Biomacromolecules

Article

Cite This: Biomacromolecules 2020, 21, 89−94

pubs.acs.org/Biomac

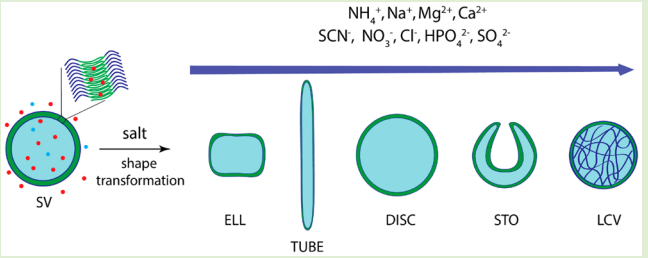
Tailoring Polymersome Shape Using the Hofmeister Effect

Yongjun Men,[§] Wei Li,[§] Coralie Lebleu, Jiawei Sun, and Daniela A. Wilson*

Radboud University, Institute for Molecules and Materials, Heyendaalseweg 135, 6525 AJ, Nijmegen, The Netherlands

Supporting Information

ABSTRACT: Reshaping polymersomes remains a challenge for both size and shape control, methodology development, and mechanism understanding, which hindered their application in nanomedicine and nanomachine. Unlike liposome, polymersomes are capable of maintaining their shape due to their rigid and glassy membrane. Here we use the Hofmeister effect to guide the shape control of polymersome by tuning the ion type and concentration. Multiple morphologies such as ellipsoid, tube, disc, stomatocytes, and large compound vesicles are found. These results give evidence of demonstrating that the shape changes are not only induced by osmotic pressure, but also by the interaction with the polymersome membranes. Additionally, this methodology provides a general tool to tailor the shape of polymersome into various morphologies.



INTRODUCTION

Cell organelles have a variety of morphologies that estimate the form in a complex environment that contains water, salts, and others. Several artificial systems are employed to mimic the morphologies of cell organelles for applications ranging from nanomedical systems to nanoreactors, such as liposomes and polymersomes.[1−7] When comparing with liposomes, polymersomes, polymeric vesicles self-assembled from synthetic amphiphilic block copolymers, have thicker membranes that display enhanced stability and membrane integrity under a wide range of conditions.[8−13] Additionally, the thicker membranes allow, from an energy point of view, the storage of more energy in the membranes transitory via chain enwind, and then the energy is slowly released by polymer rearrangement of the resulting shape changes at a time scale from minutes to days.[14] This unique property allows the various intermediate shapes at the nanoscale to be captured and visualized with the assistance of (cryogenic-)TEM.

Several pioneering studies have demonstrated that unusual shapes could be controlled uniformly by shape transformation from spherical polymersomes via osmotic pressure, magnetic field, and chemical structure changes.[15−19] For example, poly(ethylene glycol)-*block*-poly(styrene) (PEG-*b*-PS) spherical polymersomes transferred to stomatocytes, bowl-shaped polymersomes, via dialysis of their organic solvent solution against pure water, which created the osmotic pressure differences over the PS membrane that induced the shape changes.[14] The compelling dual-compartmentalized stomatocyte-structures are capable of encapsulating catalytic nanoparticles or enzymatic networks toward the formation of complex nanoreactors and nanomotors.[20,21] Further studies demonstrated that the pathway of the shape change is strongly influenced by the bending energy (E_b), which can be functionalized by three parameters: the bending rigidity (k), the mean surface curvature (C), and the spontaneous curvature (C_0), as shown in eq 1:

$$E_b = \frac{k}{2}\oint (2C - C_0)^2 \mathrm{d}A \tag{1}$$

The bending rigidity (k) is dictated by the chemical properties of the membrane; the mean surface curvature (C) is dictated by the degree of curvature at different positions on the membrane and is contingent upon the shape. Contrary to C, the spontaneous curvature (C_0) is not a consequence of the shape, but arises from asymmetry in copolymer conformation between the inner and outer surfaces and is therefore sensitive to the membrane microenvironment. A positive C_0 would promote the shape change to prolates and tubes; rather, a negative C_0 contributes to the pathway to oblates, disks, and stomatocytes.[22,23] Very recently, our research revealed that other chemical additives such as PEG can not only drive the shape change via increasing osmotic pressure, but also change the pathway to form oblates, disks, and stomatocytes and even to fuse the polymersome membrane to generate intriguing shapes such as nest and stomatocyte-in-stomatocyte in a short period of time (<1 min).[24] This result indicates that the added PEG and the PEG part of the polymersome interaction exists, which changed the values of C_0 at various conditions, leading to different shape change pathways.

