first application of photo-sensitive polymeric BCNs by combining the photo-oxidative effects of PhA with oxidation-sensitive PEG-*b*-PPS. Light-induced oxidation of PEG-*b*-PPS BCNs resulted in a triggerable *in situ* morphological transition from bicontinuous to micellar nanostructures. The PEG-*b*-PPS polymer functioned as an ROS 'sponge' to inhibit the typical cytotoxicity and pro-apoptotic effects of PhA. Photo-oxidation of PhA-BCNs within the endosomes of cells resulted in the release of cargo into the cytosol. Such endosomal escape is a critical step in the delivery of many different types of cargo but is difficult to control in an off-on fashion with most triggerable polymeric nanocarriers, as rapid degradation can occur regardless of external stimulation.

Photosensitive self-assembled nanostructures are frequently employed to enhance localized delivery of systemically administered therapeutics. While photo-oxidation

will significantly enhance delivery within irradiated tissues, self-assembled nanocarriers will still release payloads within off-target cells due to the degradative environments of the endolysosomal pathway. Here, we find that a bicontinuous nanoarchitecture enhances nanocarrier stability within cells, allowing PEG-*b*-PPS BCNs to resist degradation and stably retain payloads within cells under conditions that would otherwise rapidly cause disassembly for other self-assembled nanostructures of the same chemical composition, such as PEG-*b*-PPS polymersomes and filomicelles. We found that BCN protected cells from cytotoxic payloads, including pro-apoptotic photo-oxidizer pheophorbide A while under irradiation as well as the chemotherapeutic camptothecin. Interestingly, photo-oxidation induced a morphological transition of BCNs to a less stable micellar morphology, a process that we characterize via small-angle X-ray scattering and electron microscopy. This morphological transition induced cytosolic delivery of encapsulated camptothecin to both recover and enhance its cytotoxicity. To the best of our knowledge, this is the first demonstration of an inducible bicontinuous-to-micellar transition.

As BCNs inhibited cytoplasmic payload release under normal conditions without irradiation, this highlights the potential use of off-on triggerable photo-oxidation-sensitive platforms for improved control over cytosolic delivery. We demonstrated an advantage of this platform for the intracellular delivery of cytotoxic chemotherapeutics, significantly decreasing apoptosis under ambient conditions while maintain high efficacy upon light stimulation. Such platforms may serve to decrease off-target effects during nanotherapy as well as enhance cell-mediated drug delivery that requires payloads to remain within cells with minimal cytotoxicity, wherein cells can be loaded with therapeutics *ex vivo* and deliver payloads to specific organs and locations of disease.

Conflicts of interest

There are no conflicts to declare.

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TOC Summary: PEG-*b*-PPS bicontinuous nanospheres loaded with a photosensitizing agent demonstrate off-on inducible photo-responsive release of cytotoxic agents into the cytosol of cells. This light-responsive platform leverages the high intracellular stability and cargo-protection afforded by PEG-*b*-PPS bicontinuous nanospheres to allow for controlled delivery of biologically relevant hydrophilic and hydrophobic compounds.

Keywords: triggered delivery, stimuli-responsive, nanocarrier, controlled delivery, bicontinuous nanosphere, photo-oxidation

