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FOCUS REVIEW

Stimuli-responsive supramolecular systems guided by chemical reactions

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Abstract

This focus review describes the development of stimuli-responsive supramolecular systems, emphasizing molecular design approach used to construct constituent molecular hybrids comprising chemically reactive small molecules and potentially functional biomolecules or bio-related molecules. The construction of stimuli-responsive supramolecular hydrogels and G-quadruplex-forming nucleic acids, despite the lack of substantial similarity at the supramolecular systems level, highlights the versatile applicability of the molecular design approach for exploring a variety of supramolecular (bio)materials endowed with tailor-made stimuli responsiveness.

Introduction

The introduction of stimuli responsive features into biomolecules or bio-related molecules enables the control of their functions in response to desired stimuli. This molecular design is important since the newly developed molecules and the corresponding materials endowed with tailor-made stimuli responsiveness are potentially useful tools for valuable (bio) applications [1–4]. Molecular designs that introduce photoswitching functions by taking advantage of *trans–cis* isomerization [5] or photo-caged groups [6] have been extensively explored. When introducing photo-caged groups (photoremovable chemical groups) into a target biomolecule, the activities of the biomolecule are temporarily masked; then, these activities are restored by an external stimulus, i.e., photoirradiation, to remove the introduced chemical groups [6]. In contrast, molecular designs that introduce other stimuli-responsive features, such as chemical stimuli responsiveness, which can be utilized under biological conditions, are still in their infancy. Inspired by the molecular design taking advantage of photo-caged groups, we have focused on modular and hybrid molecular designs by coupling the chemical reactivity of artificial, small molecular groups and the functions and/or behaviors of biomolecules or bio-related molecules [7–17]. This molecular design approach has been applied to develop the following (i) stimuli-responsive supramolecular hydrogels based on self-assembling molecules [8–13] and (ii) chemical stimuli-responsive G-quadruplex-forming nucleic acids [15, 17], which are primarily discussed here. Research background of the two topics is summarized at the beginning of each section.

Stimuli-responsive supramolecular hydrogels controlled by chemical reactions

Supramolecular hydrogels comprising interwoven supramolecular nanostructures with a primarily fibrous morphology are a unique soft matter that is rapidly expanding to explorations of (bio)applications due to their promising prospects as a matrix for drug encapsulation and regenerative medicines or for encapsulating living cells [2, 3, 18–20]. These supramolecular nanostructures have been obtained as a result of supramolecular polymerizations (assembly) of small molecules and the formation of networks of the supramolecular nanostructures that entrap water molecules inside the networks and produce a hydrogel. Chemical reactions offer potential opportunities to introduce unique stimuli responsive functions

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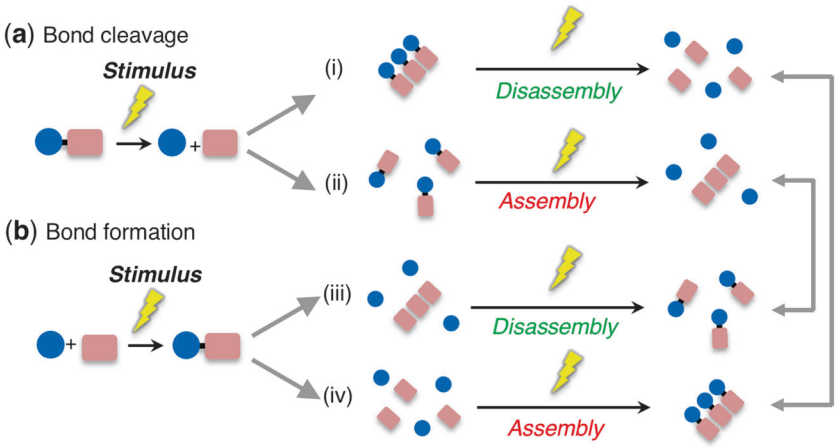

Fig. 1 Schematic showing four combination patterns of typical chemical reaction-guided supramolecular systems [**a** (i): *bond cleavage-induced assembly*, (ii): *bond cleavage-induced disassembly*, **b** (iii): *bond formation-induced assembly*, and (iv): *bond formation-induced disassembly*]. The blue-colored circles and pale orange-colored rectangles represent chemically reactive molecules and biomolecules (or bio-related molecules), respectively

into a supramolecular hydrogel system that would otherwise have been unachievable [2, 18–20]. Here, chemical reaction-guided supramolecular hydrogel systems are classified into four combination patterns based on their reaction mechanisms (*bond cleavage* or *bond formation*) and equilibrium directions of assembly (*assembly* or *disassembly*) [(i): *bond cleavage-induced assembly* (this system is categorized as class III (precursor modification followed by assembly) in the literature [21]), (ii): *bond cleavage-induced disassembly*, (iii): *bond formation-induced assembly*, and (iv): *bond formation disassembly*], as depicted in Fig. 1. Patterns "(i) and (iv)" and "(ii) and (iii)" are paired during reversible chemical reactions.

Chemical stimuli-responsive gel degradation systems (disassembly of supramolecular nanostructures responsible for hydrogel formation)

For the *bond cleavage-induced disassembly* mechanism, an example of pattern (i), we introduced chemical reaction-induced cleavable groups into potentially self-assembling biomolecules. Two distinct chemical reactive groups ("oxidation sensitive" *p*-borono-phenylmethoxycarbonyl (**BPmoc**) and "reduction sensitive" *p*-nitro-phenylmethoxycarbonyl (**NPmoc**)) were introduced into peptide molecules, as shown in Fig. 2a [8]. Research related to peptide-based supramolecular hydrogels has considerably increased due to their facile synthetic accessibility, commercial availability, and potential biocompatibility [3, 22–29].