In this context, we could propose that salts should also have the capability to induce the shape changes of polymersomes via

Special Issue: Future of Biomacromolecules at a Crossroads of Polymer Science and Biology

Received: July 2, 2019
Revised: September 13, 2019
Published: September 17, 2019

DOI: 10.1021/acs.biomac.9b00924
Biomacromolecules 2020, 21, 89−94

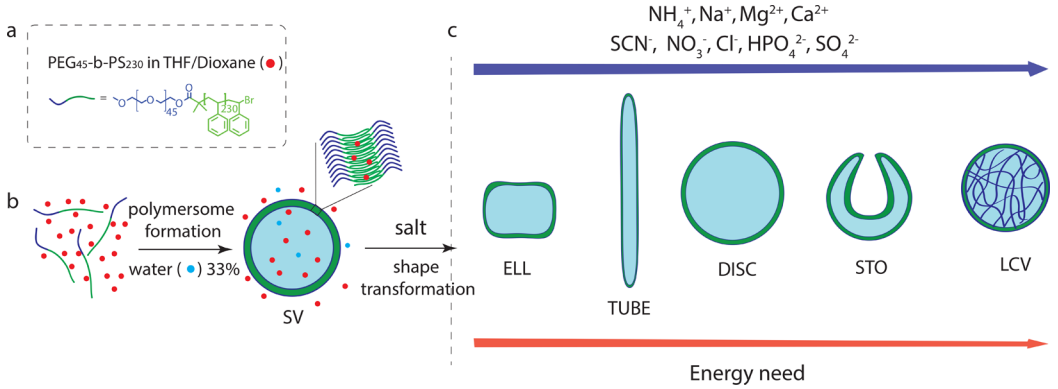
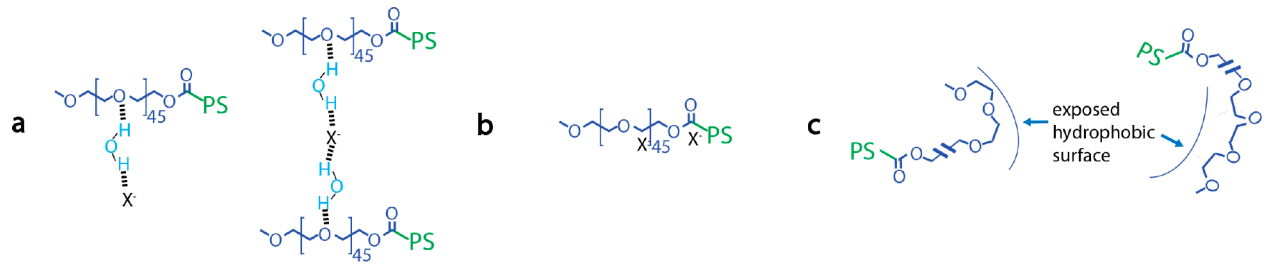
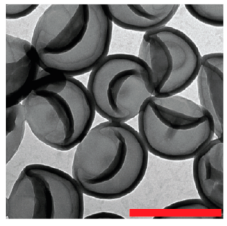


Figure 1. (a) Legend, showing the structure of the PEG-*b*-PS block copolymer building block in organic solvents. (b) Scheme of the self-assembly process. Water is slowly (1 mL·h^{-1}) added to a solution of PEG-*b*-PS in THF/dioxane until it reaches 33 vol %. Polymersomes with spherical vesicle (SV) shape was assembled after the critical aggregation point, about 20 vol % of water. (c) After addition of salts containing various cations (NH^{4+}, Na^{+}, Mg^{2+}, Ca^{2+}) and anions (SCN^{-}, NO^{3-}, Cl^{-}, HPO_4^{2-}, SO_4^{2-}) into the polymersome solution, the SV shape changed to ellipsoid (ELL), tube, disc, stomatocytes (STO), and large compound vesicles (LCV). The capacity of ions to induce these shape change follows the order of the Hofmeister series, as the arrow pointed.



d

salt \ conc. (shape)	1×10^{-2} M	1×10^{-3} M	1×10^{-4} M	1×10^{-5} M
NaSCN	ELL	SV	SV	SV
$NaNO_3$	ELL	ELL	SV	SV
NaCl	STO	ELL	SV	SV
Na_2HPO_4	LCV	STO	DISC&TUBE	SV
Na_2SO_4	LCV	STO	ELL&TUBE	SV


