Article

Modular Platform of Carbohydrates-modified Supramolecular Polymers Based on Dendritic Peptide Scaffolds

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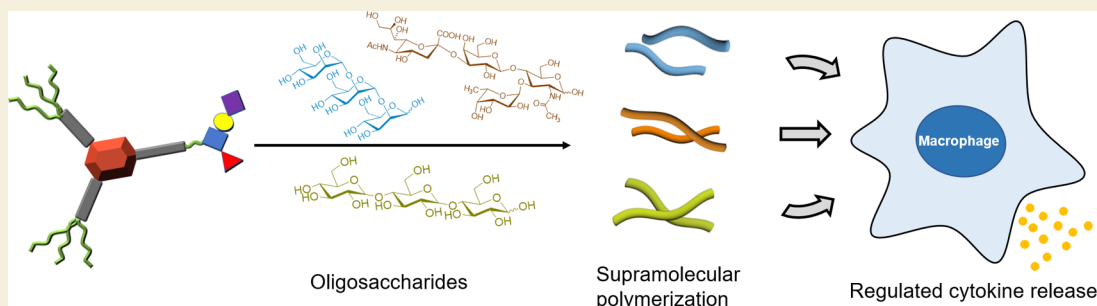Cite This: ACS Polym. Au 2022, 2, 478−485

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Oligosaccharides — Supramolecular polymerization — Regulated cytokine release

ABSTRACT: Glycopeptide supramolecular polymers displaying multivalent carbohydrates are particularly suitable for immune-relevant biomaterials, due to the important functions of carbohydrates in mediating cell-cell communication and modulating immune responses. However, the diversity and complexity of carbohydrates limited the generation of glycopeptide supramolecular monomers. Thereby, a modular platform of presenting various carbohydrates, especially more complex oligosaccharides, is highly desirable but remains underexplored. Here, we first prepared the linear amphiphilic glycopeptides that self-assembled into spherical nanoparticles and worm-like nanoparticles. Furthermore, the dendritic glycopeptides that self-assembled into uniform nanorods were designed to generate modular supramolecular polymers with variable functionality, via redesigning the molecular backbone. With various functional oligosaccharide-modified supramolecular polymers, the in vitro studies further indicated that these polymers were not cytotoxic to macrophages, and significantly modulated the production of proinflammatory cytokines. These findings provide a promising platform to develop supramolecular glycopeptide biomaterials with potential applications in immunomodulation and immunotherapy.

KEYWORDS: supramolecular polymers, self-assembly, carbohydrates, dendritic peptide scaffolds, immune-related biomaterials

■ INTRODUCTION

Supramolecular polymers refer to arrays of repeating units linked by reversible and directional noncovalent interactions such as hydrogen bonding, metal coordination, host-guest interaction, electrostatic interaction or $\pi−\pi$ stacking.[1−5] Although these non-covalent interactions are typically weaker than covalent bonds, they support unified orientation, thereby, generating highly ordered nanostructures.[1−3,6−13] More importantly, the inherently dynamic properties of these interactions enable building blocks to undergo reversible monomer-supramolecular polymer transitions that are essential for many cellular machineries and living systems.[14] For example, the assembly and disassembly of microtubules constructed by tubulins are dynamic and reversible, which is critical for microtubules to support the morphology and movement of cells and control the directional movement of intracellular particles and organelles.[15] Inspired by the important role of supramolecular architectures in biological systems, many artificial supramolecular biomaterials with

impressive properties and functions have been developed for biomedical applications.[16−22] In the past decades, peptide-based assemblies have received extensive attention due to their biomimetic chemical and mechanical properties, high biodegradability, and good biocompatibility.[23−25] Since the first example of supramolecular polymers based on amphiphilic peptide molecules was reported by Stupp and co-workers,[26] various supramolecular interactions have been successively applied to peptide-based self-assembly systems, resulting in spherical, cylindrical, sheet and tubular polypeptide assemblies.[26−34] Meanwhile, well-established synthetic methods of peptide building blocks enable the introduction of other
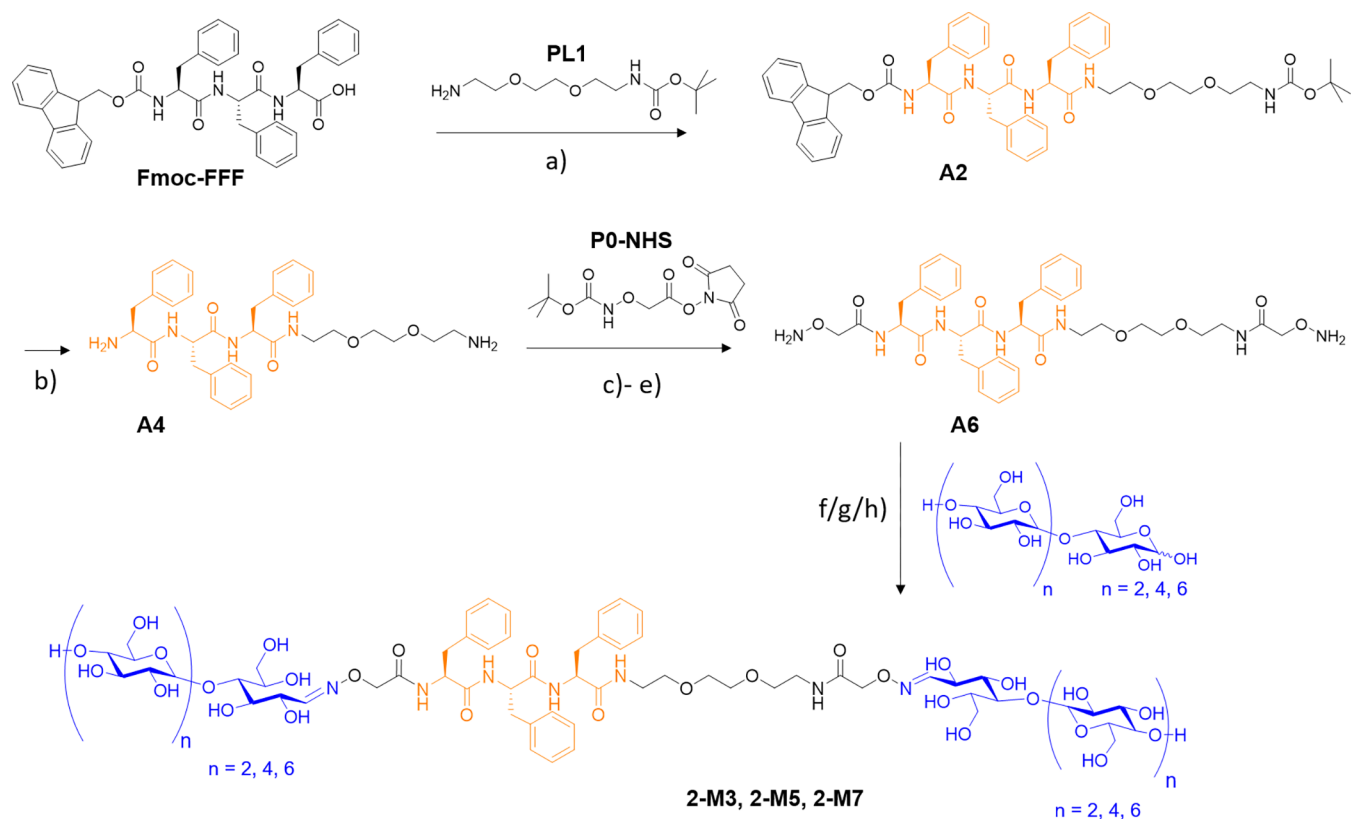
Scheme 1. Scheme for the Synthesis of Linear Oligosaccharides-tripeptide Supramolecular Polymer Monomers[a]