Phenylalanine dipeptides bearing **BPmoc** and **NPmoc** groups (hereafter referred to as **BPmoc-FF** (oxidation responsive) and **NPmoc-FF** (reduction responsive), F: phenylalanine) show hydrogelation ability, i.e., a self-assembling propensity to form supramolecular nanofiber networks under aqueous conditions [8]. Transmission electron microscopy (TEM) and atomic force microscopy (AFM) observations revealed the presence of entangled nanofiber networks. The self-assembled structure of **BPmoc-FF** in a crystal state was unambiguously elucidated to be a parallel β-sheet structure via an X-ray analysis of a single-crystal (Fig. 2b), which was consistent with the results from circular dichroism (CD) and infrared (IR) spectroscopy analyses of the hydrogel state. The self-assembled structures in crystal (solid) states, which are mostly thermodynamically stable states, are generally different from the structures in the hydrogel states that are usually kinetically trapped (thermodynamically metastable) states. Nevertheless, the **BPmoc** group could play an important role in the construction of the supramolecular nanofiber networks predominantly by participating in hydrogen bond interactions and van der Waals interactions, based on the structural characterization. Indeed, the removal of the **BPmoc** group from **BPmoc-FF**, which proceeded even in its self-assembled state (i.e., in the hydrogel state), is triggered by the oxidant hydrogen peroxide (H_2O_2), eventually causing the H_2O_2-responsive degradation of the supramolecular hydrogel, as shown in Fig. 2c. According to an HPLC analysis, the gel-to-sol transition (or gel degradation) occurred when the **BPmoc-FF** concentration was less than the *critical gelation concentration* (*CGC*, defined as the lowest concentration of the (hydro)gelator that leads to (hydro)gel formation).

The abovementioned H_2O_2-responsive gel degradation of **BPmoc-FF** was successfully converted to other chemical stimuli-responsive functions via enzymatic reactions. For instance, glucose oxidase (GOx) catalyzes the selective oxidation of D-glucose, which simultaneously produces H_2O_2. Therefore, the encapsulation of GOx into the H_2O_2-responsive **BPmoc-FF** gel facilitated the construction of a glucose-responsive supramolecular hydrogel in a predictable and rational manner. As shown in our previous study, **BPmoc-FFF** (Fig. 2a, tripeptide instead of dipeptide) displays an excellent hydrogel formation ability (low *CGC* even at neutral pH) compared with **BPmoc-FF** [9]. Therefore, the **BPmoc-FFF** gel enabled the encapsulation of various enzymes while retaining their activities. A variety of biomolecules, including choline, uric acid, and sarcosine,

