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Introduction

A key scientific challenge of widespread current interest involves the creation of well-defined functional materials that exist on a length-scale of nanometers to microns. The solution self-assembly of amphiphilic block copolymers (BCPs) offers a convenient and scalable route to core-shell nanoparticles derived from soft matter (micelles) with potential applications in fields ranging from phase transfer catalysis to drug delivery.¹ However, a major problem in this field is the limited ability to create particles with predictable shape, dimensions, and spatially-confined chemistry.

Recent work has demonstrated that when the core-forming block of a BCP is able to crystallize, morphologies of low interfacial curvature, such as cylinders and platelets, are generally favoured. Furthermore, using a seeded growth approach, the length and/or width of the micelles can be increased by epitaxial crystallization of added BCP.²⁻⁶ The dimensional increase is linearly related to the ratio of existing micelles to added BCP, leading to the descriptor "Living Crvstallization-Driven Self-Assembly" (CDSA) by analogy to the living covalent polymerization of molecular monomers.^{5,6} We have developed this synthetic platform to yield highly monodisperse cylinders of variable length that exhibit lyotropic liquid crystalline phases,⁷ core and coronal segmented comicelles of potential use as heterojunctions and barcodes,⁸ and other well-defined complex architectures.^{4,6} Living CDSA has also enabled the preparation of amphiphilic block comicelles, which themselves are capable of hierarchical selfassembly to yield supermicelles and superlattices.9 To date, a variety of different BCPs with a crystallizable core-forming block has been shown to undergo living CDSA,¹⁰ and the process is also applicable to planar species¹¹ that form π -stacked fibre-like structures. BCPs with а poly(ferrocenyldimethylsilane) (PFS) core-forming block have been the most widely studied.

The realization of complex self-assembled structures in solution requires access to more advanced analytical techniques that permit the study of systems in their native environment. Current methods for micelle characterization either provide ensemble measurements (e.g. static light scattering or bulk fluorescence), possess limited resolution (e.g. laser scanning confocal microscopy, LSCM), or require the invasive removal of solvent prior to imaging (e.g. electron microscopy).¹² Super-resolution fluorescence microscopy techniques¹³ offer an attractive solution to these problems.¹⁴

Methods

Structured illumination microscopy (SIM) was performed using a Zeiss Elyra PS. 1 microscope with a $100 \times$ oil immersion objective lens (NA = 1.46), with fluorophores from R, G and B polymers excited by diode lasers operating at 642 nm, 488 nm and 405 nm, respectively. Raw images were acquired at three grating angles, and with observation windows of 655–800 nm, 495–575 nm and 420–480 nm for R, G and B. The resulting outputs from both LSCM and SIM were obtained as digital false-colour images, and colour coded as red, green and blue, respectively.

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Results and Discussion

Recent research in our group has enabled the fabrication of uniform rectangular 2D platelet materials prepared from the seeded growth of homopolymer/BCP blends (Figure 1).¹⁵





Replacement of the PFS_{28} -*b*-PDMS₅₆₀ BCP with the fluorescently labelled BCP, PFS_{29} -*b*-(PDMS₆₅₂/DYE₁₉) (Figure 2) enabled the formation of concentric rectangular platelets of segmented functionality. PFS_{29} -*b*-(PDMS₆₅₂/DYE₁₉) was synthesized through the reaction of a BCP functionalized with pendent amine groups and then reacted with the *N*-hydroxysuccinimidyl (NHS) esters of fluorophores based on a 4,4-difluoro-4-bora-3a,4a-diaza-s-indacene (BODIPY) core.⁶ Using this method, approximately 3% dye molecules are randomly distributed along the coil block.



Figure 2: Structure of PFS₂₉-*b*-(PDMS₆₅₂/DYE₁₉), labelled with red (R), green (G), and blue (B), BODIPY dyes.

Using the fluorescent BCPs, PFS_{29} -*b*-($PDMS_{652}/DYE_{19}$), a range of single- and multi-colour rectangular platelet strucutres were prepared, which were around 10 μ m in length. These structures were then visualized using both LSCM and SIM (Figure 3). Using SIM technique we were able to resolve the different fluorescent domains without having to employ non-fluorescent domains (spacers) in between the fluorescent segments and produce superior images to those we obtained using LSCM.

Conclusions

We have successfully employed SIM to image nanostructures prepared from the living CDSA of fluorescent BCPs. This technique enabled the structures to be studied in their native environment at higher resolution than afforded by LSCM.



Figure 3: Uniform multiblock rectangular platelets selectively functionalized using fluorescent PFS BCPs. a) Schematic representations b) LSCM and c) SIM images of typical rectangular platelet block comicelles, with segregated regions composed of nonfluorescent P2VP coronas and multiple dye-functionalized fluorescent PDMS coronas. The PDMS coronas with red, green, and blue fluorescence are denoted as PDMS-R, PDMS-G, and PDMS-B, respectively. Figure reproduced with permissions from Ref. [15].

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