[a](a) N,N-dimethylformamide (DMF), ethyl cyanoglyoxylate-2-oxime, N,N′-diisopropylcarbodiimide (DIC), **PL1**. (b) Trifluoroacetic acid (TFA), dichloromethane (DCM). (c) Piperidine, DMF. (d) DMF, **P0-NHS**. (e) TFA, DCM. (f) DMF, maltotriose. (g) DMF, maltopentaose. (h) DMF, maltoheptaose

biomolecules, such as nucleic acids, lipids, and carbohydrates, for transducing biological signals and modulating cellular behavior.[35−41] Among them, the carbohydrate-peptide supramolecular polymers are especially highlighted by the structural diversity and complexity.[38−42] Moreover, carbohydrates play an important role in mediating intercellular communication and modulating immune responses,[43,44] and small molecular glycopeptides show excellent properties in anti-pathogen and intracellular delivery.[45−48] Therefore, the glycopeptide supramolecular polymers with multivalent presentation of carbohydrates are highly available for the fabrication of immune-relevant biomaterials to address cancer, bacterial and viral pathogens.[42,49] However, due to the complexity of carbohydrates, the generation of appropriate glycopeptide monomers is the foundation as well as the limiting step of fabricating diverse glycopeptide supramolecular polymers.[35−41] Therefore, a modular approach to present the various carbohydrates, especially oligosaccharides that are more complex than simple monosaccharides, is highly desirable and challenging.

On the basis of previous work focusing on a single type of oligosaccharide,[38,40] in order to further develop a versatile platform for oligosaccharide decorated supramolecular polymers, we first designed and synthesized a linear amphiphilic triphenylalanine backbone with two side-arms, which was further modified with a variety of oligosaccharides including maltotriose, maltopentaose and maltoheptaose, through an oxime-mediated strategy.[50] These glycopeptides self-assemble in water to form spherical nanoparticles or worm-like nanoparticles. Then, to further optimize the supramolecular

morphology, dendritic triazine-branched nonaphenylalanines were employed as backbone, as they were able to direct supramolecular polymerization of saccharides into uniform nanorods.[51] Furthermore, these nanorods were functionalized with immune-relevant oligosaccharides such as mannotriose and sialyl Lewis-X. In an *in vitro* study, these carbohydrate-modified nanorods showed no cytotoxicity and significantly modulated inflammatory cytokine released by macrophages, suggesting a potential application in immunomodulation and immunotherapy. Indeed, with the exchangeable carbohydrate motifs, this work will provide a modular platform of supramolecular polymers for exploring the biological and medical application of various complex oligosaccharides.

■ RESULTS AND DISCUSSION

To generate the amphiphilic glycopeptide for supramolecular self-assembly, we first designed the linear monomers in which the oligosaccharides were conjugated to hydrophobic scaffolds as water-soluble moieties. In this monomer, triphenylalanine was selected as the hydrophobic core because of its demonstrated potential to drive supramolecular self-assembly.[52−54] Furthermore, as a model, malto-oligosaccharides were introduced as the hydrophilic shell due to the relatively simple structure and abundant sources. Meanwhile, an oligo(ethylene glycol) chain aimed to improve solubilizing properties and steric requirements, was employed to link the triphenylalanine and oligosaccharides. To prepare the designed oligosaccharide-modified peptide supramolecular polymer, Fmoc-protected triphenylalanine (**Fmoc-FFF**) was first conjugated with the

Boc-protected ethylene glycol chain (**PL1**) via an amidation reaction (**A2**). After deprotection, the purified product (**A4**) was reacted with **P0-NHS**, which further extended the spacer and improved the grafting efficiency of the oligosaccharide moiety. After removal of the Boc group, the obtained tripeptide (**A6**) with two alkoxyamine groups that could be modified with two oligosaccharides at the same time, underwent a condensation reaction with maltotriose, malto-pentaose, and maltoheptaose, separately, under acidic conditions (Scheme 1).[55−57]

