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Review

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# Cyclodextrin Molecules, Polymers and Nanomaterials

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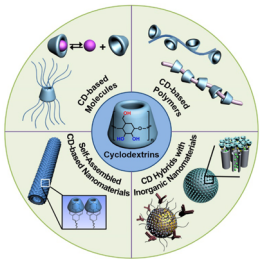
**Abstract:** Cyclodextrins (CDs) have been widely utilized for myriad applications due to their unique structural features such as hydrophobic cavity for inclusion complex formation and numerous hydroxyl groups for functionalization. In this review, we discuss various applications of CD-based molecules, polymers and nanomaterials. CD-based molecules have been developed for pharmaceutic applications and drug/gene carriers. CD-based polymers have been adopted for drug delivery, stimuli-responsive hydrogels and self-healing materials. CD hybrids with inorganic nanoparticles have been employed for stimuli-responsive carriers.




**Keywords:** cyclodextrins, inclusion complex, self-assembled structure, stimuli-responsiveness, therapeutic materials.

## 1. Introduction

Cyclodextrins (CDs) are macrocyclic oligosaccharides consisted of 6-8 $\alpha$-D-glucopyranoside unit linked by $\alpha$-(1,4) bonds, which are named $\alpha$-, $\beta$- and $\gamma$-CDs, respectively. Villiers *et al.* first discovered the CDs in 1891,[1,2] and Schardinger named these molecules as $\alpha$- and $\beta$-CDs after several decades.[3] In 1942, the structures of $\alpha$- and $\beta$-CDs were discovered using x-ray diffraction analysis.[4] In 1948, the $\gamma$-CD structure was also discovered and the inclusion complex formation of CDs originated from the hollow nature of CDs was recognized.[4] CDs have been utilized in myriad areas such as food, chemical industry, pharmaceutics, agriculture, deodorizing agent, environmental engineering.[4] In particular, pharmaceutical uses of CDs have attracted great interest because of their unique characteristics such as drug encapsulation, solubilization, controlled release of guest molecules, stabilization, penetration of body tissues.[5,6] Furthermore, CDs could be utilized as building blocks of self-assembled nanostructures for drug or gene delivery applications.[7-11] The hydrophobic cavity could be used for encapsulation of a variety of guest molecules. Amphiphilic CDs with cationic nature could be utilized for gene carriers by electrostatic complexation with negatively charged DNAs or RNAs. Furthermore, the capability of host-guest complex formation of CDs by molecular recognition could provide a unique way to form or to transform self-assembled nanostructures with diverse functions. The numerous hydroxyl groups of CD molecules could be utilized for formation of myriad polymers with diverse chemical structures such as linear CD-polymers, CD-terminated polymers, CD-centered star polymers and polymers with pendant CDs.[12,13] The unique chemical structure of CD-containing polymers provides diverse opportunities in numerous applications.[14,15] In particular, the CD-containing polymers could be adopted to produce stimuli-responsive and self-healable hydrogel by specific host-guest complexation. The CD building blocks could be employed to construct multifunctional self-assembled nanotubes as well as organic-inorganic hybrid nanoparticles.[16-25] In this review, we describe numerous applications of unique CD-containing molecules, polymers and nanomaterials.

## 2. Chemical and physical properties of CD molecules

CDs have donut-like molecular structure with hydrophilic larger and smaller rims exposed to the solvent due to their macrocyclic nature. The molecular structures and physical properties of $\alpha$-, $\beta$-, and $\gamma$-CDs are summarized in Figure 2.[26,27] Because the wide rim and narrow rim of $\alpha$-, $\beta$-, and $\gamma$-CDs have 12-16 secondary hydroxyl groups and 6-8 primary hydroxyl groups as

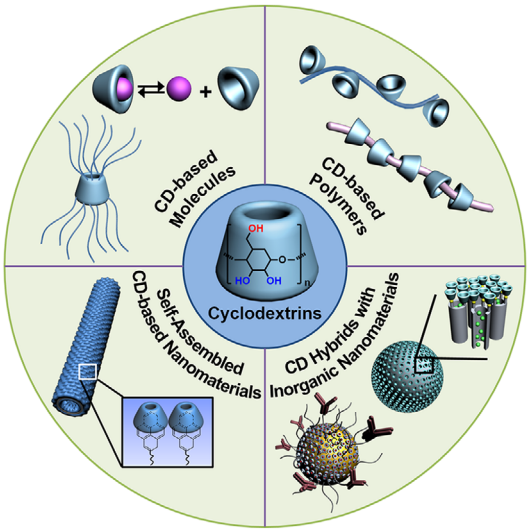



**Figure 1.** Schematic representation of CD-based molecules, polymers and nanomaterials.

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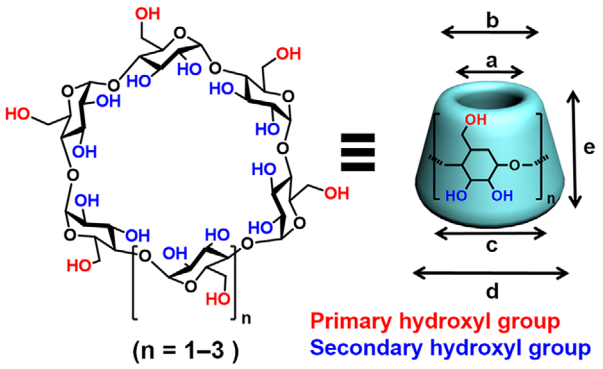


| Property | α-CD | β-CD | γ-CD |
|---|---|---|---|
| Number of Glucose Units | 6 | 7 | 8 |
| a (Å) | 4.5 | 6.1 | 7.7 |
| b (Å) | 13.2 | 14.9 | 16.1 |
| c (Å) | 5.7 | 7.8 | 9.5 |
| d (Å) | 13.7 | 15.3 | 16.9 |
| e (Å) | 7.8 | 7.8 | 7.8 |
| Solubility (g/100 mL) | 14.5 | 1.85 | 23.2 |
| Molecular Weight (g/mol) | 972 | 1135 | 1297 |
| Cavity Volume (Å$^3$) | 174 | 262 | 427 |

**Figure 2.** Molecular structures and physical properties of $\alpha$-, $\beta$-, and $\gamma$-CDs.

shown in Figure 2, respectively, the CDs have hydrophilic outer surface while the inner cavity is relatively hydrophobic. The water solubilities of $\alpha$-, $\beta$-, and $\gamma$-CDs are 15, 2 and 23 wt%, respectively.[26] The low solubility of $\beta$-CD is originated from the formation of intramolecular hydrogen bonding between the secondary hydroxyl groups (neighboring C2-OH and C3-OH).[26,28] The hydrogen bonding yields the complete secondary belt with rigid structure.[29] Around the dissolved $\beta$-CD molecules, high water density with strong ordered structure has been found by molecular dynamic simulations, which indicates that the water molecules around the dissolved $\beta$-CD have low entropy and unfavorable enthalpy to hinder spontaneous solubilization.[28] Therefore, via modification of secondary hydroxyl group, the water solubility of $\beta$-CD could be improved.[30] In contrast, $\alpha$-CD with incomplete belt of hydrogen bond and $\gamma$-CD with non-coplanar structure exhibit higher solubility in water.[29]

The most characteristic aspect of CDs is their capability to construct inclusion complex with myriad guest molecules owing to the hydrophobic nature of the CD cavity providing a microenvironment where the hydrophobic guests with appropriate size could be included to produce host-guest complex. The release of water molecules from the hydrophobic cavity is the major driving force to form host-guest inclusion complex.[29] Due to their different cavity size, $\alpha$-, $\beta$-, and $\gamma$-CDs have different ranges of guest molecules entrapped into their cavity. Moreover, as shown in Figure 3, various stoichiometry between host and guest molecules could be formed such as 1:1, 1:2, 2:1, 2:2 or even more multiple host-guest complex such as polyrotaxanes and polypseudorotaxanes with necklace-like structure.

The inclusion complex formation in the cavity of CDs greatly alters the physical and chemical characteristics of the guest molecules, especially in water solubility, which is an important

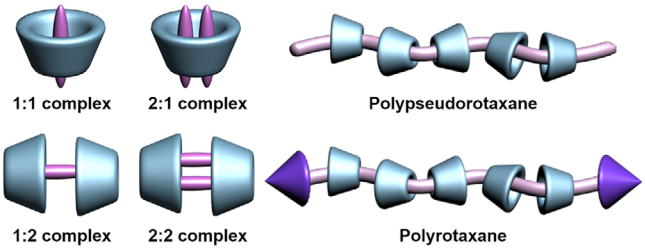


**Figure 3.** Schematic representation of various CD inclusion complexes with guest molecules.

feature that the CDs have attracted much interest in numerous applications. The encapsulated guest molecules could be released by heating, pH change, or enzymes.[31]

The chemical structure of CDs could be modified to alter their binding property, solubility and even toxicity.[6,32] Most modifications of CDs utilize the primary (C-6 position) and secondary hydroxyl groups (C-2 and C-3 position) of the glucose repeating unit. Among many hydroxyl groups, the primary hydroxyl groups at C-6 position are the most nucleophilic, hence, the most reactive. The secondary hydroxyl groups at C-3 position are difficult to access and, hence, least reactive.[32] The controlled functionalization methods can be found in other reviews.[32]

The safety information of $\alpha$-, $\beta$-, and $\gamma$-CDs upon administration have been reported.[33,34] In general, the route of administration primarily determines the safety profiles of CDs. Practically, the oral administration of CDs shows no toxicity because the absorption of CDs in the gastrointestinal tract is low and CDs can be degraded by amylase in the digestive tract or by bacteria in colon and cecum.[4,35] Subcutaneous administration of $\beta$-CD in high doses in animals might cause nephrotoxicity, body weight decrease, and liver weight decrease.[36] Intravenous administration of $\beta$-CD also shows similar effects due to the complex formation with several blood components such as cholesterol, which induces destabilization of erythrocytic membrane by inclusion complex formation.[37] Some CD derivatives could be utilized as alternatives with lower toxicity to the parent CDs. For example, 2-hydroxypropyl-$\beta$-CD (2-HP-$\beta$-CD) is well tolerated in animals and humans.[38,39] Furthermore, sulfobutylether $\beta$-CD (SBE-$\beta$-CD) is well tolerated after intravenous or oral administration.[39]

## 3. CD molecules for pharmaceutical uses by inclusion complex formation

One of the most important characteristics of CDs in pharmaceutical application is enhancing apparent aqueous solubility of drugs with low solubility through inclusion complex formation. CDs can serve as dissolution enhancers in aqueous phase or hydrophilic carriers for drugs with low water solubility.[40] Poor biopharmaceutical properties such as solubility or permeability are huge hurdle in over 40% of drug candidates.[41] For these candidates, CDs can play an important role by enhancing the apparent aqueous solubility.[42-44]

Furthermore, inclusion complex formation with CDs can increase the bioavailability of hydrophobic drugs by enhancing not only solubility but also permeating amount of drugs. The inclusion complex formation of drugs with CDs enhances the