SV

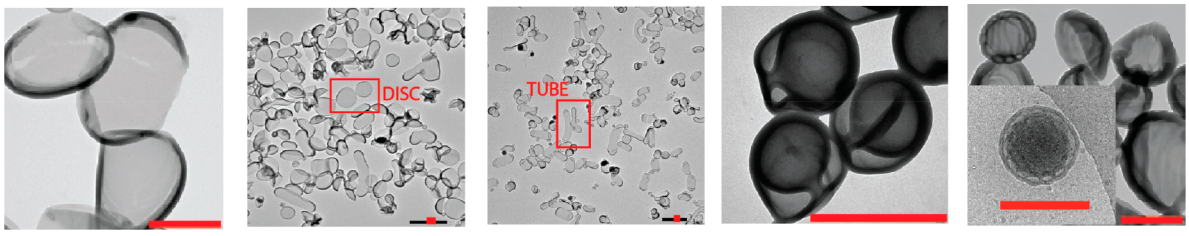
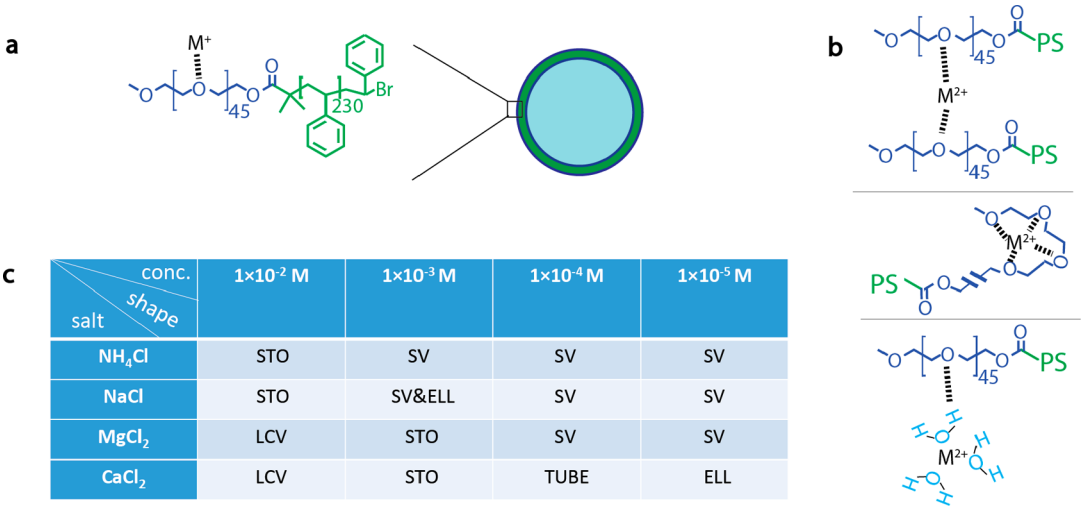


Figure 2. (a–c) Interactions among anions, PEG, and hydration waters. (a) Hydrogen bonds between water molecules and the ethylene glycol side chains are destabilized through polarization by the anion X^{-}; (b) direct binding of the anion to the polymer, leading to ion accumulation at the polymer/water interface; (c) the anions can interfere with the hydrophobic hydration of the polymer backbone by increasing or decreasing the surface tension at the polymer/water interface. (d) Shapes obtained from various sodium salts at different concentrations (10^{-2}–10^{-5} M). The TEM images of SV, ELL, STO, DISC, and LCV (cryo-TEM image inserted); scale bar (red): 500 nm. The whole shape images are presented in Figure S1. The cyro-TEM images of SV and ELL were shown in Figure S3.

an increase of osmotic pressure and to influence the pathway via affecting the C_0 values though polymer–ion or ion–hydrated polymer interaction, and this interaction should be dependent on the specific ion types. To demonstrate this hypothesis, herein, we employed eight types of common salts to test their capabilities of inducing shape change of polymersome (PEG-*b*-PS). The morphologies were followed by TEM after quenching samples into a large amount of water. Cation and anion variations were studied separately at different concentrations. Interestingly, we found that, at the same

DOI: 10.1021/acs.biomac.9b00924
Biomacromolecules 2020, 21, 89–94



salt \ conc. (shape)	1×10^{-2} M	1×10^{-3} M	1×10^{-4} M	1×10^{-5} M
NH_4Cl	STO	SV	SV	SV
NaCl	STO	SV&ELL	SV	SV
$MgCl_2$	LCV	STO	SV	SV
$CaCl_2$	LCV	STO	TUBE	ELL

Figure 3. (a, b) Interactions among cations, PEG corona, and water. (a) Monocations interact directly with the PEG oxygen atom; (b) dications interact with PEG oxygen atom from nearby two chains or from nearby ethylene glycol units of a signal chain like crown ethers, as well the highly hydrated divalent cations indirectly interact with PEG. (c) Shapes obtained from variation of the cations at different the concentration ranging from 10^{-2} M to 10^{-5} M. The whole shape images are presented in Figure S2.

concentration, different salts can induce polymersomes to transform to different shapes, while the same salt at different concentrations can lead to multiple shape changes.