The obtained three glycopeptide amphiphiles (**2-M3**, **2-M5**, **2-M7**) were directly dissolved in water and sonicated to prepare an aqueous solution of glycopeptide assemblies. Dynamic light scattering (DLS) was employed to characterize the hydrodynamic diameter of the assembly. As shown in Figure 1a, the assembly formed by the maltotriose-modified
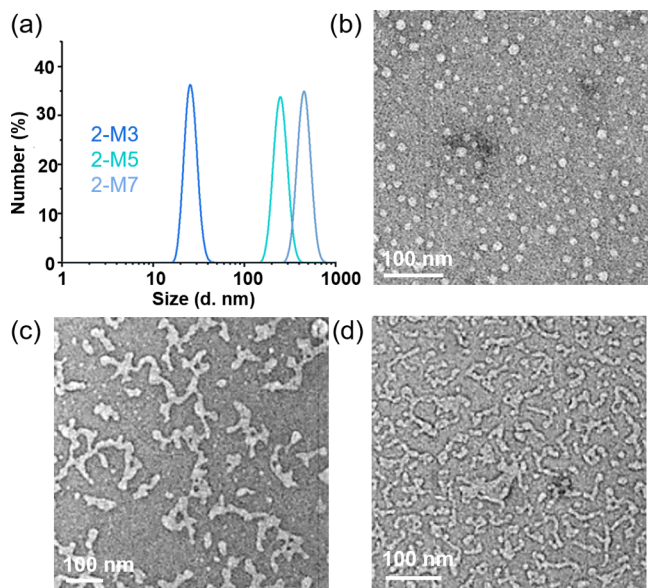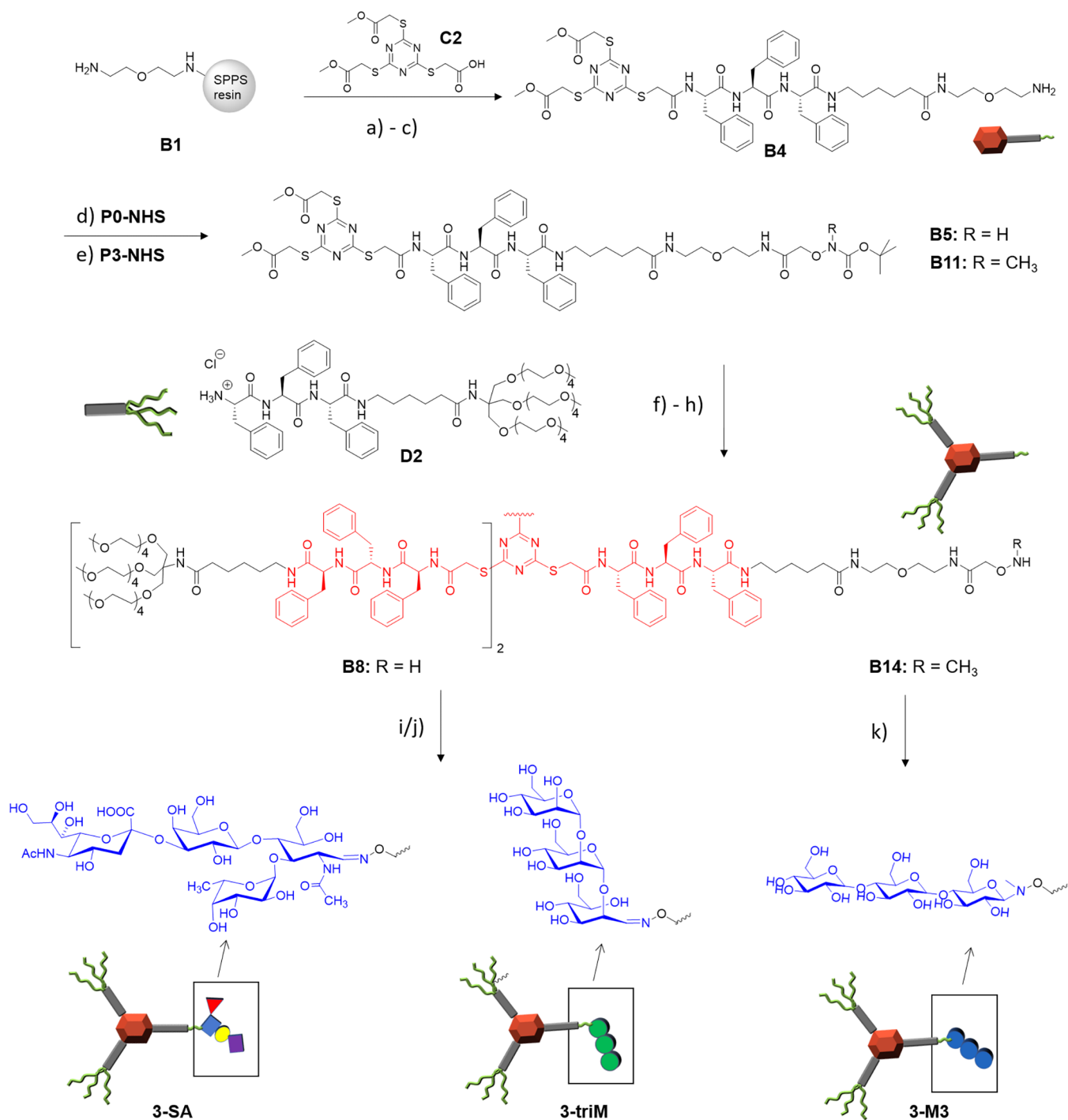

Figure 1. Hydrodynamic sizes and TEM images of linear amphiphilic glycopeptide assemblies. Assemblies were generated at room temperature using 25 μM aqueous solutions (water) of **2-M3**, **2-M5**, and **2-M7** monomers, respectively. (a) Hydrodynamic sizes of **2-M3**, **2-M5**, and **2-M7** assemblies. (b) Spherical nanoparticles of **2-M3**. (c) Worm-like nanoparticles of **2-M5**. (d) Worm-like nanoparticles of **2-M7**.

tripeptide (**2-M3**) assembled in aqueous solution exhibited the smallest hydrodynamic diameter (about 20 nm), while the maltopentaose (**2-M5**) and maltoheptaose-modified tripeptides (**2-M7**) generated larger assemblies with hydrodynamic diameters (about 240 nm and 450 nm, respectively). This suggests that oligosaccharide moieties of different lengths could lead to diverse morphologies of the assemblies. Therefore, transmission electron microscopy (TEM) was employed to further investigate the morphology of the assembly. The observed images showed that the **2-M3** was self-assembled into spherical nanoparticles with a diameter of about 20 nm (Figure 1b), which was consistent with the results of DLS. While **2-M5** and **2-M7** generated worm-like nanoparticles with sizes exceeding 200 nm (Figure 1c,d). It suggested that the linear malto-oligosaccharide-modified triphenylalanines were capable of supramolecular polymerization, albeit with less regularity of assemblies. Furthermore, since these three glycopeptide molecules mainly differed in the

lengths of malto-oligosaccharide repeats, we speculated that the carbohydrate-carbohydrate interactions and steric hindrance were very different in **2-M3**, **2-M5**, and **2-M7**, thereby affecting the morphological transition from spherical nanoparticles to worm-like nanoparticles.[58−60]