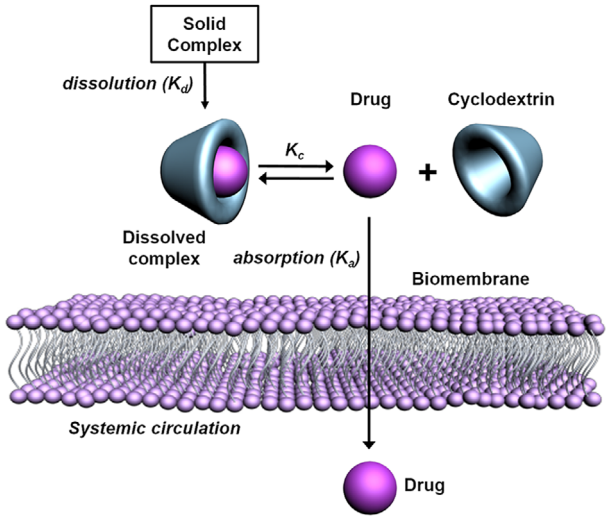


**Figure 4.** Schematic representation of the absorption of drugs from inclusion complex with CDs through biomembranes.

permeating amount of drugs with low solubility by making the drugs available on the surface of the biological barriers.[45] The drugs in the hydrophobic cavity of CDs would be partitioned into the biomembranes without disrupting the lipid bilayers of the membrane (Figure 4).[45] Only the free drugs in equilibrium with the inclusion complexes with CDs have capability of penetrating hydrophobic membranes because the CDs hardly penetrate the biomembranes due to their chemical structure with hydrophilic surface, molecular weight and very low octanol/water partition coefficient.[46] In this mechanism, excess CDs without forming complex may decrease the drug availability by increase of drugs partitioned into the cavity of CDs. Thereby, just enough stoichiometric CDs to solubilize the drug in water should be used to maximize the bioavailability of drugs.[47-49]

Due to the aforementioned powerful advantage of using CDs, numerous pharmaceutical drugs in an inclusion complex form with CDs have been developed and marketed. The initial marketed products with native CDs were developed with various prostaglandins.[50,51] In 1976, the first $\beta$-CD complex of prostaglandin E2 (PGE2) with potent oxytocin-like effects for the induction of labor in parturition was approved and marketed in Japan.[6,52,53] Although the PGE2 is highly unstable, its inclusion complex with $\beta$-CD exhibits significantly improved stability with high effectiveness. Piroxicam, a nonsteroidal anti-inflammatory drug, was also marketed using complexation with native CDs. While the piroxicam shows poor dissolution in aqueous phase due to its poor solubility and high crystallinity,[54-57] the included form of piroxicam in $\beta$-CD showed improved apparent solubility and safety without reduction in pharmacokinetic characteristics.[6]

Among the numerous commercially available CDs, randomly methylated CD with a low degree of substitution (RM-$\beta$-CD) shows the most powerful solubilizing effect.[45] Due to its good biocompatibility and complexing efficiency, RM-$\beta$-CD is used in composition of various marketed drugs including an eye drop of the antibiotic chloramphenicol in Portugal.[58] HP-$\beta$-CD with a low molar substitution exhibited the best complexing ability with low surface activities. Complexation of guests and HP-$\beta$-CD with a low degree of substitution is highly similar to parent $\beta$-CD.[45] The drugs included in HP-$\beta$-CD could be administrated in a wide range of route including rectal, oral, intravenous, buccal and ophthalmic ways.[59] Itraconazole is the most widely used drug complexed with HP-$\beta$-CD, which is administered by oral and intravenous route.[60] Upon complexation with HP-$\beta$-CD, the apparent aqueous solubility of itraconazole increased over five orders of magnitude.[61-63] SBE-$\beta$-CD is an excellent solubilizer for several drugs, of which effectiveness is higher than that of $\beta$-CD.[64] The complexation with SBE-$\beta$-CD greatly enhanced the stability of diverse chemically unstable drugs than other CDs.[64] SBE-$\beta$-CD was formulated with voriconazole, an antifungal agent.[6] The defect of voriconazole including poor water solubility and low stability in aqueous solutions due to its inactive enantiomer formation was overcome by complexation with SBE-$\beta$-CD.[65]

## 4. Self-assembled CD-based materials

### 4.1. Self-assembled amphiphilic CD nanoparticles

Amphiphilic CDs have been synthesized to overcome the limit of natural CDs in pharmaceutical applications, which includes (1) enhancement of interaction with biomembranes, (2) additional interaction of long alkyl chains at the CD surface for hydrophobic drugs, and (3) spontaneous nanoparticle formation through self-assembly of amphiphilic CDs.[8,10,66] Among them, the capability of self-assembly is the primary advantage of amphiphilic CDs. These unique properties can improve the capacity of drug encapsulation, interaction with cellular membranes, release profiles of entrapped drugs and cytotoxicity.[8,66]

Non-ionic amphiphilic CDs with self-assembly capability have been employed as delivery carriers of various drugs. Amphiphilic $\beta$-CDs functionalized with multivalent mannose units were developed for tumor-targeted drug carrier as shown in Figure 5.[11] The C-6 hydroxyl units of $\beta$-CD were modified by mannose units and the C-2 and C-3 position were modified by propionyl units to produce $C_3$-CD-Man7. The modified non-ionic amphiphilic CD was self-assembled in aqueous phase to form nanoparticles with about 112 nm of hydrodynamic diameter. The $C_3$-CD-Man7

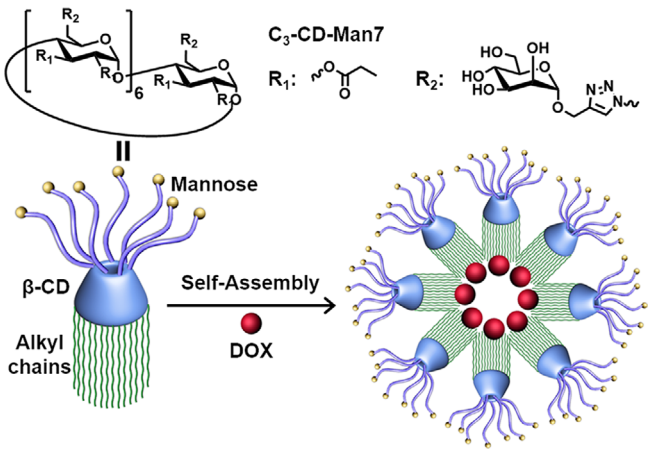


**Figure 5.** Chemical structure and self-assembled structure of $C_3$-CD-Man7 for targeted drug delivery.

 

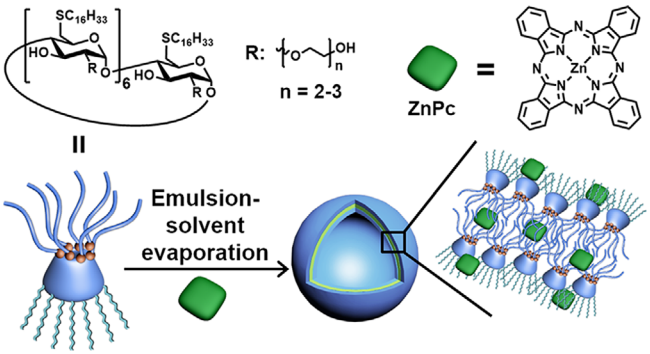


**Figure 6.** Non-ionic amphiphilic CDs encapsulating ZnPc photosensitizer for photodynamic therapy of cancer cells.

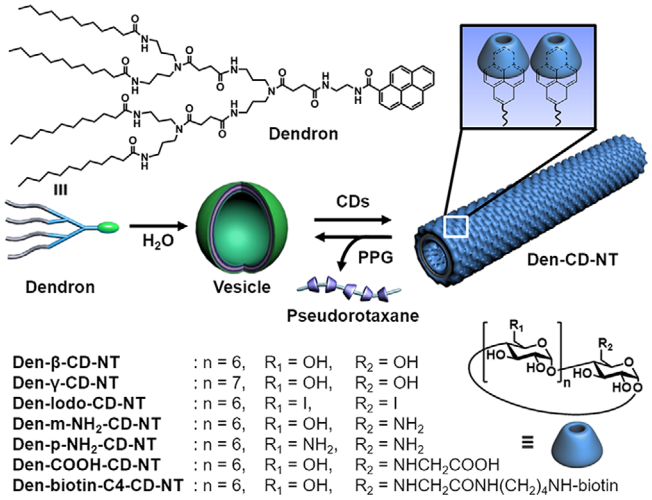


**Figure 7.** Formation of hierarchically self-assembled nanotubes by molecular recognition of CDs and amide dendrons.

nanoparticles could encapsulate doxorubicin (DOX, an anticancer drug), which showed slightly increased hydrodynamic diameter (199 nm). The DOX-loaded CD nanoparticles exhibited a pH-responsive drug-release property, which was originated from the protonation of the amino groups of DOX and resulting electrostatic repulsion inside the nanoparticles. Furthermore, these nanoparticles with mannose targeting units were efficiently internalized into MDA-MB-231 breast cancer cells with high expression of mannose receptor by receptor-mediated endocytosis. Consequently, the self-assembled CD nanoparticles encapsulating DOX could sufficiently inhibit the growth of MDA-MB-231 cells *in vitro* and *in vivo*.

Furthermore, the non-ionic amphiphilic CDs were adopted for the delivery of zinc(II)-phthalocyanine (ZnPc) as a photosensitizer in photodynamic therapy of cancer cells as shown in Figure 6.[67] Here, the C-2 and C-6 positions of $\beta$-CD were modified by hydrophilic oligo(ethylene glycol) and hydrophobic hexadecylthioether units, respectively. The resulting CDs could self-assemble into nanoparticles entrapping ZnPc. In the nanoparticles, the ZnPc molecules exhibited high tendency not to form aggregation but to remain active monomeric form, which endowed ZnPc with high propensity to produce singlet oxygen upon irradiation. The nanoparticles showed photodynamic therapeutic effect on HeLa cancer cells.

Cationic amphiphilic CDs have been widely employed as a gene delivery vector.[8-10] The positive charge of cationic amphiphilic CDs was adopted for electrostatic complexation with negatively charged DNAs or RNAs. Polycationic amphiphilic $\beta$-CDs (paCD) with cationic amino groups at C-6 and hydrophobic alkyl chains at C-2 and C-3 position were developed for specific delivery of DNAs.[68] Furthermore, the mannose moiety was introduced to paCD for specific targeting of macrophage (glyco(paCD)). By electrostatic interaction with DNAs, the glyco(paCD) formed a nanocomplex with multivalent surface mannose units which enhanced specific interaction with complementary lectin receptors on the surface of macrophage. Furthermore, the surface mannose units decreased the non-specific interaction with surface proteoglycans of cells by shielding the positive charge of the nanocomplex.

### 4.2. Hierarchically self-assembled CD-based materials

The hierarchical self-assembly of diverse CD building blocks provides a unique route to CD-based multifunctional materials.[16-19,69-73] In particular, the inclusion complex formation by specific molecular recognition of CDs could be utilized for tuning the balance between the hydrophilicity and the hydrophobicity of the self-assembling building block. Furthermore, the molecular recognition of CDs could be utilized for the non-covalent surface functionalization. As shown in Figure 7, amide dendrons with focal pyrene unit were reported as a guest for CDs to produce dendron-CD complexes which serve as a building block to construct dendron-CD nanomaterials.[16,17] The amide dendrons self-assembled into a vesicular structure with about 210-260 nm of diameter in aqueous phase. This vesicular solution of dendron exhibited a broad excimeric emission of pyrene units at 420-550 nm, which indicated that the focal pyrene units of the dendron were stacked by $\pi$-$\pi$ interaction to form excimers. Upon addition of $\beta$- or $\gamma$-CDs into the vesicular solution, the excimeric emission of the focal pyrene units were disappeared. This result indicated that the hydrophobic pyrene units were included into the cavity of CDs by specific molecular recognition and thereby the portion of the hydrophilicity of the self-assembling building block increased. Consequently, after this inclusion complex formation, the self-assembled structures were transformed to the nanotubular structures with the outer and inner surfaces covered by CDs (Den-CD-NT). Furthermore, the surface CDs could be removed by addition of poly(propylene glycol) (PPG) for pseudorotaxane formation with CDs. Consequently, the nanotubular structure was transformed back to the dendron vesicular structure.