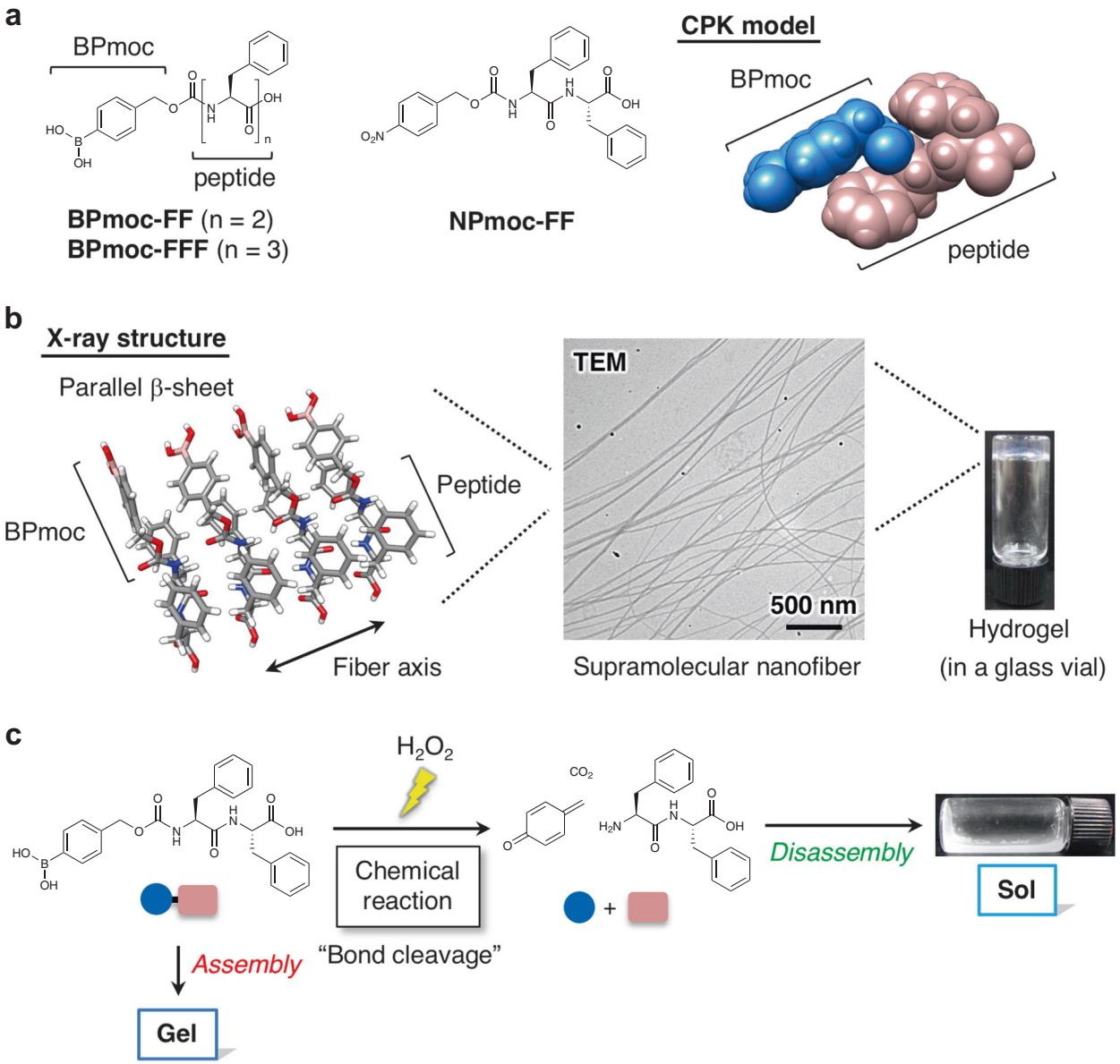

Fig. 2 **a** Chemical structures of **BPmoc-FF**, **BPmoc-FFF** and **NPmoc-FF**, and a space-filling model of **BPmoc-FF**. **b** X-ray structure of **BPmoc-FF** displaying a self-assembled (parallel β-sheet) structure that would likely be the core (cross β-spine) of supramolecular fibers observed under the microscope. A typical photograph and TEM image of a **BPmoc-FF** gel is shown. **c** Schematic showing bond cleavage chemical reaction-induced disassembly (pattern (i), Fig. 1) of **BPmoc-FF** to afford a H_2O_2-responsive supramolecular hydrogel

induce gel degradation upon the encapsulation of the corresponding oxidases (Fig. 3a). Other enzymes have been encapsulated with the oxidases to further expand the number of chemical stimuli that induce gel degradation. For example, acetylcholine esterase (AChE), a hydrolase, has been encapsulated with choline oxidase (COx), extending the H_2O_2-responsiveness of the **BP-FFF** gel to acetylcholine-responsiveness since acetylcholine is hydrolyzed to choline by AChE and choline subsequently induces the production of H_2O_2 by COx (Fig. 3b).

Similarly, **NPmoc-FF** gave rise to a reduction-responsive supramolecular hydrogel, where the removal of the **NPmoc** group in response to either chemical reduction (typically sodium dithionite ($Na_2S_2O_4$)) or enzymatic reduction (nitroreductase (NTR) coupled with nicotinamide adenine dinucleotide (NADH)) is essential for gel degradation. The H_2O_2-responsive **BPmoc-FFF** gel was then mixed with this reduction-responsive **NPmoc-FF** gel, along with two enzymes (GOx and NTR). Excitingly, this supramolecular hydrogel–enzyme hybrid material displayed Boolean AND logic-gated responses toward two biomolecules, namely, glucose and NADH. Gel degradation was not observed by the addition of either one of the two biomolecules. On the other hand, the addition of both glucose and

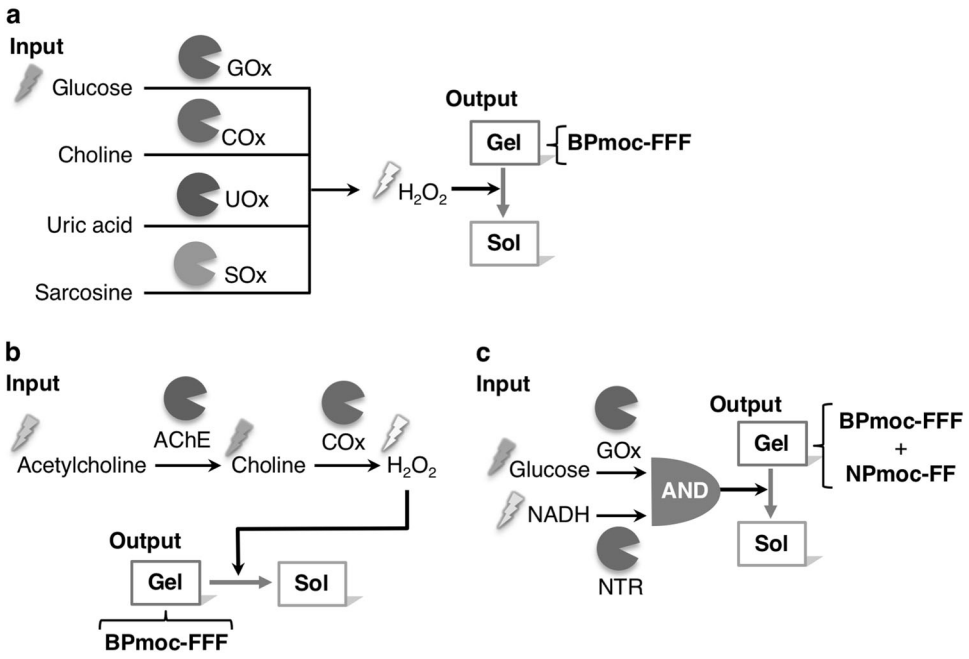

Fig. 3 **a** Expansion of the variety of chemical stimuli used to induce gel degradation (gel-to-sol transition, disassembly). GOx: glucose oxidase, COx: choline oxidase, UOx: urate oxidase, SOx: sarcosine oxidase. **b** Serial coupling of two enzymatic reactions (AChE and COx) to extend the chemical stimuli responsiveness from H_2O_2 to acetylcholine. **c** Scheme showing Boolean AND logic-gated responses of a supramolecular hydrogel composed of **NPmoc-FF** and **BPmoc-FFF** toward glucose and NADH as inputs

NADH was necessary to induce gel degradation. This finding was ascribed to the induction of the two chemical reactions (the removal of **BPmoc** and **NPmoc**) upon the addition of glucose and NADH to cause the disassembly of **BPmoc-FFF** and **NPmoc-FF** that is necessary to induce complete the degradation of the hybrid material (Fig. 3c).