Given that the linear glycopeptides were able to form spherical and worm-like nanoparticles, we further attempted to obtain more regular supramolecular polymers by redesigning the molecular backbone. Therefore, the dendritic "three-branched" structure was selected as the scaffold, as the molecules might provide stronger $\pi-\pi$ stacking between the phenylalanine moieties, and more pronounced directionality in the supramolecular organisation.[61−63] Furthermore, considering that increasing the content of phenylalanine will result in a strongly hydrophobic structure, two hydrophilic dendritic motifs containing three tetraethylene glycol arms were designed to increase its solubility in water and colloid-stability in supramolecular polymers. Meanwhile, only one arm was used to introduce the oligosaccharide groups, as the linear assembly results indicated that carbohydrate-carbohydrate interactions and steric hindrance may arise when multiple modification sites were involved, affecting the self-assembled morphology (Figure 1). Structurally, it contained three critical moieties: the dendritic glycopeptides containing three triphenylalanine arms, could be conjugated with two dendritic hydrophilic arms and one oligosaccharide hydrophilic arm; as a core to generate C_3 or C_2 symmetry, triazine was selected for its potential in fabricating supramolecular polymers; as another indispensable element, mannotriose and sialyl-Lewis X were employed to generate functionalized dendritic glycopeptides. Among the oligosaccharide moieties, the mannoside residue of mannotriose is known for its targeting properties to antigen presenting cells (such as macrophages and dendritic cells) of the innate immune system.[42−44] Notably, via binding to the C-type lectin receptors (CLRs) on antigen presenting cells, various mannose-modified macromolecular or supramolecular scaffolds have been reported to enhance immune responses in cancer immunotherapy.[42−44] Conversely, sialyl-Lewis X is a high-affinity ligand for selectins (E-, P-, and L-selectin), and has the ability to inhibit CD62-mediated neutrophil recruitment to sites of inflammation.[64,65] Furthermore, sialyl-Lewis X is overexpressed in many cancer cells and closely related to tumor invasion and metastasis.[66−68]

The backbone of dendritic glycopeptide was synthesized by solid phase peptide synthesis (SPPS). Starting from a resin loaded with bis-(2-aminoethyl)-ether (**B1**), **B1** was elongated using the Fmoc-aminohexanoic acid and 3 × Fmoc-L-Phe-OH to obtained **B2**. Meanwhile, the core of triazine (**C2**) and the hydrophilic dendritic motifs containing three triethylene glycol arms (**D2**) was synthesized, according to previous report.[51,69] The prepared triazine **C2** with two substituents being methyl-2-thioglycolate and one substituent being thioglycolic acid was conjugated with the **B2** in DMF to prepare **B3**. Then the **B4** could be obtained by cleavage from the resin using a mixture of TFA/TIPS/H$_2$O (9.5/0.25/0.25). The purified product (**B4**) was reacted with **P3-NHS** or **P0-NHS** to extend the spacer (**B5** or **B11**), and followed by methyl ester hydrolysis with 0.1 M LiOH in tetrahydrofuran (THF) to generate dicarboxylated **B6** or **B12**. The **P3-NHS** formed an *N*-methyl alkoxyamine group after deprotection to maintain the cyclic form of the saccharide after conjugation.[70] However, in the model reaction, the *N*-methyl alkoxyamine-modified dendritic peptide scaffolds (**B14**) were employed to conjugate with maltotriose, resulting

Scheme 2. Scheme for the Synthesis of Dendritic Oligosaccharides-tripeptide Supramolecular Polymer Monomers[a]

[a](a) Fmoc-aminohexanoic acid, Fmoc-L-Phe-OH, HBTU, HOBt, DIPEA, piperidine, DMF. (b) HATU, HOAt, DMF, **C2**. (c) TFA/TIPS/H2O 9.5:0.25:0.25. (d) DMF, **P0-NHS**. (e) DMF, **P3-NHS**. (f) MeOH, THF, LiOH 0.1 M. (g) HOBt, DMF, PyBOP, DIPEA, **D2**. (h) TFA, DCM. (i) DMF, sialyl Lewis-X. (j) DMF, mannotriose. (k) DMF, maltotriose.

in low yields of **3-M3**. Therefore, **P0-NHS**, which can form alkoxyamine groups after deprotection, was selected as a functional group to improve the reaction efficiency with oligosaccharides. Furthly, **B6** was double amidated by hydrophilic dendritic motifs **D2** to give **B7**, followed by deprotection leading to the formation of alkoxyamine group (**B8**). In the final step, the alkoxyamine group was conjugated with oligosaccharides (**3-triM** and **3-SA**) in DMF at 45 °C (Scheme 2).[55−57]

From previous reports,[51,69,71,72] we could tentatively speculate that this designed glycopeptide may direct supramolecular polymerization into nanorod-like polymers in aqueous solution. Therefore, the **3-M3** with maltotriose as a model of C_3 symmetric glycopeptides was first dissolved in aqueous solution and sonicated to prepare assemblies. The DLS results suggested the generation of assemblies at 200 nm (Figure S1). TEM further confirmed the self-assembly of **3-M3** into nanorod-like materials (Figure S1). It was confirmed that

the oligosaccharide-modified peptides with C_3 symmetrical cores were capable of preparing regular supramolecular polymers. Similarly, the functional oligosaccharide-peptides, **3-triM** and **3-SA**, were also prepared by dissolution and sonication. The DLS results showed that hydrodynamic diameter of **3-triM** assembly was about 200 nm (Figure 2a),
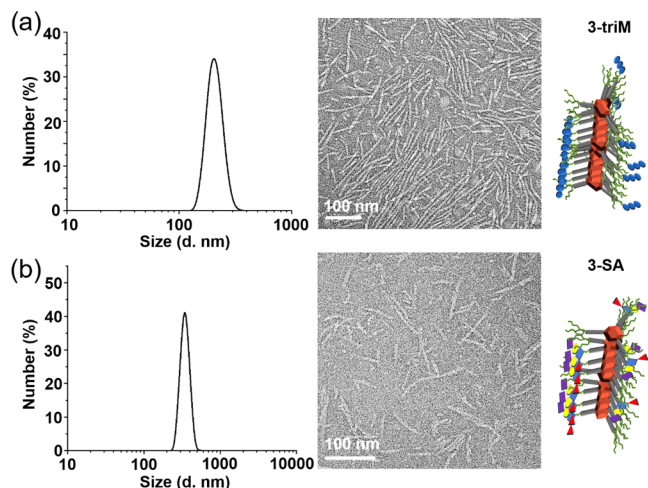
Figure 2. Hydrodynamic sizes and TEM images of dendritic glycopeptide assemblies. Assemblies were generated at room temperature using 25 μM aqueous solutions (water) of **3-triM**, and **3-SA** monomers, respectively. Nanorod-shaped supramolecular polymers of (a) **3-triM**, and (b) **3-SA**.

and TEM results showed that it self-assembled in water to form uniform supramolecular nanorods with an average diameter of 7 nm and an average length of 114 nm (Figure 2a). Meanwhile, the **3-SA** could self-assemble in solution to form a hydrodynamic diameter of about 300 nm (Figure 2b). In the TEM experiments, the assembled morphology was a nanorod structure with an average diameter of 7 nm and an average length of 66 nm (Figure 2b). Similar supramolecular architectures prepared from **3-M3**, **3-triM**, and **3-SA** suggested that the dendritic glycopeptides were able to generate repeatable morphology of supramolecular polymers even with diverse oligosaccharide modules. To further evaluate the universal applicability of this supramolecular monomer including the possibility of imaging functions, an alternative route for synthesizing a fluorescent label (FITC) was investigated. The **B4** and FITC were dissolved in DMF and catalyzed with TEA overnight, followed by demethylation and amidation with **D2**. More importantly, the TEM confirmed that the **3-FITC** self-assembled into nanorods similar to

glycopeptides (Figure S2), indicating the high diversity and replaceability of oligosaccharide moieties.