The surface functionality of hierarchically self-assembled Den-CD-NT could be modified in a facile way by using C-6 modified CDs during inclusion complex formation. As shown in Figure 7, a wide range of functional groups including amine, carboxylic, iodo and biotin units could be introduced onto the surface of Den-CD-NT by using various C-6 functionalized CDs.[17,69] Therefore, this non-covalent surface functionalization strategy could provide an opportunity for various applications of Den-CD-NTs. For example, the charged metal precursors could be selectively bound onto the surface of Den-CD-NTs with positive or negative charge by electrostatic interaction. Therefore, the subsequent

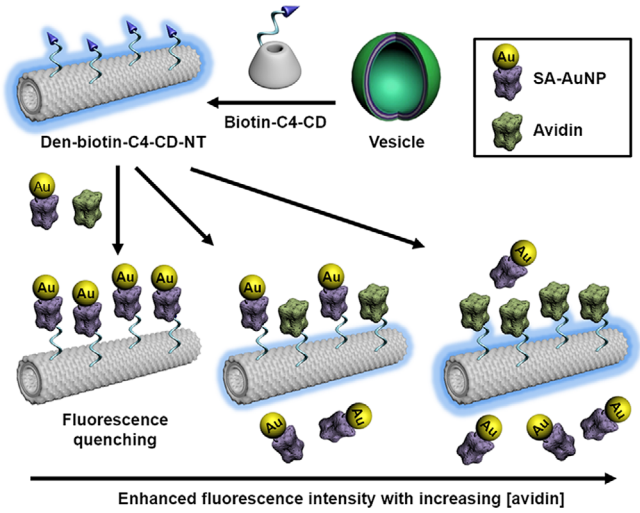


**Figure 8.** Hierarchically self-assembled Den-CD-NTs for a bio-sensory platform.

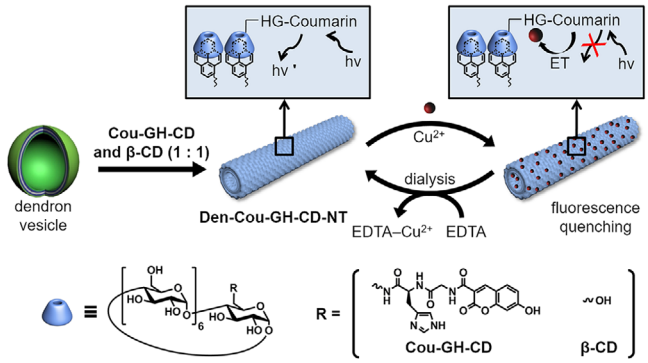


**Figure 9.** Hierarchically self-assembled Den-CD-NTs for a selective and recyclable metal ion sensory platform.

reduction of metal precursors resulted in the formation of metal nanoparticles along with the nanotube. The surface metal nanoparticles could also electronically interact with the pyrene units of Den-CD-NTs, which resulted in quenching of the pyrene fluorescence.[17] Using this fluorescent quenching by interaction between pyrenes and metal nanoparticles, Den-CD-NTs could be employed as a biosensory platform of avidin or Concanavalin A.[17,70] As shown in Figure 8, for the construction of avidin sensory platform, the biotin-functionalized CD was utilized for the formation of Den-biotin-C4-CD-NT. The resulting nanotube exhibited specific binding capability to streptavidin (SA) and avidin. For utilization of the nanotube as an avidin sensory platform in an inhibitory approach, a gold nanoparticle was conjugated with SA unit (SA-AuNP). Upon treatment of SA-AuNP into the nanotube solution, the pyrenic fluorescence of the nanotube was quenched by photo-induced electron transfer from the focal pyrene of dendron unit to the gold nanoparticle of SA-AuNP. Furthermore, upon treatment of SA-AuNP and avidin into Den-biotin-C4-CD-NT solution, the higher concentration of avidin resulted in the higher fluorescence intensity of the nanotube. Therefore, this concept provided a unique route to construct biosensory platforms using hierarchically self-assembled CD-based materials.

Furthermore, the surface of Den-CD-NTs could be functionalized with diverse peptide units for utilization as a sensory platform of metal ions as shown in Figure 9.[19] Den-CD-NT with a surface coumarin unit linked by a GlyHis dipeptide spacer was synthesized by a hierarchical self-assembly and molecular recognition of functional CDs (Den-Cou-GH-CD-NT). The functional groups introduced at the C-6 position of *β*-CD was exposed on the surface of Den-CD-NTs. The GlyHis dipeptide unit originated from the amino terminal Cu and Ni ion binding site was introduced for selective cupric ion binding. Upon addition of cupric ion into the nanotubular solution, the cupric ion was selectively bound to the dipeptide unit on the surface of the nanotube and the fluorescence of coumarin was quenched by

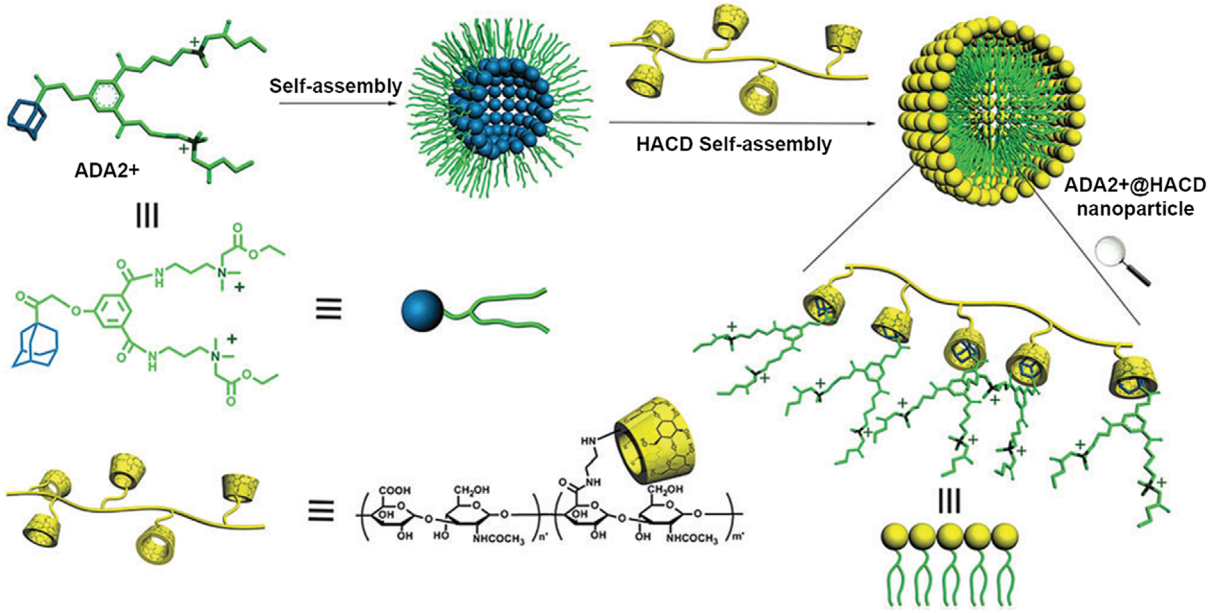


**Figure 10.** Self-assembled CD-based materials for gene carriers. Adapted with permission from ref 74. Y. Yang *et al.*, *Chem. Commun.*, **54**, 8713 (2018). ©2018, The Royal Society of Chemistry.

 

photo-induced electron transfer to cupric ion. Den-Cou-GH-CD-NT showed selective sensory capability towards cupric ion and no fluorescence quenching upon addition of other metal ions. The fluorescence of the pyrene unit in the cavity of CDs was not affected by cupric ion binding and provided a reference fluorescent signal of sensory platform. Furthermore, the nanotubular sensor could be successfully recycled due to the self-assembled architecture.

Self-assembled CD-based materials could also be utilized as a gene carrier.[73,74] The adamantane unit with two positively charged moieties (ADA2+) and hyaluronic acid with $\beta$-CDs (HACD) at the side chain were adopted for controllable gene binding as shown in Figure 10. The ADA2+ molecule self-assembled into a huge micellar structure with a diameter of 600 nm in aqueous phase. After mixing ADA2+ and HACD, the supramolecular nanostructure (ADA2+@HACD) with a diameter of about 130 nm was constructed due to the formation of inclusion complex between the adamantane unit and $\beta$-CD. The nanostructure possessed negatively charged HACD on the shell and positively charged ADA2+ molecules at the core. After addition of plasmid DNA, the positively charged core formed an electrostatic complex with the DNA. Then, hydrolysis of the ester group of ADA2+ induced a controlled release of encapsulated DNA. The ADA2+@HACD could be utilized as a platform for targeted and controlled delivery of DNA.

## 5. CD-based polymers

Along with the capability to form inclusion complex, CDs have been employed for the preparation of polymers with unique structures.[12,13] The CD-based polymers have been utilized for various applications including environmental, catalysis and drug delivery.[12] For the synthesis of CD-based polymers, a variety of controlled polymerization methods could be introduced. CD-based polymers could be classified into several types: linear CD-polymers, CD-terminated polymers, CD-centered star polymers, polymers with pendant CDs and CD-threaded polyrotaxanes.[13]

### 5.1. Linear CD-polymers

Linear CD-polymers contain CD moieties in the main backbone. Using linear CD-polymer (CALAA-01) as shown in Figure 11(a), the first example of systemic administration of cationic polymer/siRNA complex to humans was reported.[75,76] The linear CD-polymer consisted of $\beta$-CDs and cationic linear chain formed inclusion complexes with adamantane-conjugated PEG (PEG-Ad) and human transferrin (Tf-PEG-Ad). Then, the polymer was complexed with siRNA which was designed to inhibit M2 subunit of ribonucleotide reductase, a well-known anticancer target. Although this linear CD-polymer showed successful delivery of siRNA to the cancer cells upon systemic administration, this clinical trial was terminated because the patients suffered from dose-limiting toxicity such as acute immune responses and hypersensitivity.[76,77]

Linear polymers with a backbone consisted of $\beta$-CDs and PEG and side chains of camptothecin (CPT) anticancer drugs were reported as shown in Figure 11(b).[78,79] After conjugation to the linear CD-polymer, the CPT solubility was enhanced more than three orders of magnitude when 6 wt% of drug was conjugated onto the polymer. The conjugated drug was released by hydrolysis at a greatly accelerated rate in human plasma than in buffer solution. Furthermore, the CPT-conjugated linear CD-polymer showed excellent antitumor activity against LS174T human colon carcinoma tumors xenografted in nude mice superior to that of free CPT.

