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<th>Engineered Hybrid Nanoparticles for On-Demand Diagnostics and Therapeutics</th>
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<td>Nguyen, Kim Truc; Zhao, Yanli</td>
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Engineered Hybrid Nanoparticles for On-Demand Diagnostics and Therapeutics

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CONSPECTUS: Together with the simultaneous development of nanomaterials and molecular biology, the bionano interface brings about various applications of hybrid nanoparticles in nanomedicine. The hybrid nanoparticles not only present properties of the individual components but also show synergistic effects for specialized applications. Thus, the development of advanced hybrid nanoparticles for targeted and on-demand diagnostics and therapies of diseases has rapidly become a hot research topic in nanomedicine. The research focus is to fabricate novel classes of programmable hybrid nanoparticles that are precisely engineered to maximize drug concentrations in diseased cells, leading to enhanced efficacy and reduced side effects of chemotherapy for the disease treatment. In particular, the hybrid nanoparticle platforms can simultaneously target diseased cells, enable the location to be imaged by optical methods, and release therapeutic drugs to the diseased cells by command.

This Account specially discusses the rational fabrication of integrated hybrid nanoparticles and their applications in diagnostics and therapeutics. For diagnostics applications, hybrid nanoparticles can be utilized as imaging agents that enable detailed visualization at the molecular level. By the use of suitable targeting ligands incorporated on the nanoparticles, targeted optical imaging may be feasible with improved performance. Novel imaging techniques such as multiphoton excitation and photoacoustic imaging using near-infrared light have been developed using the intrinsic properties of particular nanoparticles. The use of longer-wavelength excitation sources allows deeper penetration into the human body for disease diagnostics and at the same time reduces the adverse effects on normal tissues. Furthermore, multimodal imaging techniques have been achieved by combining several types of components in nanoparticles, offering higher accuracy and better spatial views, with the aim of detecting life-threatening diseases before symptoms appear. For therapeutics applications, various nanoparticle-based treatment methods such as photodynamic therapy, drug delivery, and gene delivery have been developed. The intrinsic ability of organic nanoparticles to generate reactive oxygen species has been utilized for photodynamic therapy, and mesoporous silica nanoparticles have been widely used for drug loading and controlled delivery. Herein, the development of controlled-release systems that can specifically deliver drug molecules to target cells and release then upon triggering is highlighted. By control of the release of loaded drug molecules at precise sites (e.g., cancer cells or malignant tumors), side effects of the drugs are minimized. This approach provides better control and higher efficacy of drugs in the human body. Future personalized medicine is also feasible through gene delivery methods. Specific DNA/RNA-carrying nanoparticles are able to deliver them to target cells to obtain desired properties. This development may create an evolution in current medicine, leading to more personalized healthcare systems that can reduce the population screening process and also the duration of drug evaluation. Furthermore, nanoparticles can be incorporated with various components that can be used for simultaneous diagnostics and therapeutics. These multifunctional theranostic nanoparticles enable real-time monitoring of treatment process for more efficient therapy.

1. INTRODUCTION

The development of modern analytical systems allows us to obtain data with more accuracy and higher resolution, paving the way for tremendous investigations of nanomaterials. New nanomaterials are continuously being created and present a wide range of applications in electronics, catalysis, energy harvesting, biomedicine, etc.¹,² Nanomaterials can be categorized into different shapes, sizes, and material components. By tuning of these aspects, new properties of nanomaterials can be generated for specific applications. In particular, the integration of biological molecules with nanomaterials is highly expected to bring about new prospects for nanotechnology in future medicine.³

When the ease of synthesis and marvelous functions that different types of nanomaterials can provide are taken into account, the development of various nanoparticles for applications in medicine is a major research focus. Nano-
particles with a suitable size range have been fabricated for blood circulation and cell uptake (Figure 1). In this Account, we mainly discuss the bioapplications of some representative hybrid nanoparticles, including mesoporous silica nanoparticles (MSNPs) and organic/metalllic nanoparticles. Each type of nanoparticle possesses unique features for modification and utilization in diagnostics and therapeutics. In the following subsections, we briefly introduce these unique properties and functionalities that enable medical applications of these integrated nanoparticles.