Encouraged by these results, the application of these glycopeptide supramolecular polymers as immune-related biomaterials was tested on RAW264.7 macrophages. As a crucial factor limiting the applicability, the cytotoxicity of the assemblies was first examined by *in vitro* incubation with RAW264.7 at serial dilution concentrations ranging from 125 to 7.5 μg/mL for 24 h. It showed that all assemblies had no obvious cytotoxicity even at concentrations up to 125 μg/mL (Figure S3), suggesting good biocompatibility of these oligosaccharide-modified supramolecular polymers. Subsequently, RAW264.7 cells were incubated with these assemblies at final concentrations of 10 and 100 μg/mL, to further evaluate their effects on macrophage bioactivity. After 24h of incubation, proinflammatory cytokines including tumor necrosis factor α (TNF-α) and interleukin 6 (IL-6) in the supernatant were detected. In most samples, the secretion of TNF-α and IL-6 were significantly increased with increasing concentration of assemblies (Figures 3 and S3). The results suggest that the supramolecular glycopeptide arrays provide significant immunostimulatory effects *in vitro*. Interestingly, compared with other glycopeptides, the **3-SA** self-assembled nanorods had negligible increases in the production of TNF-α and IL-6 (Figure 3). Furthermore, the reduced secretion of TNF-α and IL-6 at 100 μg/mL confirmed a concentration-dependent behavior (Figure 3). These results suggested that the sialyl-Lewis X modified supramolecular polymer might be able to suppress rather than induce immune responses. Although the detailed biological mode of action needs further investigations, these promising preliminary results support the potential of supramolecular glycopeptide polymers as immunomodulating biomaterials.

■ CONCLUSIONS

In summary, amphiphilic glycopeptide molecules had been designed and synthesized, in which the linear glycopeptides could self-assemble into spherical nanoparticles and worm-like nanoparticles in aqueous solution. Furthermore, in order to prepare a modular glycopeptide monomer that could easily fabricate similar supramolecular polymers with various complex oligosaccharides, via redesigning the peptide backbone, a dendritic backbone was employed to synthesize more versatile glycopeptide molecules. These dendritic glycopeptide monomers were able to perform self-assembly in aqueous solution to generate uniform supramolecular nanorods. More importantly, the similar morphologies in diverse oligosaccharide-modified supramolecular polymers confirmed the high adaptability to different oligosaccharide moieties. Finally, the
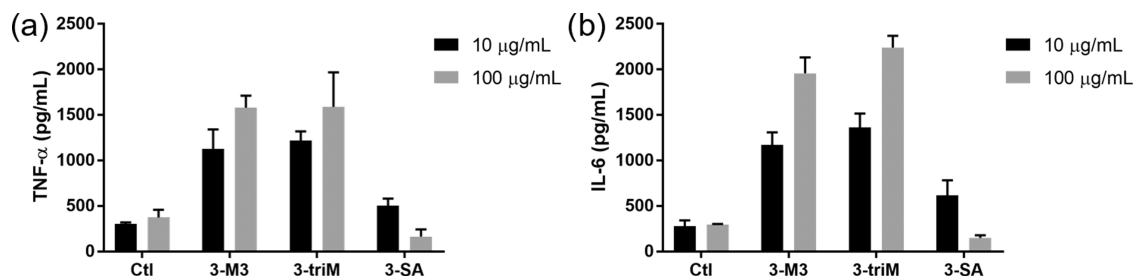
Figure 3. Cytokine release of macrophages induced by the dendritic glycopeptide supramolecular polymers. The production of (a) TNF-α, and (b) IL-6 after 24h incubation with **3-M3**, **3-triM**, and **3-SA**. The control group used PBS buffer.

incubation of supramolecular polymers with macrophages significantly affected the expression of proinflammatory cytokines. These findings provide a versatile strategy for conjugation of various oligosaccharides onto the same backbone via the same chemical method, resulting in comparable morphologies at nanoscale, which will be a promising platform for the development of artificial self-assembled glycopeptide biomaterials, and show potential applications in immunomodulation and immunotherapy.

■ ASSOCIATED CONTENT

ⓢ Supporting Information

The Supporting Information is available free of charge at https://pubs.acs.org/doi/10.1021/acspolymersau.2c00032.

> Abbreviations, extended materials and characterization methods (SI), Hydrodynamic sizes and TEM images of **3-M3** assemblies (Figure S1), TEM images of **3-FITC** assemblies (Figure S2), cytotoxic effect of glycopeptide supramolecular polymers on macrophages (Figure S3), cytokine release of macrophages induced by the two-branched glycopeptide supramolecular polymers (Figure S4), and detailed reaction process (PDF)

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CRediT: **Moritz Urschbach** data curation (equal); **David Straßburger** data curation (equal).

Notes

The authors declare no competing financial interest.

■ REFERENCES

(1) Lehn, J. M. Supramolecular polymer chemistry-scope and perspectives. *Polym. Int.* **2002**, *51*, 825−839.

(2) Aida, T.; Meijer, E. W.; Stupp, S. I. Functional supramolecular polymers. *Science* **2012**, *335*, 813−817.

(3) Tayi, A. S.; Kaeser, A.; Matsumoto, M.; Aida, T.; Stupp, S. I. Supramolecular ferroelectrics. *Nat. Chem.* **2015**, *7*, 281−294.

(4) Zhang, X.; Wang, L.; Xu, J.; Chen, D.; Shi, L.; Zhou, Y.; Shen, Z. Polymeric Supramolecular Systems: Design, Assembly and Functions. *Acta Polym. Sin.* **2019**, *50*, 973−987.