### 5.2. CD-terminated polymers

The terminal CD moieties could be employed for the surface functionalization of the assembled structure by inclusion complex formation with the hydrophobic cavity of CDs. A CD-terminated polymer was reported to produce supramolecular triblock copolymer as shown in Figure 12.[80] The CD-terminated polymer ($\beta$-CD-PDMAEMA) with poly(*N*,*N*-dimethylaminoethyl methacrylate) was prepared by atom transfer radical polymerization (ATRP) at the C-6 position of one glucopyranoside unit of $\beta$-CD. Poly($\varepsilon$-caprolactone) with methacrylate and adamantane moiety at the both terminal respectively (Ada-PCL-MAA) was synthesized by ring-opening polymerization (ROP). Poly(*N*-isopropylacrylamide) with trithiocarbonate (CTA-PNIPAM) was prepared by reversible addition-fragmentation chain transfer polymerization (RAFT). The trithiocarbonate unit of CTA-PNIPAM was a reversible chain transfer agent for RAFT and could be converted to a thiol group by aminolysis. Under basic condition with trimethylamine and ethylene diamine, CTA-PNIPAM was reacted with

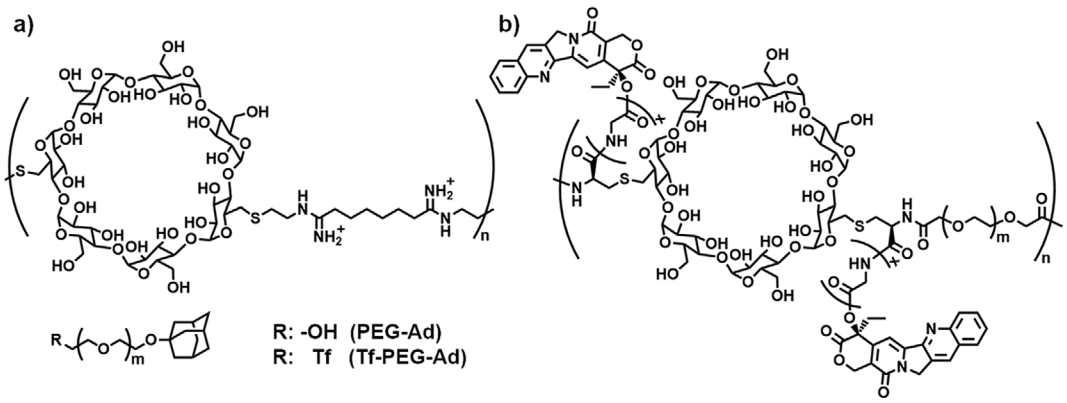


**Figure 11.** Linear CD-polymers for gene (a) and drug (b) delivery.

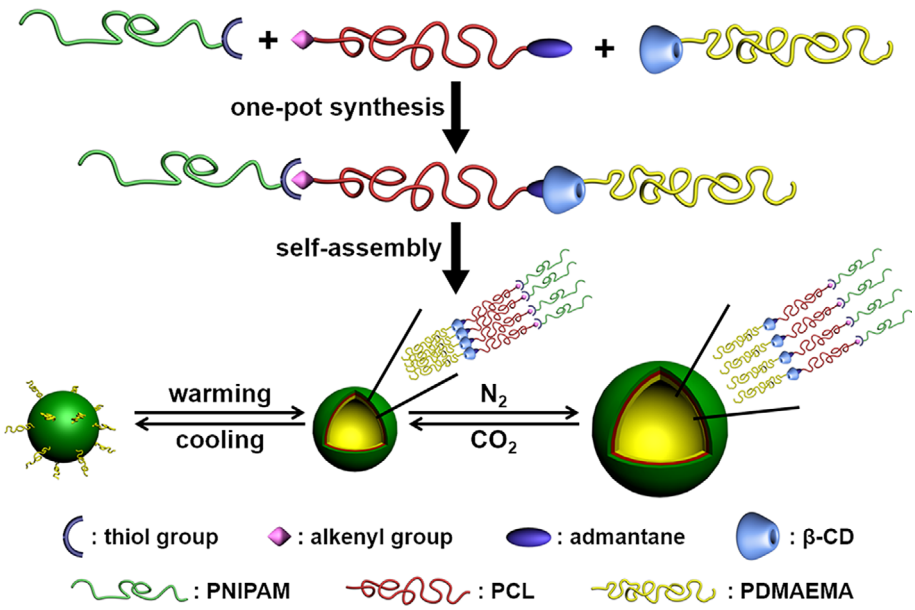


**Figure 12.** Stimuli-responsive supramolecular triblock copolymer by using CD-terminated polymer.

Ada-PCL-MAA to produce PCL-*b*-PNIPAM diblock copolymer with adamantane unit by thiol formation of CTA-PNIPAM and subsequent thiol-ene addition to methacrylate unit. Then, sequential addition of *β*-CD-PDMAEMA resulted in a supramolecular triblock copolymer PNIPAM-*b*-PCL-*b*-PDMAEMA by inclusion complex formation of adamantane unit with CDs. The resulting triblock copolymer self-assembled into a vesicular structure with stimuli-responsive characteristics. The vesicular structure was reversibly changed in response to $CO_2$ gas and temperature. Upon addition of $CO_2$ gas or increase of temperature from 25 to 40 ℃, the vesicular structure was swelled or transformed into spherical micellar structure, respectively. Furthermore, the triblock copolymer showed low cytotoxicity and capability of controlled release triggered by $CO_2$ for utilization as drug carriers.

### 5.3. Polymers with pendant CDs

Polymers with pendant CDs are macromolecules containing numerous CDs at the side chains. Namgung *et al*. reported a self-assembled drug delivery system by using multivalent host-guest complexation between a polymer with pendent CDs and a polymer conjugated with paclitaxel (PTX) drugs.[81] The polymer with pendent CDs (pCD) was prepared by esterification of hydroxyl groups of *β*-CDs with anhydride groups of poly(isobutylene-*alt*-maleic anhydride). The PTX-conjugated polymer with AP-1 targeting peptide (AP-1-pPTX) was synthesized by the reaction of poly(methyl vinyl ether-*alt*-maleic anhydride) with PTX and targeting peptide. AP-1 peptide was introduced for specific targeting of IL-4 receptor of the cancer cell surface. Upon mixing of the two polymers with 1:1 molar ratio of PTX and CD, a self-assembled nanoparticle was produced by multivalent inclusion complex formation between CDs and PTX. The nanoparticles could deliver the PTX drugs into the target cells by both active and passive targeting ways. The ester linkage conjugating PTX and polymers allowed the efficient degradation-induced release of PTX in the cytoplasm of the cancer cells. Therefore, the Ap-1-pPTX/pCD nanoparticles selectively inhibited the growth of tumor with IL-4 receptor *in vivo*.

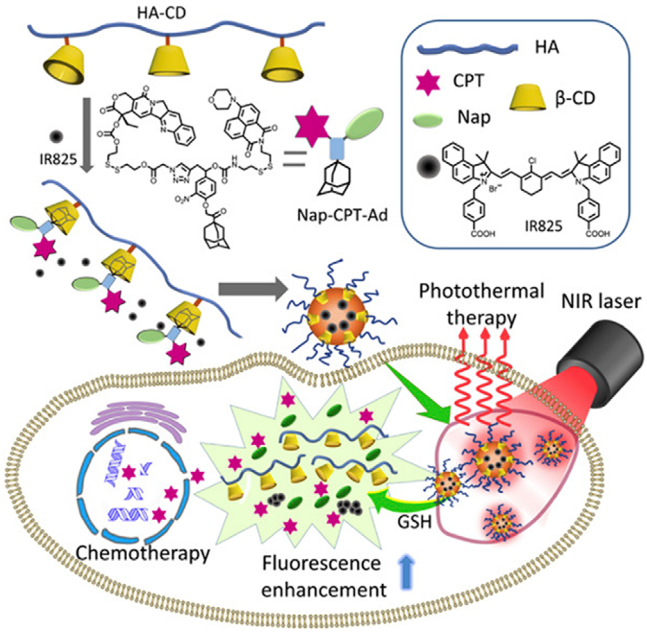


**Figure 13.** HA with pendant CDs for combinational anticancer treatment for chemotherapy and photothermal therapy. Adapted with permission from ref 82, Y. Zhang *et al*., *Biomaterials*, **163**, 14 (2018). ©2018, Elesvier Ltd.

*β*-CDs were introduced at the side chain of hyaluronic acid (HA-CD) for combinational anticancer treatment by chemotherapy and photothermal therapy (Figure 13).[82] The CPT drugs, naphthalimide (Nap) dye and nitrobenzene moiety were conjugated with adamantane unit by disulfide bond (Nap-CPT-Ad). Then, Nap-CPT-Ad and near-IR photosensitizer IR825 were mixed with HA-CD to form nanoparticles. Upon treatment of the nanoparticles into cancer cells, the quenched fluorescence of Nap moiety by nitrobenzene unit was enhanced by intracellular glutathi-

 

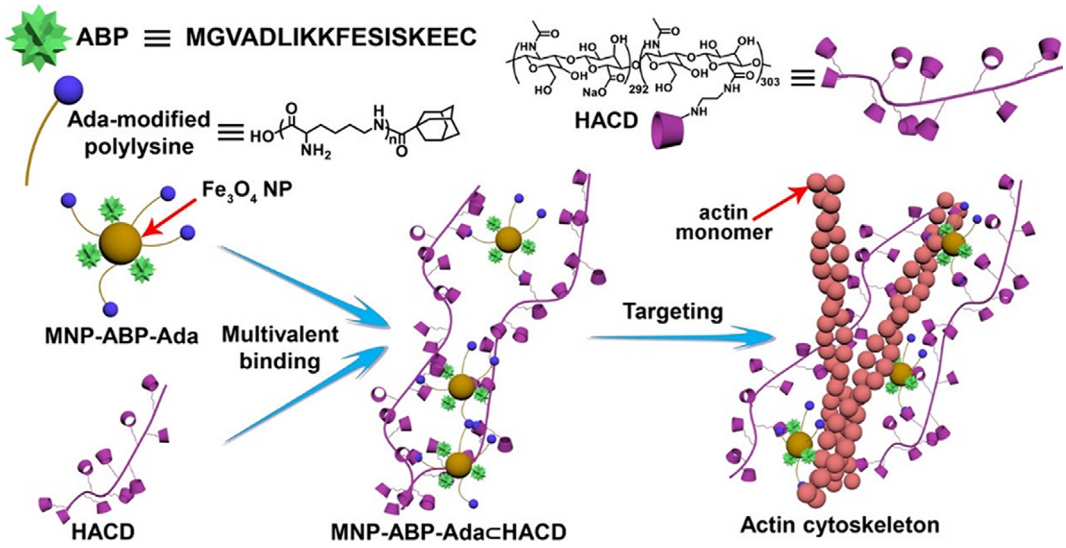


**Figure 14.** HA with pendant CDs for cancer therapy by disruption of actin cytoskeleton. Adapted with permission from ref 83, Q. Yu *et al.*, *ACS Appl. Mater. Interfaces*, **12**, 13709 (2020). ©2020, American Chemical Society.

one (GSH) which cleaves the disulfide bond. Furthermore, the CPT drug molecules are released into the cytoplasm of the cancer cells. Upon irradiation of near-IR laser, the entrapped IR825 photosensitizer effectively convert the near-IR light into heat for photothermal effect. Therefore, the combinational treatment of chemotherapy and photothermal therapy resulted in the effective inhibition of the tumor growth in mice bearing U14 murine cervix cancer cell.