These supramolecular hydrogel–enzyme hybrid materials highlight the potential of chemical reaction-guided stimuli-responsive supramolecular systems. Rational expansion of a variety of chemical stimuli to modulate the self-assembly and installation of logic-gated stimuli-responsive functions may have potential (bio)applications, such as intelligent drug releasing reservoirs. Furthermore, in principle, the stimuli are amplified using positive feedback loops [30]. Negative feedback loops and combinations may also enable researchers to construct further sophisticated stimuli-responsive supramolecular (hydrogel) systems. Moreover, photo-responsive supramolecular hydrogels, including less explored two-photon excitation sensitive hydrogels, have been constructed by introducing photo-caged groups into the N-terminus of dipeptides using a similar molecular design approach [8, 11].

Heat stimuli-responsive gel formation systems based on a retro-Diels-Alder reaction (growth of supramolecular nanostructures responsible for hydrogel formation)

For *bond cleavage-induced assembly*, an example of pattern (ii), we used a thermally reversible, retro-Diels-Alder (rDA) reaction. As shown in Fig. 4, an unprecedented class of heat-set supramolecular hydrogels has been designed, in which the rDA reaction is coupled with the growth of supramolecular nanostructures by detaching a hydrophilic group from precursor molecules [12, 13]. For instance, we developed a glycolipid **1** (Fig. 4a) bearing a maleimide group (dienophile) that self-assembles to form supramolecular hydrogel ($CGC = 0.2$ wt% (3.6 mM), T_{gel} (gel-to-sol transition temperature, above which the gel state turns into sol state) = 80 °C at 0.2 wt%, in 200 mM HEPES (pH 7.2)) [12]. The glycolipid **1** then reacted with a hydrophilic furane **2** (diene) to afford a water-dispersible precursor **3** (cycloadduct) via the DA reaction. The precursor **3** self-assembled to form nanosheets but did no exhibit a hydrogel formation ability. When the dispersed aqueous solution of compound **3** was heated at 60 °C (below T_{gel}), a hydrogel was formed after 5 h (without cooling). HPLC analyses revealed that precursor **3** was converted to the original glycolipid **1** (hydrogelator), and the amount exceeded the approximate *CGC* value when the hydrogel formed. Microscopic observations using TEM and AFM revealed a morphological transformation from nanosheets of ~100–200 nm to a network of nanofibers with a diameter of ca. 35 nm after heating, which is likely responsible for hydrogel formation. Overall, a heat-induced rDA (bond-cleavage) reaction triggered the growth of supramolecular nanostructures, which allowed the molecular design to develop the heat-set supramolecular hydrogel systems in a semi-rational manner (Fig. 4b). As shown in our subsequent study, a peptide-based self-assembling molecule **4** (Fig. 4c) bearing a furan (dienophile) group (instead of maleimide (diene)) was capable of forming thermally stable supramolecular hydrogels at a notably lower concentration (CGC = 0.06 wt% (1.0 mM), $T_{gel} = 92$ °C at 0.06 wt%) [13]. A