(5) Zhang, X. Supramolecular Polymer Chemistry: Past, Present, and Future. *Chin. J. Polym. Sci.* **2022**, *40*, 541−542.

(6) Dong, S.; Luo, Y.; Yan, Y.; Zheng, B.; Ding, X.; Yu, Y.; Ma, Z.; Zhao, Q.; Huang, F. A dual-responsive supramolecular polymer gel formed by crown ether based molecular recognition. *Angew. Chem., Int. Ed.* **2011**, *50*, 1905−1909.

(7) Wijnands, S. P. W.; Engelen, W.; Lafleur, R. P. M.; Meijer, E. W.; Merkx, M. Controlling protein activity by dynamic recruitment on a supramolecular polymer platform. *Nat. Commun.* **2018**, *9*, No. 65.

(8) Jung, S. H.; Bochicchio, D.; Pavan, G. M.; Takeuchi, M.; Sugiyasu, K. A Block Supramolecular Polymer and Its Kinetically Enhanced Stability. *J. Am. Chem. Soc.* **2018**, *140*, 10570−10577.

(9) Shao, L.; Sun, J.; Hua, B.; Huang, F. An AIEE fluorescent supramolecular cross-linked polymer network based on pillar[5]arene host-guest recognition: Construction and application in explosive detection. *Chem. Commun.* **2018**, *54*, 4866−4869.

(10) Qiao, F.; Zhang, L.; Lian, Z.; Yuan, Z.; Yan, C.; Zhou, S.; Zhou, Z.; Xing, L. Construction of artificial light-harvesting systems in aqueous solution: Supramolecular polymers based on host-enhanced $\pi-\pi$ interaction with aggregation-induced emission. *J. Photochem.* **2018**, *355*, 419−424.

(11) Zhang, Z.; Luo, Y.; Chen, J.; Dong, S.; Yu, Y.; Ma, Z.; Huang, F. Formation of Linear Supramolecular Polymers That Is Driven by C-H···π Interactions in Solution and in the Solid State. *Angew. Chem., Int. Ed.* **2011**, *50*, 1397−1401.

(12) Ji, X.; Yao, Y.; Li, J.; Yan, X.; Huang, F. A supramolecular cross-linked conjugated polymer network for multiple fluorescent sensing. *J. Am. Chem. Soc.* **2013**, *135*, 74−77.

(13) Lange, R. F. M.; Van Gurp, M.; Meijer, E. W. Hydrogen-bonded supramolecular polymer networks. *J. Polym. Sci., Part A: Polym. Chem.* **1999**, *37*, 3657−3670.

(14) Tu, Y.; Peng, F.; Adawy, A.; Men, Y.; Abdelmohsen, L. K. E. A.; Wilson, D. A. Mimicking the cell: bio-inspired functions of supramolecular assemblies. *Chem. Rev.* **2016**, *116*, 2023−2078.

(15) Goodson, H. V.; Jonasson, E. M. Microtubules and Microtubule-Associated Proteins. *Cold Spring Harbor Perspect. Biol.* **2018**, *10*, No. a022608.

(16) Yan, X.; Wang, F.; Zheng, B.; Huang, F. Stimuli-responsive supramolecular polymeric materials. *Chem. Soc. Rev.* **2012**, *41*, 6042−6065.

(17) Burnworth, M.; Tang, L.; Kumpfer, J. R.; Duncan, A. J.; Beyer, F. L.; Fiore, G. L.; Rowan, S. J.; Weder, C. Optically healable supramolecular polymers. *Nature* **2011**, *472*, 334−337.

(18) Dong, R.; Zhou, Y.; Huang, X.; Zhu, X.; Lu, Y.; Shen, J. Functional supramolecular polymers for biomedical applications. *Adv. Mater.* **2015**, *27*, 498−526.

(19) Radvar, E.; Azevedo, H. S. Supramolecular Peptide/Polymer Hybrid Hydrogels for Biomedical Applications. *Macromol. Biosci.* **2019**, *19*, No. 1800221.

(20) Webber, M. J.; Appel, E. A.; Meijer, E. W.; Langer, R. Supramolecular biomaterials. *Nat. Mater.* **2016**, *15*, 13−26.

(21) Petkau-Milroy, K.; Sonntag, M. H.; Brunsveld, L. Modular Columnar Supramolecular Polymers as Scaffolds for Biomedical Applications. *Chem. - Eur. J.* **2013**, *19*, 10786−10793.

(22) Li, Y.; Tian, R.; Xu, J.; Hou, C.; Luo, Q.; Liu, J. Protein Supramolecular Polymers and Their Applications. *Acta Polym. Sin.* **2022**, *53*, 1−23.

(23) Makam, P.; Gazit, E. Minimalistic peptide supramolecular co-assembly: expanding the conformational space for nanotechnology. *Chem. Soc. Rev.* **2018**, *47*, 3406−3420.

(24) Dikecoglu, F. B.; Topal, A. E.; Ozkan, A. D.; Tekin, E. D.; Tekinay, A. B.; Guler, M. O.; A, D. Force and time-dependent self-assembly, disruption and recovery of supramolecular peptide amphiphile nanofibers. *Nanotechnology* **2018**, *29*, No. 285701.

(25) Adler-Abramovich, L.; Gazit, E. The physical properties of supramolecular peptide assemblies: from building block association to technological applications. *Chem. Soc. Rev.* **2014**, *43*, 6881−6893.

(26) Hartgerink, J. D.; Beniash, E.; Stupp, S. I. Self-assembly and mineralization of peptide-amphiphile nanofibers. *Science* **2001**, *294*, 1684−1688.

(27) Handelman, A.; Beker, P.; Amdursky, N.; Rosenman, G. Physics and engineering of peptide supramolecular nanostructures. *Phys. Chem. Chem. Phys.* **2012**, *14*, 6391−6408.

(28) Jiao, D.; Geng, J.; Loh, X. J.; Das, D.; Lee, T.; Scherman, O. A. Supramolecular peptide amphiphile vesicles through host-guest complexation. *Angew. Chem., Int. Ed.* **2012**, *51*, 9633−9637.

(29) Lim, Y.-b.; Lee, E.; Lee, M. Controlled bioactive nanostructures from self-assembly of peptide building blocks. *Angew. Chem., Int. Ed.* **2007**, *119*, 9169−9172.

(30) Kol, N.; Adler-Abramovich, L.; Barlam, D.; Shneck, R. Z.; Gazit, E.; Rousso, I. Self-assembled peptide nanotubes are uniquely rigid bioinspired supramolecular structures. *Nano Lett.* **2005**, *5*, 1343−1346.