EXPERIMENTAL SECTION

Materials. All reagents and chemicals were purchased from commercial sources and used as received. Milli-Q-water (18.1 MΩ) was used throughout the experiments. Molecular weights of the block copolymers were measured on a Shimadzu Prominence GPC system equipped with a PL gel 5 μm mixed D column (Polymer Laboratories) and differential refractive index and UV (254 nm) detectors. THF was used as an eluent with a flow rate of 1 mL/min. NMR spectra were performed on a Varian Inova 400 spectrometer with $CDCl_3$ as a solvent. Transmission electron microscopy (TEM) samples were prepared in the following way: a solution of sample (6 μL) was air-dried on a carbon-coated Cu TEM grid (200 mesh). A TEM JEOL 1010 microscope at an acceleration voltage of 60 kV was used to perform the measurements. Sonicator VWR USC300TH was used for the sonication experiments at room temperature. A JEOL 2100 cryo-Transmission Electron Microscope was used for characterization of polymersome structures. Poly(ethylene glycol) macroinitiators and block copolymers, poly(ethylene glycol)-*b*-polystyrene (PEG-*b*-PS) were used the one reported previously.[12]

Preparation of Polymersomes. Modified from the former literature report,[14] a typical procedure is described: PEG_{45}-*b*-PS_{230} (20 mg) was dissolved in a solvent mixture of tetrahydrofuran (THF) and 1,4-dioxane (dioxane) (2 mL, 4:1 by volume) in a 15 mL capped vial with a magnetic stirrer. After dissolving the solution for 1 h at room temperature, a syringe pump equipped with a syringe with a needle was calibrated to deliver water with a speed of 1 mL/h. The needle from the syringe was inserted into the vial of which the cap was replaced by a rubber septum. A total of 1 mL of water was pumped into the organic solution with vigorous stirring (900 rpm). When finishing the water addition, 50 μL of the suspension was dropped at once into 1 mL of pure water with stirring, which ensured a rapid quenching of the PS domain within the bilayer of the polymersomes.

Salt-Induced Reshaping Polymersome. Polymersome suspension (200 μL) in organic/water solution was loaded in a 1.5 mL Eppendorf centrifugation tube. A total of 10 μL of salt aqueous solution (0.2−2.0 × 10^{-5} M) was added into the suspension under a shaking speed of 1200 rmp. After 1 min, 1 mL of ultrapure water was added one time in the solution to freeze the structure.

RESULTS AND DISCUSSION

Polymersome Preparation. The experimental procedure is described in Figure 1. Spherical polymersomes were assembled from 20 mg of PEG45-*b*-PS230 ($Đ$ = 1.09, the number-average molecular weight of PS was calculated via ^{1}H NMR spectroscopy) in 2 mL of THF/1,4-dioxane = 4:1 (v/v) via slow addition of water at a rate of 1 mL/h.[12] The suspension became turbid when 0.44 mL of water was added. When the volume of water reached 1 mL, 200 μL of the polymersome suspension was transferred to a centrifuge tube. Due to the relatively high organic solvent content (67 vol %), the polymersome membrane is flexible and permeable to the solvent, allowing the shape transition to occur. Then 10 μL of salts aqueous solutions was added into the suspension at once, followed by 1 min shaking for shape transition. A fraction of this solution (50 μL) was taken from this suspension and added at once to 1 mL of pure water to rapidly freeze the shape. The suspension was purified 3× by ultracentrifugation to remove the added salt, leaving for TEM sample preparation.

Shape Transformation. The results observed from TEM images demonstrated that all eight types of salts can induce the shape change of the polymersomes at certain concentrations above 1 × 10^{-5} M, following a route of ellipse, tube, disc, stomatocyte, and LCV, as shown in Figures 2d and 3c. Without considering the salt and polymer interaction, the driving force of these shape changes would be osmotic pressure. In our experiment, the salts were shortly added into the polymersome suspension that immediately caused large osmotic pressure differences over the polymersome membrane, especially at a high salt concentration, leading to the organic solvent/water squeezed out from the cavity to release the osmotic energy via deflation. The more salt was added, the bigger osmotic pressure was induced, causing the larger reduced volume (deflation) of the polymersome. This volume reduced sequence is corresponding to a shape-change sequence of SV, ELL, DISC, and STO (Figure 2d). During the deflation, the osmotic energy decreases, but the bending energy increases until it becomes a domain parameter for the shape. To minimize the bending energy polymers in the polymersome need to adjust the number proportion between the inner and

DOI: 10.1021/acs.biomac.9b00924
Biomacromolecules 2020, 21, 89−94

the outer layers, so-called surface area difference, to form the final kinetic shapes. But only osmotic pressure cannot explain the shape change at a very low salt concentration and the shape variations at the same salt concentration, but different salt types. For example, at 0.001 M NaSCN cannot change the shape while NaCl can elongate the polymersome to ELL. We suppose that the difference is mainly caused by the interaction of ions and polymer (PEG).[25] Thus, the interaction of the ions and PEG should be a coexisting effecter of the shape change. Interestingly, we found the shape change triggered by salt follows a sequence exactly the same as Hofmeister series.