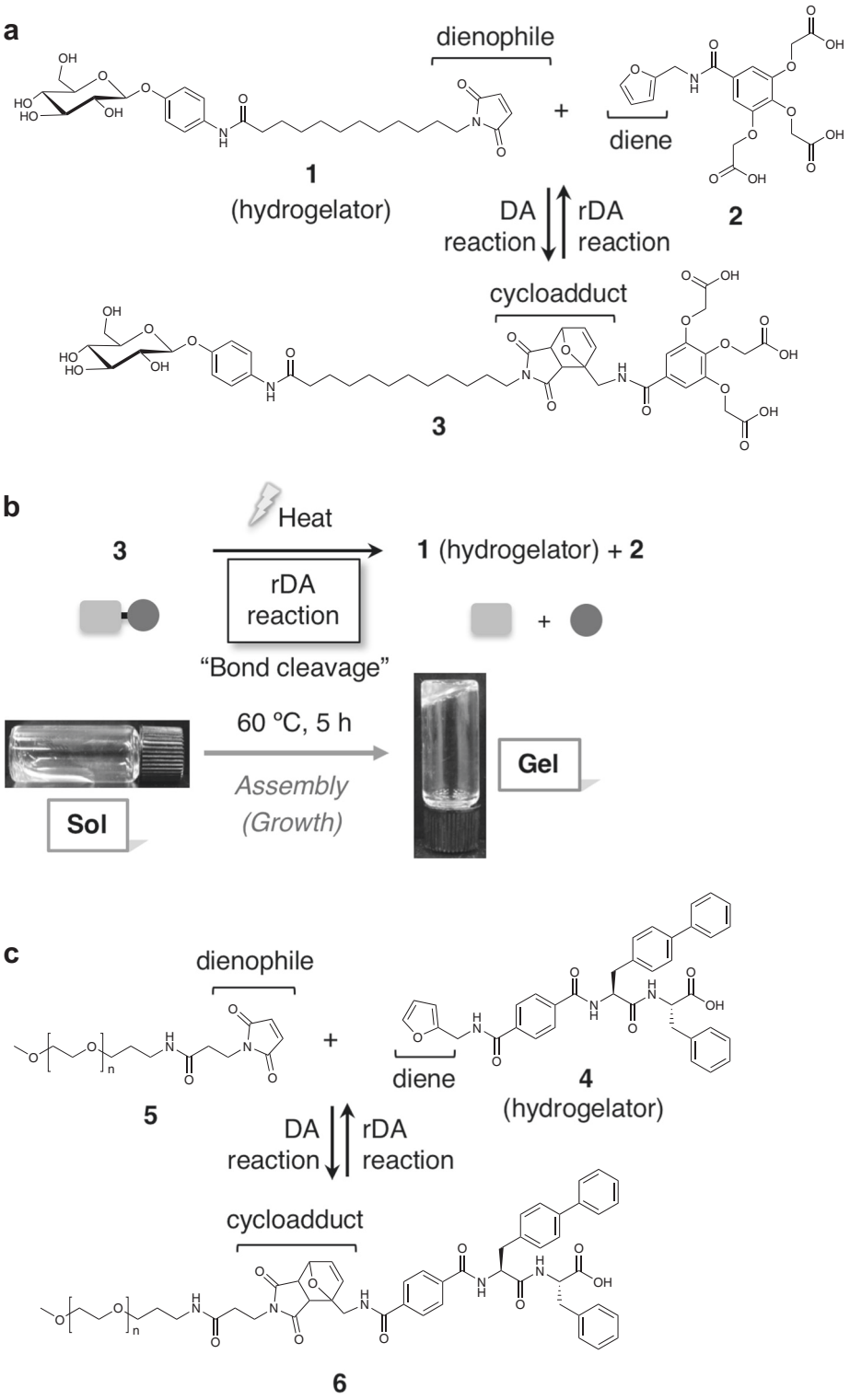

Fig. 4 **a** Chemical structures of glycolipid **1** bearing a maleimide (dienophile) and compounds **2** and **3**. **b** Schematic showing the bond cleavage chemical reaction-induced assembly (pattern (ii), Fig. 1) of precursor **3** into a heat-set supramolecular hydrogel. **c** Chemical structures of peptide **4** bearing a furan (diene) and compounds **5** and **6**

precursor **6** was then constructed via the DA reaction, with a hydrophilic maleimide **5** bearing a PEG chain. In this case, hydrogel formation was accelerated due to the superior gelation ability of the peptide derivative **4**. Here, self-assembling molecules (hydrogelators) can be modified with either diene (maleimide) or dienophile (furan), underscoring the flexibility of this simple, modular, and hybrid molecular design to construct heat-set, self-assembling systems guided by a chemical reaction.

Several examples of chemical reaction-guided supramolecular hydrogel systems developed in previous studies are briefly described below. As the seminal example of a paired