(31) Kumaraswamy, P.; Lakshmanan, R.; Sethuraman, S.; Krishnan, U. M. Self-assembly of peptides: influence of substrate, pH and medium on the formation of supramolecular assemblies. *Soft Matter* **2011**, *7*, 2744−2754.

(32) Hamley, I. W. Peptide nanotubes. *Angew. Chem., Int. Ed.* **2014**, *53*, 6866−6881.

(33) Krieg, E.; Bastings, M. M. C.; Besenius, P.; Rybtchinski, B. Supramolecular polymers in aqueous media. *Chem. Rev.* **2016**, *116*, 2414−2477.

(34) Görbitz, C. H.; Rise, F. Template-directed supramolecular assembly of a new type of nanoporous peptide-based material. *J. Pept. Sci.* **2008**, *14*, 210−216.

(35) Ustun Yaylaci, S.; Ekiz, M. S.; Arslan, E.; Can, N.; Kilic, E.; Ozkan, H.; Orujalipoor, I.; Ide, S.; Tekinay, A. B.; Guler, M. O. Supramolecular GAG-like self-assembled glycopeptide nanofibers induce chondrogenesis and cartilage regeneration. *Biomacromolecules* **2016**, *17*, 679−689.

(36) Liu, J.; Sun, Z.; Yuan, Y.; Tian, X.; Liu, X.; G, D.; Yang, Y.; Yuan, L.; Lin, H.; X, L. Peptide glycosylation generates supra-molecular assemblies from glycopeptides as biomimetic scaffolds for cell adhesion and proliferation. *ACS Appl. Mater. Interfaces* **2016**, *8*, 6917−6924.

(37) Lee, S. S.; Fyrner, T.; Chen, F.; Alvarez, Z.; Sleep, E.; Chun, D. S.; Weiner, J. A.; Cook, R. W.; Freshman, R. D.; Schallmo, M. S.; Katchko, K. M.; Schneider, A. D.; Smith, J. T.; Yun, C.; Singh, G.; Hashmi, S. Z.; McClendon, M. T.; Yu, Z.; Stock, S. R.; Hsu, W. K.; Hsu, E. L.; Stupp, S. I. Sulfated glycopeptide nanostructures for multipotent protein activation. *Nat. Nanotechnol.* **2017**, *12*, 821−829.

(38) Liu, Y.; Zhang, Y.; Wang, Z.; Wang, J.; Wei, K.; Chen, G.; Jiang, M. Building Nanowires from Micelles: Hierarchical Self-Assembly of Alternating Amphiphilic Glycopolypeptide Brushes with Pendants of High-Mannose Glycodendron and Oligophenylalanine. *J. Am. Chem. Soc.* **2016**, *138*, 12387−12394.

(39) Restuccia, A.; Seroski, D. T.; Kelley, K. L.; O'Bryan, C. S.; Kurian, J. J.; Knox, K. R.; Farhadi, S. A.; Angelini, T. E.; Hudalla, G. A. Hierarchical self-assembly and emergent function of densely glycosylated peptide nanofibers. *Commun. Chem.* **2019**, *2*, No. 53.

(40) Liu, R.; Zhang, R.; Li, L.; Kochovski, Z.; Yao, L.; Nieh, M. P.; Lu, Y.; Shi, T.; Chen, G. A Comprehensive Landscape for Fibril Association Behaviors Encoded Synergistically by Saccharides and Peptides. *J. Am. Chem. Soc.* **2021**, *143*, 6622−6633.

(41) Brito, A.; Salma, K.; Rui, L. R.; Rein, V. U.; Ricardo, A. P.; Iva, P. Carbohydrate amphiphiles for supramolecular biomaterials: Design, self-assembly, and applications. *Chem* **2021**, *7*, 2943−2964.

(42) Su, L.; Feng, Y.; Wei, K.; Xu, X.; Liu, R.; Chen, G. Carbohydrate-Based Macromolecular Biomaterials. *Chem. Rev.* **2021**, *121*, 10950−11029.

(43) Keumatio Doungstop, B. C.; van Vliet, S. J.; van Ree, R.; de Jong, E. C.; van Kooyk, Y. Carbohydrates in allergy: from disease to novel immunotherapies. *Trends Immunol.* **2021**, *42*, 635−648.

(44) Mantuano, N. R.; Natoli, M.; Zippelius, A.; Laubli, H. Tumor-associated carbohydrates and immunomodulatory lectins as targets for cancer immunotherapy. *J. Immunother. Cancer* **2020**, *8*, No. e001222.

(45) Motiei, L.; Rahimipour, S.; Thayer, D. A.; Wong, C.; Ghadiri, M. R. Antibacterial cyclic D, L-α-glycopeptides. *Chem. Commun.* **2009**, 3693−3695.

(46) Lécorché, P.; Walrant, A.; Burlina, F.; Dutot, L.; Sagan, S.; Mallet, J.; Desbat, B.; Chassaing, G.; Alves, I. D.; Lavielle, S. Cellular uptake and biophysical properties of galactose and/or tryptophan containing cell-penetrating peptides. *Biochim. Biophys. Acta* **2012**, *1818*, 448−457.

(47) Morelli, P.; Bartolami, E.; Sakai, N.; Matile, S. Glycosylated Cell-Penetrating Poly(disulfide)s: Multifunctional Cellular Uptake at High Solubility. *Helv. Chim. Acta* **2018**, *101*, No. e1700266.

(48) Gallego, I.; Montenegro, J. Glycan shields for penetrating peptides. *Chem. Commun.* **2022**, *58*, 1394−1397.

(49) Feng, Y.; Li, L.; Du, Q.; Gou, L.; Zhang, L.; Chai, Y.; Zhang, R.; Shi, T.; Chen, G. Polymorphism of Kdo-Based Glycolipids: The Elaborately Determined Stable and Dynamic Bicelles. *CCS Chem.* **2022**, *4*, 2228−2238.

(50) Gallego, I.; Rioboo, A.; Reina, J. J.; Díaz, B.; Canales, Á.; Cañada, F. J.; Guerra-Varela, J.; Sánchez, L.; Montenegro, J. Glycosylated Cell-Penetrating Peptides (GCPPs). *ChemBioChem* **2019**, *20*, 1400−1409.

(51) Straßburger, D.; Stergiou, N.; Urschbach, M.; Yurugi, H.; Spitzer, D.; Schollmeyer, D.; Schmitt, E.; Besenius, P. Mannose-Decorated Multicomponent Supramolecular Polymers Trigger Effective Uptake into Antigen-Presenting Cells. *ChemBioChem* **2018**, *19*, 912−916.