Hofmeister series is a classification of ions in order of their ability to salt out or salt in proteins, which is generally more pronounced for anions than for cations.[26] SCN^-, NO_3^-, and Cl^- are referred to as chaotropes, which are known to destabilize folded proteins and give rise to the salting-in behavior, while HPO_4^{2-} and SO_4^{2-} are called kosmotropes, which are strongly hydrated and have stabilizing and salting-out effects on proteins and macromolecules.[27−32] Three types of interactions exist between the anions and the PEG polymer in water media. First, the anions can polarize an adjacent water molecule that is in turn involved in hydrogen bonding with the oxygen atom (Figure 2a). Second, the anions may bind directly to the PEG, leading to ion accumulation at the polymer/water interface (Figure 2b). Third, these species can interfere with the hydrophobic hydration of the polymer by increasing the surface tension of the cavity from the hydrophobic segment (Figure 2c). All of these three interactions have influence on the spontaneous curvature C_0, resulting in a change of E_b and shape variation.

Effects of Anions. Here we choose NaSCN, $NaNO_3$, NaCl, Na_2HPO_4, and Na_2SO_4 to test their differences of reshaping polymersome. As shown in Figure 2d, NaSCN could only elongate the spherical polymersome to ellipsoid even at a concentration of 1×10^{-2} M, similar to $NaNO_3$, both of which are chaotropic ions. But $NaNO_3$ performs slightly more efficient to induce the shape change at an order of magnetite lower concentration (1×10^{-3} M). NaCl is the only monovalence salt in this context that can reshape polymersome to stomatocyte at 1×10^{-2} M, demonstrating the order of anion for reshaping polymersome followed the Hofmeister series as $Cl^- > NO_3^- \approx SCN^-$. Divalent anions, comparing with monovalent anions, carry the higher capability to push the shape change further. Na_2HPO_4 and Na_2SO_4 as the kosmotropic ion can reshape polymersome to ellipsoid and tube at 1×10^{-4} M, stomatocytes at 1×10^{-3} M, and LCV at 1×10^{-2} M.

Ellipsoid, tube, disc, and stomatocytes shapes can be explained by the collective effect of osmotic pressure and bending energy. But the formation of LCV goes though membrane fusion process, which is more complex. We supposed an explanation based on the molar surface tension increment, $k_E = (\partial\Delta\gamma/\partial m)T$, of the anions. Previous research gave the averages k_E of different salt aqueous solutions, as $Na_2SO_4 \approx 2.77$ mN L m^{-1} mol^{-1}, NaCl ≈ 1.73 mN L m^{-1} mol^{-1}, $NaNO_3 \approx 1.21$ mN L m^{-1} mol^{-1}, and NaSCN ≈ 0.5 mN L m^{-1} mol^{-1}.[33] The shape change correlates very well to the anions' surface tension increment, which suggests that the removal of hydration waters from the hydrophobic polymer backbone (the mechanism of Figure 2c) plays an important role in the shape change process. This removal might generate bundling force to the individual polymer chains, which induces the membrane fusion to form LCV, as the cryo-TEM image shown as the inset photo in Figure 2d when Na_2SO_4 and Na_2HPO_4 are at 0.01 M.

Effects of Cations. Besides anions, a Hofmeister series has also been established for cations, with an order of $NH_4^+ > Na^+ > Mg^{2+} > Ca^{2+}$. NH_4^+ and Na^+ decrease the solubility of proteins, while Mg^{2+} and Ca^{2+} increase their solubility.[27,28] The effects of cations, however, tend to be much less pronounced than those of anions, because the cations are generally excluded from the polymer/water interface. Since the PEG corona of the polymersome is negatively charged in the aqueous solution, and the free electrons doublets on PEO oxygen atoms display an attractive force with respect to cationic species,[34] or interact with cations in a similar way as crown ethers, such direct cation binding would lead to additional charge and increased solubility of the polymer and thus to shape changes.