HA with pendant CDs was also employed for cancer therapy by disruption of the tumor actin cytoskeleton as shown in Figure 14.[83] The disruption of actin cytoskeleton is a promising strategy for anticancer therapy, but the severe side effect such as universal toxicity even in normal cells frequently limited its utilization.[84,85] $\beta$-CD grafted HA (HACD) was mixed with iron oxide nanoparticles (MNP-ABP-Ada) containing an actin-binding peptide (ABP) and adamantane-functionalized polylysine molecule to produce a self-assembled nanofiber structure by inclusion complex formation between CDs and adamantane. The nanofiber was selectively targeted the actin cytoskeleton in cancer cells. Furthermore, under alternating magnetic field, severe disruption of actin cytoskeleton occurred by the self-assembled nanofibers, which resulted in efficient death of cancer cells.

### 5.4. CD-threaded polyrotaxanes

CD-threaded polyrotaxanes are typical supramolecules which contain multiple CDs threaded onto the polymer chain end-capped with bulky moieties at both terminals. The mobility of threaded CDs containing multiple hydroxyl groups could provide polyrotaxanes with unique dynamic functionality such as free sliding or rotating of CDs along the polymer backbone.[86] In general, linear polymer chains such as polyethers, polyesters and $\pi$-conjugated polymers could thread CDs on their backbone, which yields polypseudorotaxanes. Then, subsequent conjugation of bulky molecules to the both terminals of the polymer chains could produce CD-threaded polyrotaxanes. The polyrotaxanes could be applied in diverse areas such as drug or gene delivery,

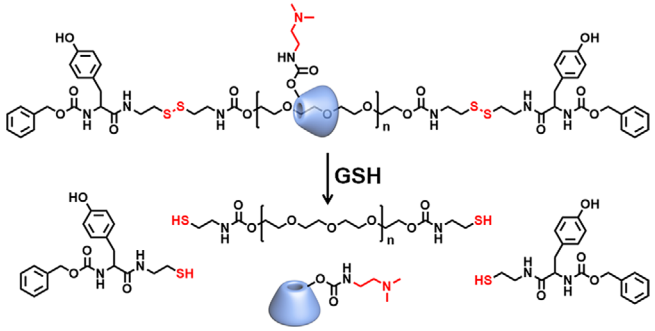


**Figure 15.** GSH-responsive degradation of end-capping molecules of CD-threaded polyrotaxane for gene delivery.

self-healing materials, slide-ring materials and molecular machines, of which details could be found in other reviews.[86-88] In particular, introduction of stimuli-responsive functional groups to polyrotaxanes is a useful way of providing valuable functions. For example, capping molecules were conjugated with redox-responsive disulfide bonds to the both end of polymer chain of *N*,*N*-dimethylaminoethyl-modified $\alpha$-CD/PEG polyrotaxane for efficient dissociation of electrostatic DNA complex in gene delivery as shown in Figure 15.[89,90] Upon internalization into GSH-upregulated cells, the disulfide bonds of polyrotaxanes complexed with DNA were cleaved and the threaded $\alpha$-CDs were removed from the PEG chain. Therefore, the complexed DNA could be released and the gene expression could be enhanced.

## 6. CD-based hydrogels

Hydrogels are crosslinked network which could hold a huge amount of water in their structure. Through introduction of CDs on their networks, the hydrogels can reversibly interact with guest molecules by specific molecular recognition. By utilizing this unique property, CD-based hydrogels with stimuli-responsive molecular recognition and self-healing properties have been widely reported.[14,91,92]

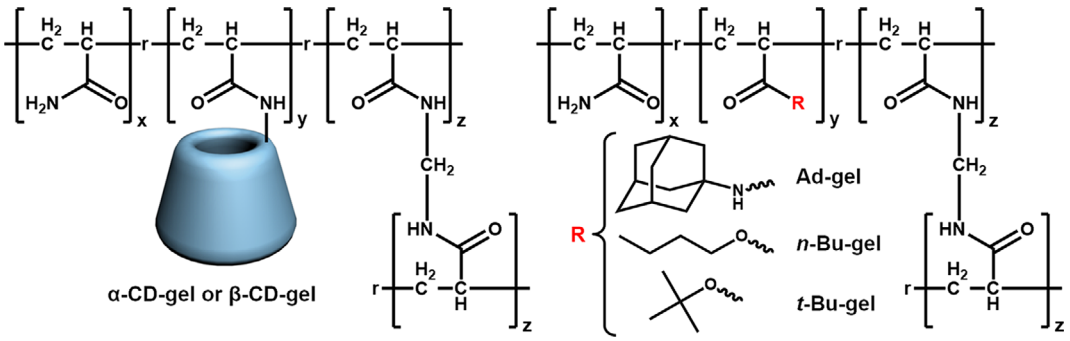


**Figure 16.** Acrylamide-based hydrogels containing CDs and guest molecules for selective macroscopic self-assembly.

### 6.1. CD-based stimuli-responsive hydrogels

Harada *et al*. reported an acrylamide-based hydrogels containing CDs and guest molecules for demonstration of macroscopic self-assembly process by molecular recognition as shown in Figure 16.[91] The hydrogels containing host (*α*- and *β*-CD-gels) and guest molecules (adamantyl, *n*-butyl and *t*-butyl gels) were synthesized from acrylamide, *N*,*N*′-methylenebis(acrylamide). In water, the strong adhesion between *β*-CD-gel and Ad-gel occurred by direct contact or shaking. The *β*-CD-gel or Ad-gel did not show adhesion to a gel with homologous functional group. In the presence of excess *β*-CD or 1-adamantanamine which could mask the adamantane groups or *β*-CD, respectively, no adhesion between Ad-gel and *β*-CD-gel could be observed. Furthermore, the dissociation of the adhesion between *β*-CD-gel and Ad-gel could be achieved at elevated temperature over 90 ℃, which indicates the reversible binding property. By shaking of *α*-CD-gel, *β*-CD-gel, *n*-Bu-gel and *t*-Bu-gel in water, selective adhesion of *β*-CD-gel/*t*-Bu-gel and *α*-CD-gel/*n*-Bu-gel could be obtained by selective molecular recognition of host and guest moieties.

By using this approach, photo-responsive CD-based hydrogel also reported.[93] The acrylamide gel containing guest molecules was prepared using azobenzene moiety (Azo-gel) which has a capability of photoisomerization between *trans*- and *cis*-isomers. The *trans*- and *cis*-azobenzene moiety showed higher affinities toward inclusion complex formation with *α*-CD and *β*-CD, respectively. In water, *α*-CD-gel adhered to *trans*-form Azo-

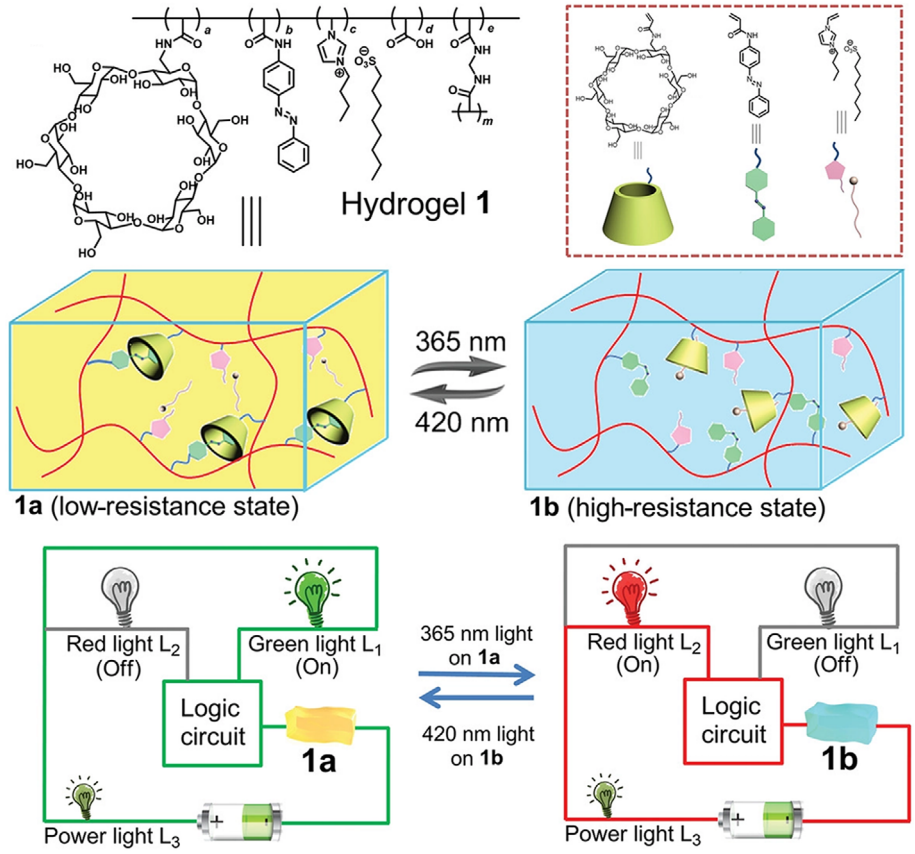


**Figure 17.** CD-based hydrogel for a reversible ion-conducting switch. Adapted with permission from ref 94, H. Wang *et al*., *Adv. Mater*., **31**, 1807328 (2019). ©2019, WILEY-VCH Verlag GmbH & Co. KGaA.

 

gel. Upon irradiation at 365 nm, the adhesion of gels was dissociated by photoisomerization of azobenzene to *cis*-form. Then, photoirradiation of visible light at 430 nm resulted in reassembly of Azo-gels (*trans*-form) and $\alpha$-CD-gels. Furthermore, in the presence of $\beta$-CD-gels, photoirradiation at 365 nm to Azo-gels adhered with $\alpha$-CD-gels induced an exchange reaction to form an adhesion of Azo-gel not with $\alpha$-CD-gel but with $\beta$-CD-gel.

Azobenzene-introduced CD-based hydrogel was adopted for the reversible ion-conducting switch as shown in Figure 17.[94] Onto the matrix of the hydrogel, $\alpha$-CD, ionic liquid and azobenzene were grafted. Upon irradiation at 365 nm, the azobenzene unit was transformed into *cis* form and the complex formation between $\alpha$-CD and the anionic part of the ionic liquid with long alkyl chain dominantly occurred. As the result, the ionic mobility of the hydrogel decreased and the electrical resistance increased. After irradiation at 420 nm, the azobenzene transformed to *trans* isomer and formed an inclusion complex with $\alpha$-CD. These results led the anionic part of the ionic liquid to be released from the CD cavity. Therefore, the resistance of the hydrogel was lowered. By using this photo-responsive conductivity change, the CD-based hydrogel was utilized as a reversible switch of an electric circuit.

Ferrocene was also adopted for the construction of stimuli-responsive CD-based hydrogels.[95] The ferrocene unit is a well-known redox-responsive molecule. The ferrocene unit could internalize into the hydrophobic cavity of $\beta$-CD. By oxidation, the ferrocene unit is converted to a cationic ferrocenium ion, which showed low tendency of inclusion complex formation with $\beta$-CD. Therefore, the polyacrylamide hydrogel containing ferrocene units (Fc-gel) could be selectively attached to the $\beta$-CD-gel by selective inclusion complex formation. After addition of an oxidizing agent (ceric ammonium nitrate, CAN), the ferrocene unit was oxidized, which resulted in disruption of adhesion between Fc-gel and $\beta$-CD-gel. In contrast, efficient adhesion was observed with a polyacrylamide hydrogel containing anionic styrenesulfonic acid sodium salt (SSNa-gel) by electrostatic interaction. By partial immersion of Fc-gel to CAN oxidizing agent, ABC-type macroscopic assembly consisted of $\beta$-CD-gel, Fc-gel and SSNa-gel could be constructed through two different non-covalent interactions.