(52) Mayans, E.; Casanovas, J.; Gil, A. M.; Jiménez, A. I.; Cativiela, C.; Puiggalí, J.; Alemán, C. Diversity and Hierarchy in Supramolecular Assemblies of Triphenylalanine: From Laminated Helical Ribbons to Toroids. *Langmuir* **2017**, *33*, 4036−4048.

(53) Brown, N.; Lei, J.; Zhan, C.; Shimon, L. J. W.; Adler-Abramovich, L.; Wei, G.; Gazit, E. Structural Polymorphism in a Self-Assembled Tri-Aromatic Peptide System. *ACS Nano* **2018**, *12*, 3253−3262.

(54) Han, T. H.; Ok, T.; Kim, J.; Shin, D. O.; Ihee, H.; Lee, H.; Kim, S. O. Bionanosphere Lithography via Hierarchical Peptide Self-Assembly of Aromatic Triphenylalanine. *Small* **2010**, *6*, 945−951.

(55) Ulrich, S.; Boturyn, D.; Marra, A.; Renaudet, O.; Dumy, P. Oxime Ligation: A Chemoselective Click-Type Reaction for Accessing Multifunctional Biomolecular Constructs. *Chem. - Eur. J.* **2014**, *20*, 34−41.

(56) Kölmel, D. K.; Kool, E. T. Oximes and Hydrazones in Bioconjugation: Mechanism and Catalysis. *Chem. Rev.* **2017**, *117*, 10358−10376.

(57) Booth, R.; Insua, I.; Bhak, G.; Montenegro, J. Self-assembled micro-fibres by oxime connection of linear peptide amphiphiles. *Org. Biomol. Chem.* **2019**, *17*, 1984−1991.

(58) de la Fuente, J. M.; Penadés, S. Understanding carbohydrate-carbohydrate interactions by means of glyconanotechnology. *Glycoconjugates* **2004**, *21*, 149−163.

(59) Fuss, M.; Luna, M.; Alcántara, D.; Fuente, J. M.; Enríquez-Navas, P. M.; Angulo, J.; Penadés, S.; Briones, F. Carbohydrate-Carbohydrate Interaction Prominence in 3D Supramolecular Self-Assembly. *J. Phys. Chem. B* **2008**, *112*, 11595−11600.

(60) Ojeda, R.; de Paz, J. L.; Barrientos, A. G.; Martín-Lomas, M.; Penadés, S. Preparation of multifunctional glyconanoparticles as a platform for potential carbohydrate-based anticancer vaccines. *Carbohydr. Res.* **2007**, *342*, 448−459.

(61) Su, H.; Jansen, S. A. H.; Schnitzer, T.; Weyandt, E.; Rösch, A. T.; Liu, J.; Vantomme, G.; Meijer, E. W. Unraveling the Complexity of Supramolecular Copolymerization Dictated by Triazine−Benzene Interactions. *J. Am. Chem. Soc.* **2021**, *143*, 17128−17135.

(62) Lou, X.; Schoenmakers, S. M. C.; van Dongen, J. L. J.; Garcia-Iglesias, M.; Casellas, N. M.; Romera, M. F. C.; Sijbesma, R. P.; Meijer, E. W.; Palmans, A. R. A. Elucidating dynamic behavior of synthetic supramolecular polymers in water by hydrogen/deuterium exchange mass spectrometry. *J. Polym. Sci.* **2021**, *59*, 1151−1161.

(63) Chidchob, P.; Jansen, S. A. H.; Meskers, S. C. J.; Weyandt, E.; van Leest, N. P.; de Bruin, B.; Palmans, A. R. A.; Vantomme, G.; Meijer, E. W. Supramolecular Systems Containing B−N Frustrated Lewis Pairs of Tris(pentafluorophenyl)borane and Triphenylamine Derivatives. *Org. Mater.* **2021**, *03*, 174−183.

(64) Polley, M. J.; Phillips, M. L.; Wayner, E.; Nudelman, E.; Singhal, A. K.; Hakomori, S.; Paulson, J. C. CD62 and Endothelial Cell-Leukocyte Adhesion Molecule 1 (ELAM-1) Recognize the Same Carbohydrate Ligand, sialyl-Lewis X. *Proc. Natl. Acad. Sci. U.S.A.* **1991**, *88*, 6224−6228.

(65) Lasky, L. A. Selectins: Interpreters of Cell-Specific Carbohydrate Information During Inflammation. *Science* **1992**, *258*, 964−969.

(66) Takada, A.; Ohmori, K.; Yoneda, T.; Tsuyuoka, K.; Hasegawa, A.; Kiso, M.; Kannagi, R. Contribution of carbohydrate antigens sialyl Lewis A and sialyl Lewis X to adhesion of human cancer cells to vascular endothelium. *Cancer Res.* **1993**, *53*, 354−361.

(67) Trinchera, M.; Aronica, A.; Dall'Olio, F. Selectin ligands sialyl-Lewis a and sialyl-Lewis x in gastrointestinal cancers. *Biology* **2017**, *6*, 16.

(68) Nguyen, M.; Strubel, N. A.; Bischoff, J. A role for sialyl Lewis-X/A glycoconjugates in capillary morphogenesis. *Nature* **1993**, *365*, 267−269.

(69) Berac, C. M.; Zengerling, L.; Straβburger, D.; Otter, R.; Urschbach, M.; Besenius, P. Evaluation of Charge-Regulated Supramolecular Copolymerization to Tune the Time Scale for Oxidative Disassembly of β-Sheet Comonomers. *Macromol. Rapid Commun.* **2020**, *41*, No. 1900476.

(70) Peri, F.; Dumy, P.; Mutter, M. Chemo- and stereoselective glycosylation of hydroxylamino derivatives: a versatile approach to glycoconjugates. *Tetrahedron* **1998**, *54*, 12269−12278.

(71) Ahlers, P.; Fischer, K.; Spitzer, D.; Besenius, P. Dynamic light scattering investigation of the kinetics and fidelity of supramolecular copolymerizations in water. *Macromolecules* **2017**, *50*, 7712−7720.

(72) Appel, R.; Fuchs, J.; Tyrrell, S. M.; Korevaar, P. A.; Stuart, M. C. A.; Voets, I. K.; Schönhoff, M.; Besenius, P. Steric constraints induced frustrated growth of supramolecular nanorods in water. *Chem. - Eur. J.* **2015**, *21*, 19257−19264.