We tested the influence of cations on the shape change of polymersome by adding different salts (cations including NH_4^+, Na^+, Mg^{2+}, and Ca^{2+}) with chloride counterions in a concentration of 1×10^{-2} to 1×10^{-5} M. For a monovalent cation, the result is presented in Figure 3c, at 1×10^{-3} M, NH_4Cl has barely any effect on the shape change, while NaCl changes a big portion to ellipsoid, which indicates that the existence of the interaction between PEG corona and ions (Figure 3a), as well Na^+ has stronger effect than NH_4^+. Comparing with monovalent ions, divalent ions exhibit higher charge-ionic radius ratio that should bind PEG stronger and more efficiency (Figure 3b), resulting in shape change at lower concentrations. The results in Figure 3c demonstrate this hypothesis. Divalent $MgCl_2$ and $CaCl_2$ initiate shape change at concentrations approximately 1 order of magnitude lower than monovalent salts. As we hypothesize above, Ca^{2+} or Mg^{2+} are able to specifically binds with the oxygen of the ethylene oxide unite, either by an increase in the out-surface area of the corona to induce shape change to stomatocytes or by facilitating fusion by associating polymer chains inner-molecularly to form membrane-fused vesicles (LCV) by intermolecular bridges in semidilute solution, resulting in the fusion of different vesicle corona.[35,36] When the concentration of $MgCl_2$ and $CaCl_2$ reached 0.01 M, the shapes changed to LCV. Thus, the ranking order of the cation ability to initiate shape changes is opposite to the Hofmeister series as $Ca^{2+} > Mg^{2+} \gg Na^+ > NH_4^+$.

CONCLUSIONS

In conclusion, we showed that we can use the Hofmeister series to precisely reshape polymersomes from spherical vesicles to ellipsoid, tube, disc, stomatocytes and large compound vesicles. These shape changes are driven by both osmotic pressure and the interactions between salt ions and PEG corona. The salt variation strongly influences the shape change. For example, at the same concentration (1×10^{-3} M), NH_4Cl presented no influence on shape, but $MgCl_2$ changed the shape to stomatocytes. Kosmotropic ions can reshape polymersome much more efficiently than chaotropic ions, since they are more polarizable, hydrate more strongly, and interact with the PEG corona more sufficiently, as well as cross-linking the nearby PEG segments inner/intermolecularly.

The addition of common additives such as salts to tune the shape of polymersome can be exploited in protein encapsulation applications, where salts present synergistic effect. To control the shape change of polymersomes, the usually used dialysis method is time costly and results in a

DOI: 10.1021/acs.biomac.9b00924
Biomacromolecules 2020, 21, 89−94

limited number of controlled shapes. The addition of salt to polymersome can not only change the shape in a much faster manner, but also enlarge the shape portfolio of polymersomes. In this way, chaotropic ions can be used to create osmotic pressure without inducing shape changes at a relatively high concentration, which is suitable for the application of crystallization, whereas the kosmotropic ions can efficiently change the shape at a very low concentration, suitable for encapsulation of ion-sensitive particles.

■ ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acs.biomac.9b00924.

TEM and cryo-TEM images of polymersomes at different concentrations with various cations and anions (PDF)

■ AUTHOR INFORMATION

Corresponding Author

*Tel.: +31 (0)24 36 52185.

ORCID

Daniela A. Wilson: 0000-0002-8796-2274

Author Contributions

§These two authors contributed equally to this work.

Notes

The authors declare no competing financial interest.

■ ACKNOWLEDGMENTS

The authors acknowledge the European Research Council ERC-StG 307679 "StomaMotors" and NWO Chemiche Wetenschappen VIDI Grant 723.015.001 for financial support. We acknowledge support from the Ministry of Education, Culture and Science (Gravitation Program 024.001.035). W.L. acknowledges the CSC scholarship for financial support. Y.M. acknowledges fruitful discussion with Prof. Jan van Hest.

■ REFERENCES

(1) Folkman, J.; Moscona, A. Role of cell shape in growth control. *Nature* **1978**, *273*, 345.

(2) van Hest, J. C. M.; Delnoye, D. A. P.; Baars, M. W. P. L.; van Genderen, M. H. P.; Meijer, E. W. Polystyrene-Dendrimer Amphiphilic Block Copolymers with a Generation-Dependent Aggregation. *Science* **1995**, *268* (5217), 1592−1595.

(3) Zhang, L.; Eisenberg, A. Multiple Morphologies of "Crew-Cut" Aggregates of Polystyrene-b-poly(acrylic acid) Block Copolymers. *Science* **1995**, *268* (5218), 1728−1731.

(4) Zhang, L.; Yu, K.; Eisenberg, A. Ion-Induced Morphological Changes in "Crew-Cut" Aggregates of Amphiphilic Block Copolymers. *Science* **1996**, *272* (5269), 1777−1779.