### 6.2. CD-based self-healable hydrogel

CD-based hydrogel with a capability of specific molecular recognition was applied for the construction of self-healable materials. Self-healing materials have been of great interest due to the capability of improvement of material life-time. Nakahata *et al.* reported self-healable hydrogels by using poly(acrylic acid) with $\beta$-CD (pAA-6$\beta$CD) and ferrocene (pAA-Fc) unit at the side chain (Figure 18).[14,96] Upon mixing of pAA-6$\beta$CD and pAA-Fc, a hydrogel was constructed by inclusion complex formation between ferrocene and $\beta$-CD. The hydrogel exhibited a reversible sol-gel transition by addition of oxidizing (NaClO) and reducing agent (GSH) because only reduced form of ferrocene could be included into the cavity of $\beta$-CD. After 24 hr of cutting in half followed by standing close together, the crack of pAA-6$\beta$CD/pAA-Fc hydrogels disappeared to form a single gel again. The mechanical strength

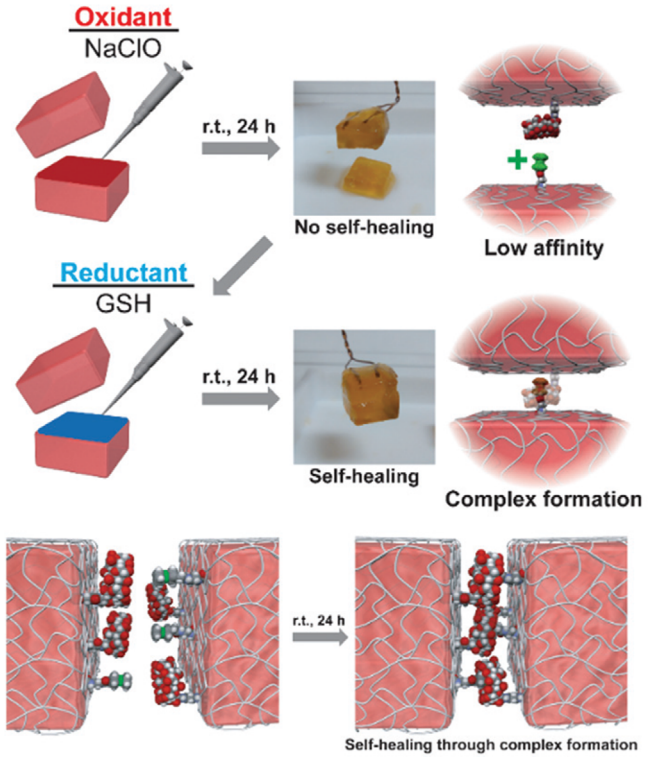


**Figure 18.** Stimuli-responsive self-healable hydrogels using poly(acrylic acid) with $\beta$-CD and ferrocene units. Adapted with permission from ref 14, G. Sinawang *et al.*, *Chem. Commun.*, **56**, 4381 (2020). ©2020, The Royal Society of Chemistry.

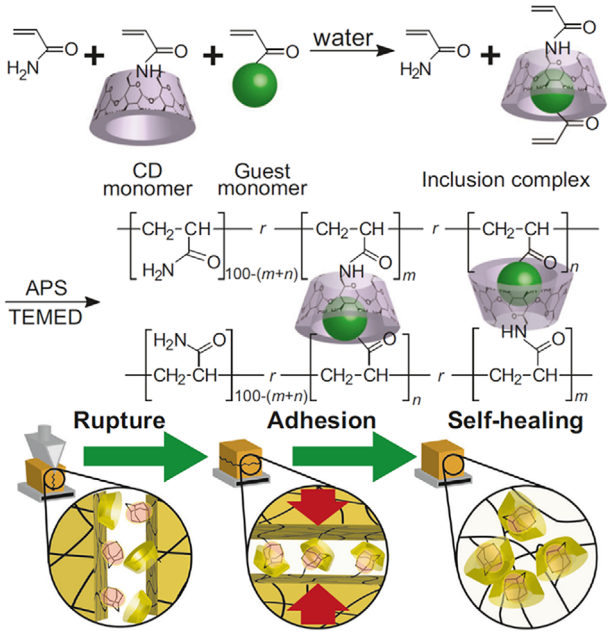


**Figure 19.** Self-healable hydrogels using CD-based polymers with both host and guest molecules at the side chain. Adapted with permission from ref 92. G. Sinawang *et al.*, *Polym. J.*, **52**, 839 (2020). ©2020, Springer Nature.

of the hydrogel was also recovered 84% of initial strength. Upon coating of the cut surface with aqueous solution of oxidizing agent (NaClO), the self-healing could not be observed even after 24 hr. After treatment of GSH solution onto the cut surface oxidized by NaClO, the surface adhered again. Therefore, the hydrogels with CDs and guest ferrocene could be utilized for the construc-

tion of stimuli-responsive self-healing materials.

CD-based polymers with both host and guest molecules at the side chain could be applied for self-healable hydrogels as shown in Figure 19.[92,97,98] For the formation of self-healable supramolecular hydrogels, polyacrylamide polymers ($\beta$CD-Ad gel) were prepared by radical copolymerization of the monomers with $\beta$-CD and adamantane unit, respectively, in aqueous phase. The polymer could form a self-standing hydrogel. The $\beta$CD-Ad gel exhibited a self-healing capability. Cube-shaped $\beta$CD-Ad gels divided into bisection adhered again immediately to form a single gel with recovery of the initial mechanical strength.

## 7. CD hybrids with inorganic nanoparticles

Recently, many researchers reported novel concepts using CDs for organic-inorganic hybrid vehicles for drug delivery. In these researches, CDs are utilized as solubilizing agent or physical trapping units to deliver drugs with low solubility to the desired target site. Furthermore, one of important features of CDs is the stimuli-responsiveness for controlled drug delivery and release on demand by using diverse stimuli such as pH, light, temperature, magnetic field, reducing agents and enzymes. In organic-inorganic hybrid systems, the inorganic nanoparticles can be used for diagnostic application as imaging agents such as computed X-ray tomography (CT), magnetic resonance imaging (MRI) and optical imaging.

### 7.1. CDs for surface functionalization of inorganic nanoparticles

#### 7.1.1. AuNPs with CDs

Gold nanoparticles (AuNPs) have recently attracted interest due to their great potential as drug carriers with unique characteristics of tunable size, stability, and biocompatibility. Furthermore, AuNPs could be utilized as a CT imaging contrast agent. Park *et al.* reported AuNPs with decorated surface by CDs for encapsulating hydrophobic anticancer drugs ($\beta$-lapachone), PEG for enhanced dispersion stability and anti-fouling effect, and anti-epidermal growth factor receptor (anti-EGFR) for specific targeting of cancer cells as shown in Figure 20.[20] In this report, CDs were introduced on the surface of AuNPs by using per-6-thio-6-deoxy-$\beta$-CD, and the $\beta$-lapachone was loaded into the hydrophobic cavity of CDs on the AuNP surface without chemical modification by noncovalent inclusion complex formation. The encapsulated $\beta$-lapachone was released on demand triggered by intracellular GSH which is overexpressed in various cancer cells. The thiol group of GSH removed the thiol ligand from the surface of AuNPs. Consequently, in A549 cells with high GSH level, treated AuNPs released $\beta$-lapachone loaded in the NPs. Furthermore, anti-EGFR active targeting ligand increased the intracellular uptake and apoptosis of cancer cells by AuNPs. The improved radiotherapeutic efficacy of $\beta$-lapachone loaded in AuNPs was demonstrated with *in vivo* study using mice bearing A549 xenograft tumors.[99]

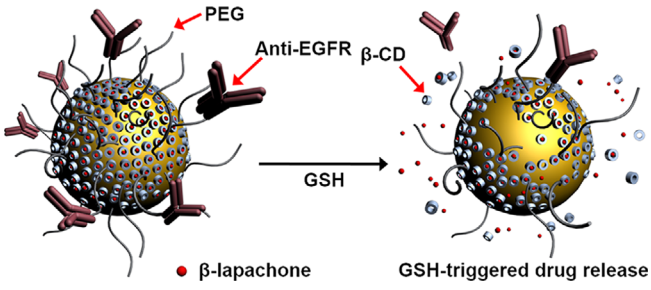


**Figure 20.** Schematic representation of AuNP with surface CDs encapsulating $\beta$-lapachone, PEG and anti-EGFR targeting ligand.

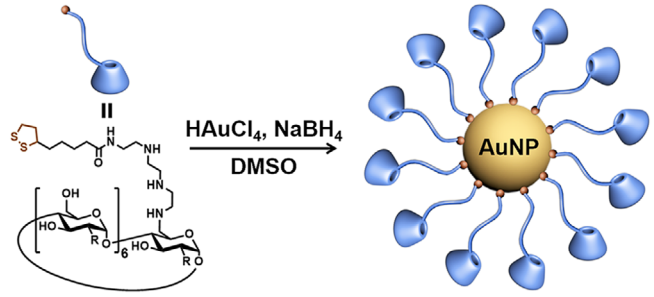


**Figure 21.** AuNPs with surface CDs for gene delivery.

Wang *et al.* reported application of AuNPs with surface CDs for gene delivery as shown in Figure 21.[100] The AuNPs and CDs were linked by oligo(ethylenediamine) chains as a spacer. The oligo(ethylenediamine) chains provided a capability of electrostatic complexation with DNA. The AuNPs formed aggregates upon mixing with DNA, which successfully delivered the plasmid DNA (pEGFPC1) into MCF-7 human breast cancer cells. Without CDs, AuNPs with only oligo(ethylenediamine) group showed weak binding effect with DNA.

Recently, AuNPs decorated with pH-responsive CDs were utilized for photodynamic and photothermal tumor therapy as shown in Figure 22.[101] In this research, the surface of AuNPs was modified by dopamine-functionalized PEG and $\gamma$-CDs with dopamine (DOPA) and amine groups (dCD-$NH_2$). Then, the surface of AuNPs were electrostatically complexed with CDs with negatively charged 2,3-dimethylmaleic acid (DMA) and chlorin e6 (Ce6) moiety (cCD-DMA). Under the tumor environment with low pH (about 6.8), the DMA groups were cleaved from cCD-DMA and the complex was deconstructed. The resulting AuNPs showed highly positive charge and extensive intratumoral uptake by electrostatic interaction with the membrane of tumor cells. Then, AuNPs and Ce6 exhibited photothermal and photodynamic therapeutic efficacy in tumor cells, respectively.