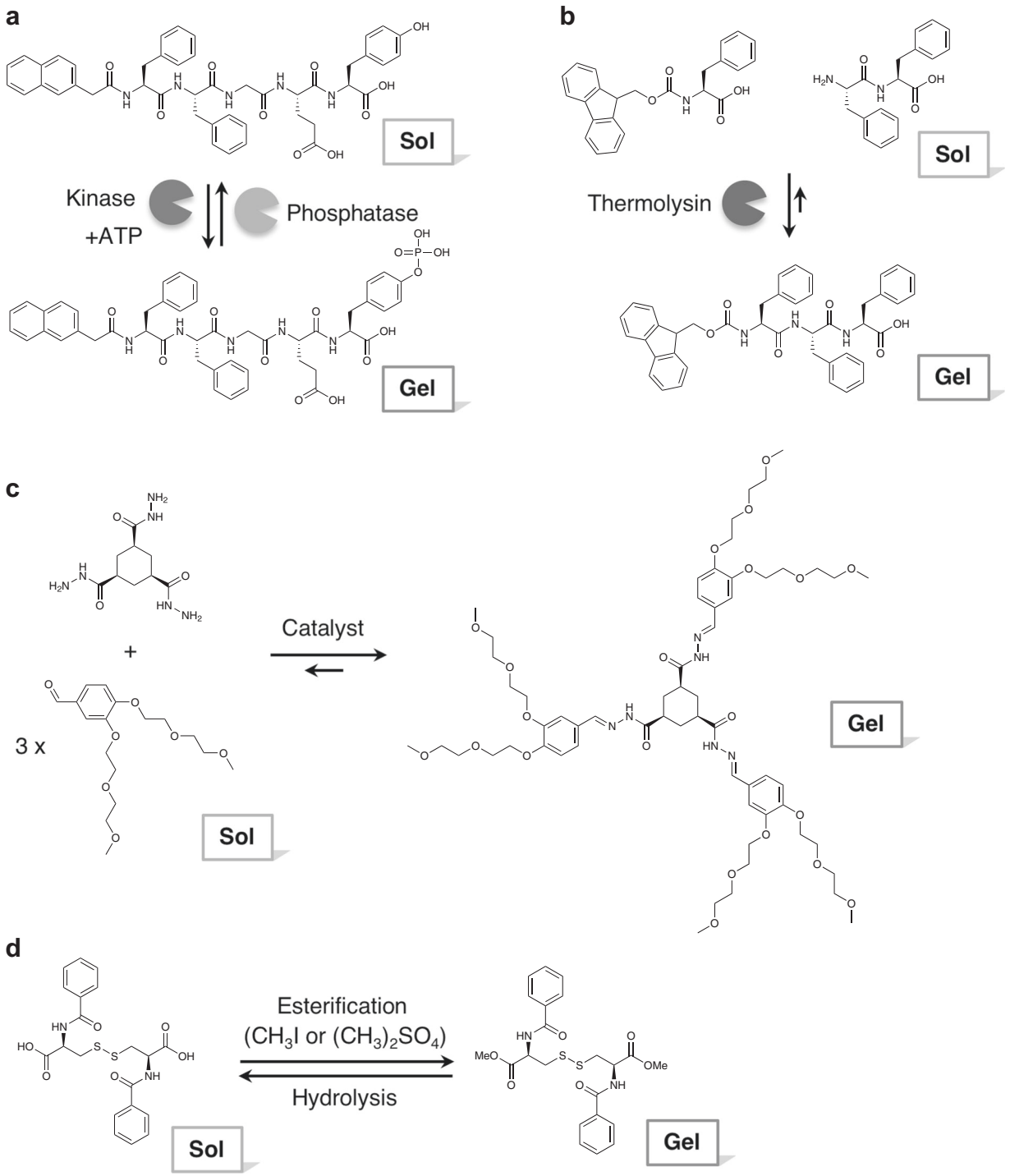

Fig. 5 **a** Enzyme-instructed self-assembly (EISA) of peptide derivatives bearing a tyrosine residue (patterns (ii) and (iii), Fig. 1). **b** Construction of self-assembling peptides (supramolecular hydrogelators) from principally non-self-assembling amino acids and/or peptides (pattern (iv), Fig. 1). **c** Hydrazone bond formation reaction among non-self-assembling molecules to construct a supramolecular hydrogelator and the corresponding hydrogel state (pattern (iv), Fig. 1). **d** Conditional and reversible ester bond formation to modulate self-assembly, giving rise to the transient formation of a supramolecular hydrogel (patterns (i) and (iv), Fig. 1)

pattern (ii) *bond cleavage-induced assembly* and (iii) *bond formation-induced disassembly*, Xu et al. elegantly reported the enzyme-instructed self-assembly (EISA) of peptide derivatives bearing a tyrosine residue, which is phosphorylated (bond-formation) or dephosphorylated (bond-cleavage) using the corresponding enzymes (kinase or phosphatase), as shown in Fig. 5a [31]. This class of EISA systems based on the activities of kinases/

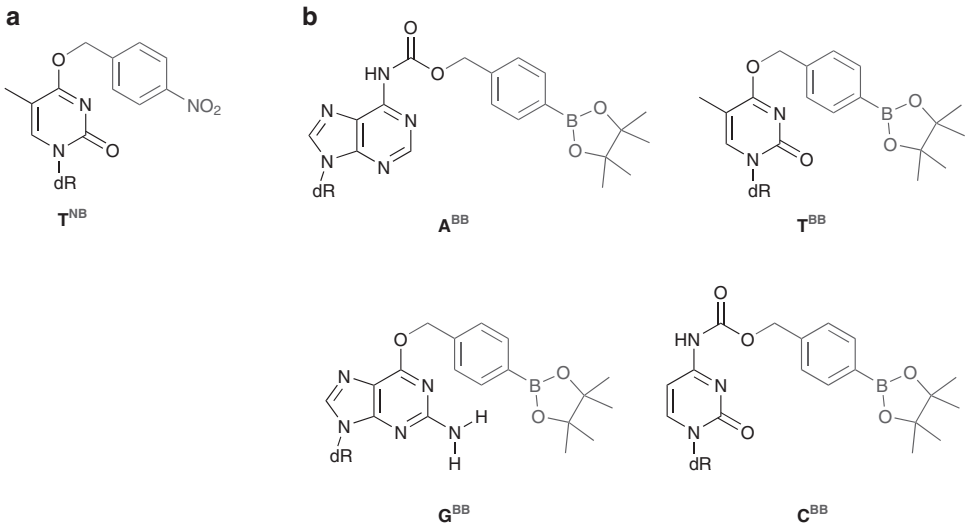

Fig. 6 Chemical structures of (**a**) $\mathbf{T}^{\mathbf{NB}}$ [39] and (**b**) $\mathbf{N}^{\mathbf{BB}}$ (N = A, T, G, C) [40]

phosphatases and other enzymes has been extensively studied. Furthermore, several of these supramolecular systems have been applied as unique tools capable of executing biological functions (e.g., inhibition of cell growth), even *in vivo* [32]. In a study describing a series of examples of pattern (iv) *bond-formation-induced assembly* by Ulijn et al., peptide synthesis catalyzed by proteases (reversed hydrolysis) constructed self-assembling peptides (supramolecular hydrogelators) from principally non-self-assembling amino acids and/or peptides, as shown in Fig. 5b [33]. As another example of pattern (iv), van Esch and Eelkema et al. reported hydrazone bond-formation reactions among non-self-assembling molecules (i.e., one molecule bearing three hydrazide groups and three molecules bearing one aldehyde group) to construct a supramolecular hydrogelator and the corresponding hydrogel state, as shown in Fig. 5c [34]. The mechanical properties of the resulting supramolecular hydrogels (network structure and bundling tendency of supramolecular nanofibers) were controlled by the reaction rate of the hydrazone bond formations catalyzed upon the addition of aniline under acidic pH. In the context of transient self-assembly [35, 36] and paired pattern (i) and (iv), the same group published intriguing findings and utilized reversible ester bond-formation reactions to modulate molecular self-assembly, as shown in Fig. 5d [37, 38].