(5) Discher, B. M.; Won, Y.-Y.; Ege, D. S.; Lee, J. C.-M.; Bates, F. S.; Discher, D. E.; Hammer, D. A. Polymersomes: Tough Vesicles Made from Diblock Copolymers. *Science* **1999**, *284* (5417), 1143−1146.

(6) Percec, V.; Wilson, D. A.; Leowanawat, P.; Wilson, C. J.; Hughes, A. D.; Kaucher, M. S.; Hammer, D. A.; Levine, D. H.; Kim, A. J.; Bates, F. S.; Davis, K. P.; Lodge, T. P.; Klein, M. L.; DeVane, R. H.; Aqad, E.; Rosen, B. M.; Argintaru, A. O.; Sienkowska, M. J.; Rissanen, K.; Nummelin, S.; Ropponen, J. Self-Assembly of Janus Dendrimers into Uniform Dendrimersomes and Other Complex Architectures. *Science* **2010**, *328* (5981), 1009−1014.

(7) Leong, J.; Teo, J. Y.; Aakalu, V. K.; Yang, Y. Y.; Kong, H. Engineering Polymersomes for Diagnostics and Therapy. *Adv. Healthcare Mater.* **2018**, *7*, 1701276.

(8) Blanazs, A.; Madsen, J.; Battaglia, G.; Ryan, A. J.; Armes, S. P. Mechanistic Insights for Block Copolymer Morphologies: How Do Worms Form Vesicles? *J. Am. Chem. Soc.* **2011**, *133* (41), 16581−16587.

(9) Du, J.; Tang, Y.; Lewis, A. L.; Armes, S. P. pH-Sensitive Vesicles Based on a Biocompatible Zwitterionic Diblock Copolymer. *J. Am. Chem. Soc.* **2005**, *127* (51), 17982−17983.

(10) Ladmiral, V.; Semsarilar, M.; Canton, I.; Armes, S. P. Polymerization-Induced Self-Assembly of Galactose-Functionalized Biocompatible Diblock Copolymers for Intracellular Delivery. *J. Am. Chem. Soc.* **2013**, *135* (36), 13574−13581.

(11) Wang, J.; Liu, K.; Xing, R.; Yan, X. Peptide self-assembly: thermodynamics and kinetics. *Chem. Soc. Rev.* **2016**, *45* (20), 5589−5604.

(12) Men, Y.; Peng, F.; Tu, Y.; van Hest, J. C. M.; Wilson, D. A. Methods for production of uniform small-sized polymersome with rigid membrane. *Polym. Chem.* **2016**, *7* (24), 3977−3982.

(13) Yan, X.; Zhu, P.; Li, J. Self-assembly and application of diphenylalanine-based nanostructures. *Chem. Soc. Rev.* **2010**, *39* (6), 1877−1890.

(14) Kim, K. T.; Zhu, J.; Meeuwissen, S. A.; Cornelissen, J. J. L. M.; Pochan, D. J.; Nolte, R. J. M.; van Hest, J. C. M. Polymersome Stomatocytes: Controlled Shape Transformation in Polymer Vesicles. *J. Am. Chem. Soc.* **2010**, *132* (36), 12522−12524.

(15) Rikken, R. S. M.; Engelkamp, H.; Nolte, R. J. M.; Maan, J. C.; van Hest, J. C. M.; Wilson, D. A.; Christianen, P. C. M. Shaping polymersomes into predictable morphologies via out-of-equilibrium self-assembly. *Nat. Commun.* **2016**, *7*, 12606.

(16) Zhu, J.; Zhang, S.; Zhang, K.; Wang, X.; Mays, J. W.; Wooley, K. L.; Pochan, D. J. Disk-cylinder and disk-sphere nanoparticles via a block copolymer blend solution construction. *Nat. Commun.* **2013**, *4*, 2297.

(17) Rikken, R. S. M.; Kerkenaar, H. H. M.; Nolte, R. J. M.; Maan, J. C.; van Hest, J. C. M.; Christianen, P. C. M.; Wilson, D. A. Probing morphological changes in polymersomes with magnetic birefringence. *Chem. Commun.* **2014**, *50* (40), 5394−5396.

(18) van Oers, M. C. M.; Rutjes, F. P. J. T.; van Hest, J. C. M. Tubular Polymersomes: A Cross-Linker-Induced Shape Transformation. *J. Am. Chem. Soc.* **2013**, *135* (44), 16308−16311.