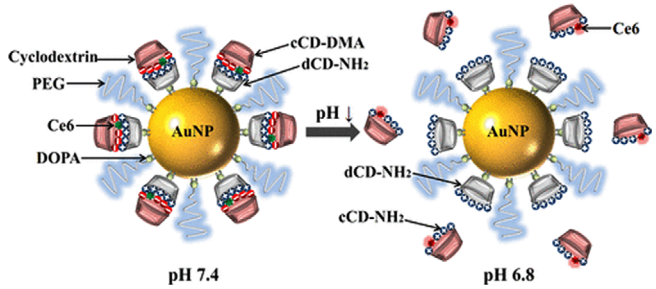


**Figure 22.** AuNPs with pH-responsive CDs for photo-therapeutic efficacy. Adapted with permission from ref 101, M. Koo *et al.*, *ACS Appl. Mater. Interfaces*, **10**, 24450 (2018). ©2018, American Chemical Society.

 

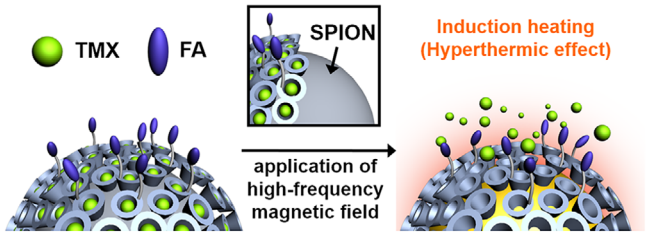


**Figure 23.** Schematic representation of controlled drug release from SPIONs covered with $\beta$-CDs triggered by high-frequency magnetic field.

### 7.1.2. Iron oxide nanoparticles with surface CDs

Superparamagnetic iron oxide nanoparticles (SPIONs) covered with CDs were employed as a triggered drug carrier in response to a high-frequency magnetic field as shown in Figure 23.[102] In this work, the surface of SPIONs with a diameter of 8.2 nm was modified with $\beta$-CDs as a drug container and folic acid as an active targeting ligand for breast cancer cell. Then, tamoxifen, an anticancer drug with efficaciousness against breast cancer cells, was loaded into the CD cavity by inclusion complex formation. The modified SPIONs showed a hydrodynamic diameter of 12.4 nm, high stability in aqueous phase, and low cytotoxicity for normal cells. Using heat induction property of SPIONs upon irradiation of high-frequency magnetic field, the drugs incorporated in the CD cavities were released on demand. Furthermore, the hyperthermic effect of SPIONs showed hyperthermia treatment and, hence, enhanced efficacy of tamoxifen.

## 7.2. Stimuli-responsive CD gatekeepers

Recently, CDs were utilized as a gatekeeper for controlling release of drugs encapsulated in the pores of nanocontainers as well as a host for inclusion complex formation with guest molecules. CDs as gatekeepers could completely block the premature release of guest drug molecules during blood circulation after intravenous injection of nanoparticles. Furthermore, the CD gatekeepers could be removed on demand in response to various stimuli including pH, photo, redox, sugar, enzyme and hypoxia, which could be achieved by precise design of CD gatekeepers and linkers between CDs and nanoparticles. Mesoporous silica nanoparticles (MSNs) are very effective for these approaches due to their ordered porous nature and ease of surface functionalization. The advantages of MSNs with CD gatekeepers include high stability, biocompatibility, low cost for synthesis and controllable particle size and pore diameter.

### 7.2.1. pH-responsive CD gatekeepers

Due to lower pH condition of endo-/lysosomes (pH 4-6) and cancer microenvironment (pH~6.5) than in normal physiological conditions (pH~7.4), the pH change could be a valuable stimulus for controlled release of guest molecules. As shown in Figure 24, pH-responsive CD gatekeepers on the surface of MSNs were reported by formation of polypseudorotaxane consisted of linear poly(ethylene imine) (PEI) and CDs.[21] The orifice of MSN mesopore was modified with linear PEI. Then, the mesopores of MSNs were filled with guest calcein cargo and blocked by threading $\alpha$- or $\gamma$-CDs to the surface PEI chains. At high pH condition, the amine groups of PEI exhibited neutral charge. To the PEI chain, the CDs could be threaded to form polypseudorotaxanes on the surface of MSNs. At pH 5.5, the CDs are dethreaded from the PEI chain and the guest cargo could be released.

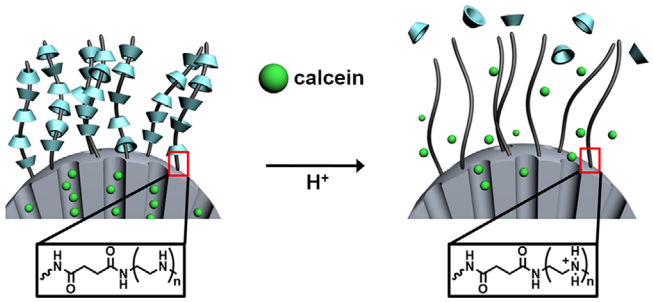


**Figure 24.** PEI-decorated MSNs with CD Gatekeepers for pH-responsive cargo release.

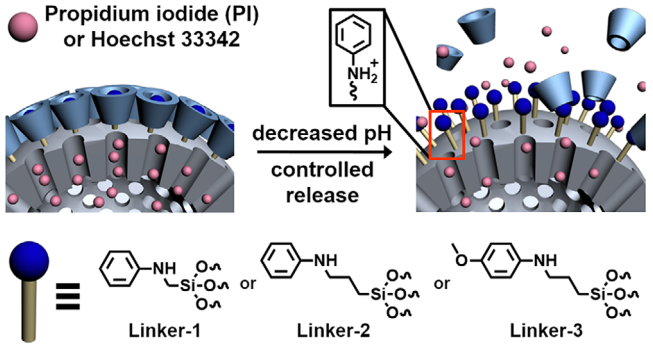


**Figure 25.** pH-responsive CD gatekeepers on hollow MSNs with different linker structures.

Hollow MSNs with pH-responsive CD gatekeepers was also reported as shown in Figure 25.[103] The CD gatekeepers were introduced by non-covalent hydrogen bonding interaction between $\alpha$-CDs and the aniline stalk moiety. The stalk moiety was conjugated to the orifice of MSN mesopores. At neutral pH, the bulky CD molecules are located near the pore orifice by inclusion complex formation with aniline stalk, which blocked the leakage of cargo molecules. At lower pH, the amine group was protonated, which decreases binding affinity of $\alpha$-CDs to the aniline moiety. Therefore, the loaded cargo molecules (PI and Hoechst 33342) were released from the mesopore. The different length of aniline stalk (linker-1 and 2) did not affect blocking and releasing cargo. The introduction of methoxy group into the aniline stalk resulted in the increase of pKa value from about 5 (aniline, linker 2) to 6 (anisidine, linker 3). Therefore, at pH 6, the MSNs with linker 2 showed slow cargo release, while fast cargo release was exhibited with linker 3 at pH 6. The hollow shape of MSNs could encapsulate more than twice cargo molecules than conventional MSNs.

CD-modified CuS nanoparticles was also employed as a gatekeeper of MSNs for imaging and photothermal application (Figure 26).[104] The ultrasmall CuS particle decorated with per-6-thio-$\beta$-CD was introduced onto the fluorescent MSNs with benzimidazole (BM)-grafted surface (FMSN @CuS). The fluorescent character of FMSN@CuS was utilized for cell imaging. The encapsulated DOX in the nanoparticle exhibited zero release at physiological pH and controlled release at a low pH environ-

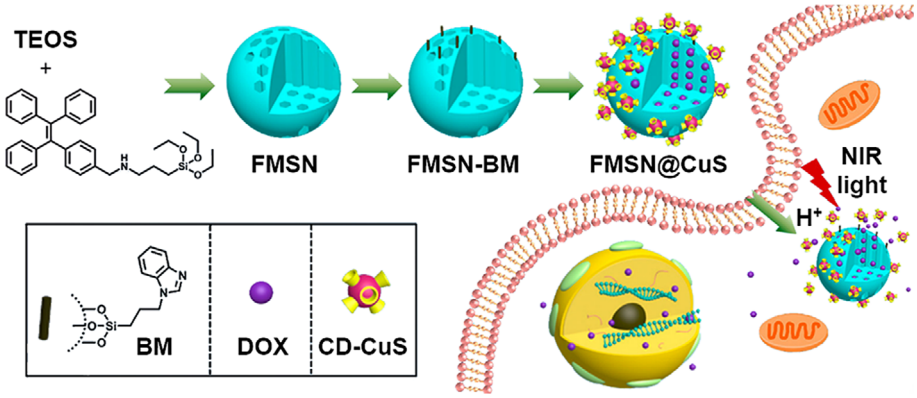


**Figure 26.** MSNs covered by CD-modified CuS nanoparticles for pH-and near-IR-responsive cancer therapy. Adapted with permission from ref 104, Q.-L. Li *et al.*, *ACS Appl. Mater. Interfaces*, **10**, 12155 (2018). ©2018, American Chemical Society.

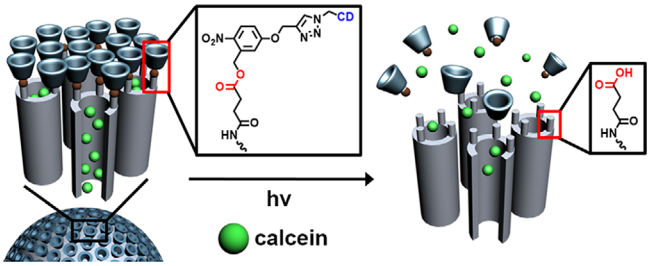


**Figure 27.** Photo-responsive CD gatekeepers on the surface of MSN for controlled cargo release.

ment. Upon irradiation of 808 nm laser light, the photothermal effect synergistically enhanced the therapeutic efficacy of the nanoparticles.

### 7.2.2. Photo-responsive CD gatekeepers

Photoirradiation was an efficient trigger for stimuli-responsive cargo release from MSNs. A photo-responsive CD gatekeeper on MSNs was reported for controlled cargo release (Figure 27).[22] The nitrobenzyl ester was introduced as a photo-responsive linker between CDs and MSNs. Without photoirradiation, the guests were not released. However, upon irradiation at 350 nm, the nitrobenzyl ester unit was cleaved and the guests were released.

Ferris *et al.* also reported photo-responsive CD gatekeepers on MSNs (Figure 28).[105] For photo-responsive cargo release, the surface of MSNs was functionalized with azobenzene moiety. After addition of $\beta$-CDs into the azobenzene-functionalized MSNs, the CD molecules were threaded onto the surface stalks containing azobenzene unit to form pseudorotaxanes. Therefore, the release of rhodamine B cargo molecules from the mesopore of MSNs was blocked by the CD gatekeeper. Upon irradiation, the photoisomerization of azobenzene unit from *trans*- to *cis*-conformation occurred, which induced dissociation of pseudorotaxnes to remove CDs from MSNs and hence the cargo was released.

### 7.2.3. Enzyme-responsive CD gatekeepers

The CD-covered MSNs could be triggered to release of encapsulated cargos by various enzymes including $\alpha$-amylase, lipase, porcine liver esterase. CD gatekeepers in response to enzymatic degradation by $\alpha$-amylase or lipase were demonstrated as shown in Figure 29.[23] The CD gatekeepers were connected to the MSN surface by ester-containing stalk moiety. Upon addition of $\alpha$-amylase which is highly associated with acute pancreatitis, the surface CD gatekeepers of MSNs were degraded and the encapsulated cargos were release from the MSN pore. On the other hand, addition of denatured $\alpha$-amylase did not induce a cargo release. Furthermore, the same MSNs with surface CD gatekeepers could exhibit lipase-responsive cargo release. Upon addition of lipase, the ester bond of the stalk part was degraded and the surface CD gatekeeper was removed from the surface of MSNs. Therefore, the MSNs with CD gatekeepers could be utilized for the controlled cargo release system by dual-enzyme responsive triggering.