Chemical stimuli-responsive folding of nucleic acids controlled by chemical reactions

Chemical reactions enable stimuli-responsive control not only over molecular self-assembly but also over other molecular behaviors, such as the folding of biomolecules (i.e., controlled conformational changes), which are indispensable for their functions (e.g., enzymatic activity, information storage, and energy conversion). We have recently been interested in controlling the three-dimensional folding of nucleic acid structures in response to desired chemical stimuli. Seminal studies on photo-caged nucleic acids, which enable photo-control over a variety of biological events [6], have been published. However, limited examples showing chemical stimuli responsiveness have been discussed [16, 39, 40]. For instance, Saneyoshi and Ono et al. developed a O^4-(4-nitrobenzyl)-modified thymine ($\mathbf{T}^{\mathbf{NB}}$, Fig. 6a) capable of showing bio-reduction-responsive transformation into thymine even when it was introduced into oligodeoxynucleotides (ODNs) [39]. Morihiro and Obika et al. also developed four nucleobases bearing a 4-boronobenzyl group ($\mathbf{N}^{\mathbf{BB}}$, Fig. 6b) and introduced $\mathbf{T}^{\mathbf{BB}}$ into antisense ODNs for hydrogen peroxide-triggered gene silencing [40]. We focused on guanine because the guanine ring is involved in unique three-dimensional folded structures, such as G-quadruplexes, which comprise stacked structures of G-tetrads in which four guanines self-assemble via the formation of eight Hoogsteen hydrogen bonds and whose central cavity is occupied by cations (typically, potassium ions) [41].

We designed and synthesized a guanine derivative ($\mathbf{G}^{\mathbf{NB}}$) bearing a reduction-responsive unit at the O^6 position, as shown in Fig. 7 [15]. Chemical and enzymatic reductions trigger the removal of the NB group from $\mathbf{G}^{\mathbf{NB}}$ to recover the original guanine at an approximately quantitative level (in a manner similar to **NPmoc-FF**). $\mathbf{G}^{\mathbf{NB}}$ was primarily introduced at the 5′-end of a G-quadruplex-forming, thrombin-binding DNA aptamer (**TBA**: 5′-GGTTGGTGTGGTTGG-3′, 15-mer) to construct $\mathbf{TBA}^{\mathbf{NB}}$ (5′-$\mathbf{G}^{\mathbf{NB}}$GTTGGTGTGGTTGG-3′). Because only a two-layer G-tetrad is required for **TBA** to fold into its functional structure (chair-type, anti-parallel G-quadruplex), the replacement of one of the eight guanines that participate in G-quartet formation is sufficient to destabilize the overall G-quadruplex structure. Indeed, Mayer and Heckel et al.

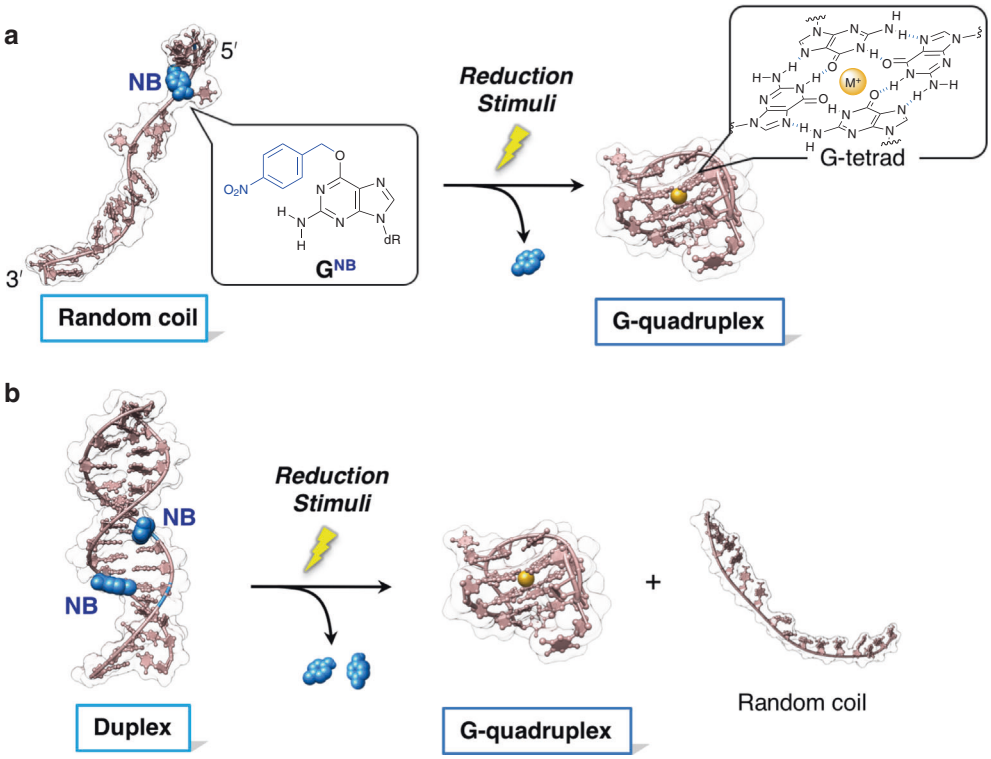

Fig. 7 Schematics depicting reduction-responsive tertiary structural transitions of nucleic acids from (**a**) a random coil (**TBA**$^{\mathbf{NB}}$) to a G-quadruplex (**TBA**) and (**b**) a duplex (**TBA**$^{\mathbf{(5,8)NB2}}$•**CS**$^{\mathbf{5G}}$) to a G-quadruplex (**TBA**). The chemical structure of **G**$^{\mathbf{NB}}$ is shown in the inset. The molecular models were visualized with the UCSF Chimera package (Chimera is developed by the Resource for Biocomputing, Visualization, and Informatics at the University of California, San Francisco (supported by NIGMS P41-GM103311)) [49] (the NB group of **G**$^{\mathbf{NB}}$ is shown in the space-filling model (blue color))