(19) Salva, R.; Le Meins, J.-F.; Sandre, O.; Brûlet, A.; Schmutz, M.; Guenoun, P.; Lecommandoux, S. Polymersome Shape Transformation at the Nanoscale. *ACS Nano* **2013**, *7* (10), 9298−9311.

(20) Tu, Y.; Peng, F.; Sui, X.; Men, Y.; White, P. B.; van Hest, J. C. M.; Wilson, D. A. Self-propelled supramolecular nanomotors with temperature-responsive speed regulation. *Nat. Chem.* **2017**, *9* (5), 480−486.

(21) Wilson, D. A.; Nolte, R. J.; van Hest, J. C. Autonomous movement of platinum-loaded stomatocytes. *Nat. Chem.* **2012**, *4* (4), 268−274.

(22) Abdelmohsen, L. K. E. A.; Williams, D. S.; Pille, J.; Ozel, S. G.; Rikken, R. S. M.; Wilson, D. A.; van Hest, J. C. M. Formation of Well-Defined, Functional Nanotubes via Osmotically Induced Shape Transformation of Biodegradable Polymersomes. *J. Am. Chem. Soc.* **2016**, *138* (30), 9353−9356.

(23) Góźdź, W. T. Spontaneous Curvature Induced Shape Transformations of Tubular Polymersomes. *Langmuir* **2004**, *20* (18), 7385−7391.

(24) Men, Y.; Li, W.; Janssen, G.-J.; Rikken, R. S. M.; Wilson, D. A. Stomatocyte in Stomatocyte: A New Shape of Polymersome Induced via Chemical-Addition Methodology. *Nano Lett.* **2018**, *18* (3), 2081−2085.

(25) Wang, L.-H.; Zhang, Z.-D.; Hong, C.-Y.; He, X.-H.; You, W.; You, Y.-Z. Anion−Dipole Interactions Make the Homopolymers Self-Assemble into Multiple Nanostructures. *Adv. Mater.* **2015**, *27* (20), 3202−3207.

DOI: 10.1021/acs.biomac.9b00924
Biomacromolecules 2020, 21, 89−94

(26) Piazza, R. Interactions and phase transitions in protein solutions. *Curr. Opin. Colloid Interface Sci.* **2000**, *5* (1), 38−43.
(27) Zhang, Y.; Cremer, P. S. The inverse and direct Hofmeister series for lysozyme. *Proc. Natl. Acad. Sci. U. S. A.* **2009**, *106* (36), 15249−15253.
(28) Zhang, Y.; Cremer, P. S. Interactions between macromolecules and ions: the Hofmeister series. *Curr. Opin. Chem. Biol.* **2006**, *10* (6), 658−663.
(29) Jaspers, M.; Rowan, A. E.; Kouwer, P. H. J. Tuning Hydrogel Mechanics Using the Hofmeister Effect. *Adv. Funct. Mater.* **2015**, *25* (41), 6503−6510.
(30) Men, Y.; Li, X.-H.; Antonietti, M.; Yuan, J. Poly-(tetrabutylphosphonium 4-styrenesulfonate): a poly(ionic liquid) stabilizer for graphene being multi-responsive. *Polym. Chem.* **2012**, *3* (4), 871−873.
(31) Men, Y.; Schlaad, H.; Voelkel, A.; Yuan, J. Thermoresponsive polymerized gemini dicationic ionic liquid. *Polym. Chem.* **2014**, *5* (11), 3719−3724.
(32) Men, Y.; Schlaad, H.; Yuan, J. Cationic Poly(ionic liquid) with Tunable Lower Critical Solution Temperature-Type Phase Transition. *ACS Macro Lett.* **2013**, *2* (5), 456−459.
(33) Pegram, L. M.; Record, M. T. Hofmeister Salt Effects on Surface Tension Arise from Partitioning of Anions and Cations between Bulk Water and the Air−Water Interface. *J. Phys. Chem. B* **2007**, *111* (19), 5411−5417.
(34) Erlander, S. R. The production of pseudo-polyelectrolytes in aqueous salt solutions of nonionic polymers. *J. Colloid Interface Sci.* **1970**, *34* (1), 53−64.
(35) Yang, Y.; Huo, H. Investigation of structures of PEO-MgCl2 based solid polymer electrolytes. *J. Polym. Sci., Part B: Polym. Phys.* **2013**, *51* (15), 1162−1174.
(36) Beaudoin, E.; Gourier, C.; Hiorns, R. C.; François, J. Structure and Properties of Hydrophobically End-Capped Poly(ethylene Oxide) Solutions in the Presence of Monovalent and Divalent Cations. *J. Colloid Interface Sci.* **2002**, *251* (2), 398−408.

DOI: 10.1021/acs.biomac.9b00924
Biomacromolecules 2020, 21, 89−94