Porcine liver esterase can serve as a trigger for stimuli-respon-

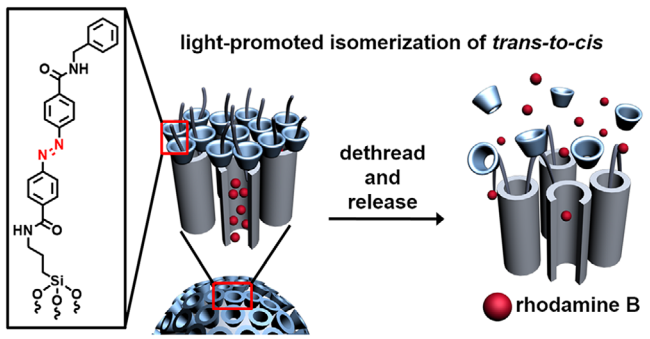


**Figure 28.** Photo-responsive CD gatekeepers on MSNs with azobenzene stalk for controlled cargo release.

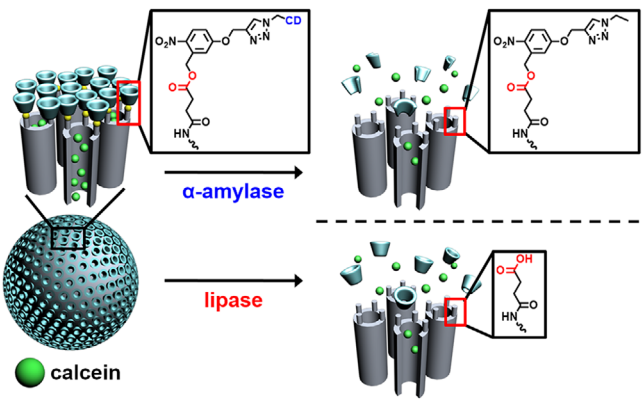


**Figure 29.** MSNs with CD gatekeepers for stimuli-responsive cargo release by $\alpha$-amylase and lipase.

 

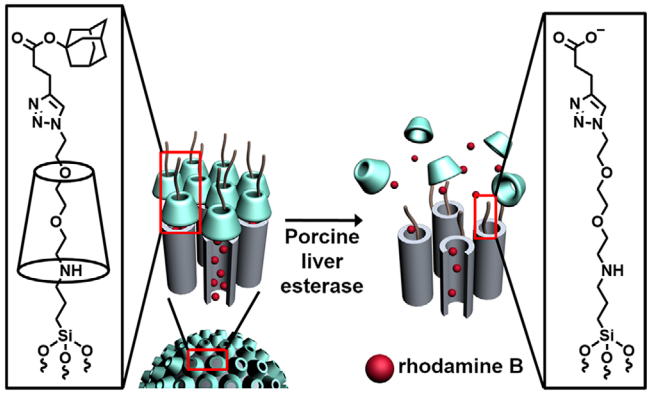


**Figure 30.** Enzyme-responsive cargo release of MSNs with CD rotaxane on the surface.

sive cargo release of MSNs with CD gatekeepers (Figure 30).[106] The MSNs with tri(ethylene glycol) stalk were mixed with $\alpha$-CDs following cargo encapsulation in the pore. Then, the adamantane unit was introduced at the terminal of tri(ethylene glycol) stalk to form rotaxane. The surface CD rotaxane blocked the cargo release without any enzymes. Upon addition of porcine liver esterase, the ester bond between adamantane and tri(ethylene glycol) was cleaved and the CD molecules of the rotaxane were removed from the MSN surface. Thereby, the entrapped cargo was released. When the ester bond of the rotaxane was replaced by amide bond, the porcine liver esterase did not trigger the cargo release.

### 7.2.4. Sugar-responsive CD gatekeepers

Various sugar molecules could trigger the cargo release from MSNs (Figure 31).[107] The phenylboronic acid at the surface of MSNs was conjugated with $\beta$-CDs (Si-PB-CD). The 1,2-diol of the glucose unit of CDs could selectively form a boronic ester with phenylboronic acid on the surface of MSNs in slightly basic condition. The CD gatekeepers on Si-PB-CD were effective to keep the guest cargos in the nanoparticle. After addition of fructose or galactose unit which shows higher binding constant toward phenylboronic acid than glucose moiety, the surface CD gatekeeper was removed by boronic ester formation of phenylboronic acid on the surface of MSNs with fructose or galactose unit. Therefore, the entrapped cargos were released. By using glucose and mannose with similar and lower binding constant, respectively, the cargo release was not triggered.

### 7.2.5. Redox-responsive CD gatekeepers

Redox potential could be an efficient stimulus for controlled release of entrapped guests from MSNs.[108] The surface of MSNs

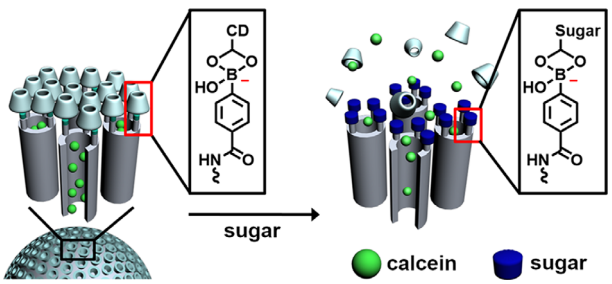


**Figure 31.** Sugar-responsive CD gatekeeper on MSN surface.

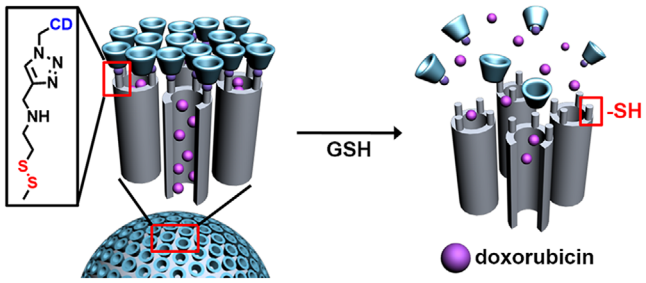


**Figure 32.** GSH-responsive MSNs with CD gatekeepers.

was decorated with ferrocenecarboxylic acid unit. After encapsulation of guest cargo in the mesopore, $\beta$-CDs were added to form a host-guest complex with the surface ferrocene units. The electrochemical oxidation of the ferrocene moiety induced dissociation of inclusion complex of ferrocene with $\beta$-CD, which resulted in the release of entrapped cargo.

GSH is a biological reducing agent upregulated in various cancer cells and is considered as a valuable target for cancer treatment. GSH could be applied as a trigger for redox-responsive cargo release from MSNs with CD gatekeepers. Kim *et al.* reported GSH-responsive MSNs by introducing disulfide bond at the stalk moiety connecting CD gatekeepers and MSN surface as shown in Figure 32.[24] The surface CD gatekeepers on MSNs (Si-SS-CD) could keep the entrapped guest from release without stimuli. Upon addition of GSH, the disulfide moiety was reduced to form two thiol units and the CD gatekeepers were removed from the MSN surface, which resulted in release of entrapped guests in the MSN pores. After treatment of PEGylated MSN (Si-SS-CD-PEG) with DOX to A549 cancer cells, the entrapped DOX was released in the cancer cells and the dose-dependent cytotoxicity toward A549 cells was observed.

### 7.2.6. Hypoxia-responsive CD gatekeepers

Hypoxia is a state with low oxygen concentration and mostly developed in malignant solid tumors which are resistant to chemo- and radiotherapy.[109-111] Hypoxic tumor tissues exhibit significant difference in reduction potential and the biological activity of numerous enzymes than normal tissues. In particular, the NAD(P)H:quinone oxidoreductase 1 (NQO1), a reductase up-regulated in various cancer cells, shows higher enzymatic activity in hypoxia than that in normoxia. Therefore, the NQO1-responsive molecules could be utilized for the selective treatment of hypoxic cancer cells. Lee *et al.* reported MSNs with CD gatekeepers with azobenzene stalk unit for hypoxia-responsive drug release and selective cancer treatment as shown in Figure 33.[25] The surface of MSNs with amino groups was functionalized with azobenzene units containing alkyne moiety. After encapsulation of cargo drugs in the mesopore of MSNs, the CD gatekeeper was introduced on the surface of MSNs followed by PEGylation to yield Si-Azo-CD-PEG. Without NQO1, CD gatekeepers of Si-Azo-CD-PEG inhibited cargo release. In addition, in normoxic condition, Si-Azo-CD-PEG showed negligible release of entrapped cargo even with NQO1. However, in hypoxic condition, the entrapped DOX in Si-Azo-CD-PEG was released by NQO1 addition. The nanoparticles exhibited selective therapeutic efficacy in MDA-MB-231 cells with overexpressed NQO1 under hypoxic condition *in vitro* and *in vivo*.

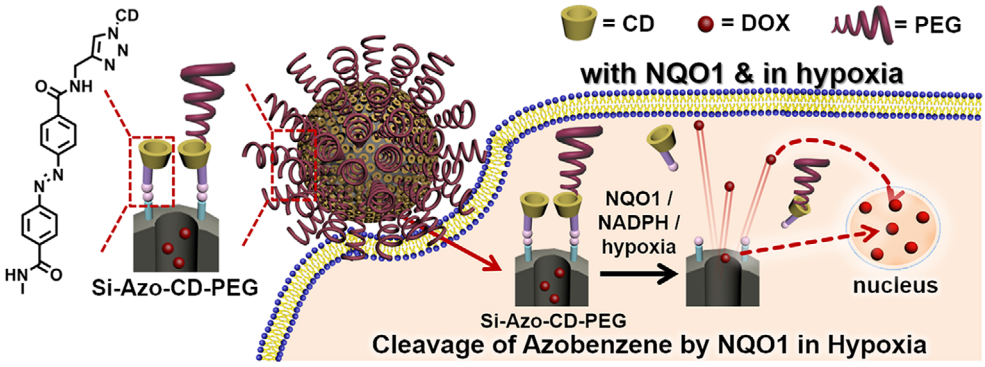


**Figure 33.** Hypoxia-targeted MSNs with CD gatekeepers.

## 8. Concluding remarks

We discussed new horizon in the application of a variety of CD-based molecules, polymers and nanomaterials. Due to the capability of inclusion complex formation, synthetic versatility to numerous architectures and high biocompatibility, CDs have been employed as a key skeleton of diverse molecular derivatives which have been widely applied in the area of biotechnology. In addition, CDs are useful building blocks for high polymers and functional materials. The selective molecular recognition could provide a unique way to construct self-assembled nanostructures or polyrotaxanes with diverse functions. The numerous hydroxyl groups of CDs could be used to produce a variety of highly functionalized molecules and polymers which are useful for diverse bio-applications. Further, CDs can be employed as valuable building blocks of self-assembled functional nanomaterials. In particular, the CD-containing materials could be adopted for unique stimuli-responsive and self-healable hydrogels by using a motif of host-guest molecular recognition. Moreover, CD hybrids with inorganic nanoparticles have been widely applied for efficient stimuli-responsive on-off drug carriers. With an advent of new methods of building up a variety of complex CD architectures in facile ways, CDs find a new way of diverse applications.

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