successfully constructed a pioneering photo-caged **TBA** capable of inducing photo-responsive G-quadruplex formation by replacing one guanine of **TBA** with a photo-caged guanine [42]. As expected and depicted in Fig. 7a, the reduction-responsive structural transition of **TBA**$^{\mathbf{NB}}$ from random coil to G-quadruplex was clearly manifested using CD spectroscopy [15]. In addition, the two other **TBA**s bearing one **G**$^{\mathbf{NB}}$ (**TBA**$^{\mathbf{5NB}}$: 5′-GGTT**G**$^{\mathbf{NB}}$GTGTGGTTGG-3′) and two **G**$^{\mathbf{NB}}$ bases (**TBA**$^{\mathbf{(5,8)NB2}}$: 5′-GGTT**G**$^{\mathbf{NB}}$GT**G**$^{\mathbf{NB}}$TGGTTGG-3′) were capable of showing reduction-responsive folding into G-quadruplex [17]. According to recent studies, DNA G-quadruplex structures potentially function in cellular processes, such as transcription [43], and they have also been frequently found in DNA aptamers [44]. Therefore, chemically caged nucleic acids bearing **G**$^{\mathbf{NB}}$ might offer a potential opportunity to install OFF-ON switching functions into several inherent and/or artificially engineered biological functions.

We unintentionally found that a duplex containing a G:G mismatch pair is unstable compared with a G$^{\mathrm{NB}}$:G mismatch [17], suggesting that a duplex structure might be destabilized after the removal of NB from the duplex in response to reduction stimulus. When a complementary sequence (**CS**$^{\mathbf{5G}}$: 3′-CCAA**G**CACACCAACC-5′) was hybridized with **TBA**$^{\mathbf{5NB}}$, a stable duplex **TBA**$^{\mathbf{5NB}}$•**CS**$^{\mathbf{5G}}$ ($T_{\mathrm{m}} = 59$ °C) was obtained. Nevertheless, the duplex was not dissociated even after the almost complete removal of NB from **TBA**$^{\mathbf{5NB}}$ upon the addition of $Na_2S_2O_4$ as a chemical reductant. This observation was attributed to the decreased stability of the G-quadruplex structure of **TBA** ($T_{\mathrm{m}} = 51$ °C) compared to the duplex **TBA**•**CS**$^{\mathbf{5G}}$ ($T_{\mathrm{m}} = 55$ °C) bearing the single G:G mismatch pair, even in the middle of the sequence. We then redesigned **TBA**$^{\mathbf{(5,8)NB2}}$ and hybridized it with an oligonucleotide (**CS**$^{\mathbf{(5,8)G7T}}$: 3′-CCAA**G**C**T**GACCAACC-5′) such that the duplex **TBA**$^{\mathbf{(5,8)NB2}}$•**CS**$^{\mathbf{5G}}$ ($T_{\mathrm{m}} = 41$ °C) contains two G:G mismatch pairs and single T:T mismatch pair. The duplex **TBA**•**CS**$^{\mathbf{5G}}$ ($T_{\mathrm{m}} = 28$ °C) obtained after the removal of two NBs would become less stable than the G-quadruplex. In fact, a clear CD spectral change was observed upon the addition of $Na_2S_2O_4$, indicating a reduction-responsive structural transition from a duplex to a G-quadruplex (Fig. 7b). As the initial conformational state, duplex structures might hold several advantages over the random coil single-stranded structure. The duplex may be multifunctional. For example, the complementary strand could be used as a carrier by designing its sequence (e.g., by attaching an aptamer sequence for desired targets in addition to the complementary sequence for duplex formation) to deliver potentially therapeutic guanine-rich oligonucleotides to a

desired location. Therefore, these findings may be useful tools to assist in the development of DNA nanoarchitectures capable of reduction-responsive functions. Since nucleic acids, particularly RNA, generally show more limited chemical stability than other biomolecules, such as peptides and proteins, the scope and limitation of chemical stimuli responsiveness to construct chemically caged nucleic acids would be important to evaluate using in vitro and in vivo experiments in the future.

Summary and outlook

In summary, modular and hybrid molecular designs used to introduce artificial chemical reactive groups into biomolecules or bio-related molecules will enable the semi-rational construction of stimuli-responsive supramolecular systems. An expansion of the modality (i.e., enrichment of applicable chemical stimuli) and diversification of the behavior of the system (e.g., supramolecular structural transformation at various hierarchies and scales) with controlled dynamics (i.e., fast or slow rate or targeting to selected pathways) would be important to construct more complex and sophisticated supramolecular systems that might exhibit valuable adaptive functions as the output in response to chemical signals and/or changes in the surrounding environments as input signals. Therefore, the coexistence of multimolecular/supramolecular systems in a confined space [2] and organic-inorganic composites [45, 46], such as biological systems, would be of great importance.

From the perspective of molecular design, the modular molecular design would be the simplest yet rational method to introduce new functions originating from the introduced functional small module (e.g., chemically reactive small molecules as the modules to attain the chemical stimuli-responsive functions discussed here). Nevertheless, this type of modular molecular design approach for introducing multiple functions would become complicated and require tedious synthetic procedures, in which the compatibility in terms of stimuli responsiveness and synthetic routes should be carefully considered and examined, and inevitably yield a larger molecular size. In natural systems, more than half of proteins do not exhibit single functions but contribute to multiple biochemical functions [47]. In addition, these natural multifunctional proteins take advantage of fused molecular designs (i.e., a single domain for multiple functions) [48]. Thus, sophisticated molecular design approaches are potentially the next target for chemists to construct complex, semi-artificial supramolecular systems inspired by biological systems created through molecular evolution.

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Compliance with ethical standards

Conflict of interest The author has no conflicts of interest to declare.

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