1.1. Mesoporous Silica Nanoparticles

MSNPs are silica materials containing mesopores with pore diameters in the range of 2 to 50 nm. Until the early 1990s, the introduction of MCM-41-type porous silica perked up the applications of MSNPs in catalysis by making use of its large surface area as well as the ease of adjusting the pore size.4 In addition to catalysis, potential medical applications of MSNPs were also recognized because of its good biocompatibility.5 MSNPs with diameters ranging between a few and hundreds of nanometers were reported to be able to circulate within blood vessels and undergo endocytosis into cells. On account of their porous structure, MSNPs can easily serve as nanocarriers in which drug molecules can be loaded inside the pores in doses sufficient for high therapeutic efficacy. Isoprofen was first successfully loaded into MCM-41 with drug/nanocarrier weight ratios of up to 30%.6 To enhance the targeting effect (i.e., the ability to deliver drugs to specific cells), the surface of MSNPs can be functionalized with particular ligands that can selectively bind to corresponding receptors on the targeted cells. Since the surface of MSNPs is covered with hydroxyl groups, the modification of ligands on MSNPs is relatively easy.7,8 Typically, side effects of anticancer drugs on noncancerous cells could be minimized by controlled drug release from MSNPs to cancer cells. MSNP-based nanocarriers are normally equipped with responsive species that can be controlled by external or internal stimuli for triggered drug release into specific cells. This controlled-release mechanism further enhances the selectivity and effectiveness of drug delivery. In addition, drug molecules are sealed inside the pores, protecting them from attack by the bloodstream environment, and then released at the desired site in the human body.

A similar type of silica material, so-called hollow mesoporous silica nanoparticles (HMSNPs), possesses the same advantages as MSNPs. The additional advantage of HMSNPs lies in their unique morphology: the core of HMSNPs is void, allowing them to accommodate more drugs.9 Therefore, HMSNPs provide much better drug loading capacity and further reduce the amount of silica used, minimizing the intake of unnecessary foreign materials into biological systems.

1.2. Organic and Metallic Nanoparticles

Another common type of nanoparticles that has been used widely in bioapplications is organic nanoparticles consisting mainly of organic species. For instance, some polymer-based nanoparticles have been approved as drug carriers by the U.S. Food and Drug Administration (FDA) and applied in clinical cancer therapy.10,11 Typically, self-assembly and host–guest interactions are the main factors that assemble organic building blocks together into organic nanoparticles. The reprecipitation technique is usually employed to prepare organic nanoparticles in aqueous media.

Metallic nanoparticles are metal or metal oxide nanocrystals. Because of their intrinsic properties, they have been used widely in diagnostic and therapeutic applications. For example, magnetic nanoparticles render magnetic resonance imaging (MRI) signal-enhanced contrast,12 gold nanorods are effective for photothermal therapy,13 and upconversion nanoparticles are useful for biolabeling and bioimaging.14 Usually, the surface of these nanocrystals is covered by organic ligands that prevent them from self-aggregation. By modifying these organic ligands, enhanced biocompatibility of metallic nanoparticles could be expected. Thus, rational modification methods for improving the bioapplicability of metallic nanoparticles are needed.

2. DIAGNOSIS

Detecting diseases at an early stage is always desired for the treatment process. Especially in cancer treatment, early detection could prevent tumor growth and metastasis, giving patients a much higher chance for complete recovery. The research focuses in diagnosis include (1) biosensing to detect disease-related molecular processes and (2) bioimaging to visualize specific tumor sites in the human body. The detection and biosensing of many disease biomarkers have been well-reported and summarized in the literature.15,16

2.1. Biosensing

2.2.1. Zinc(II) Ion Detection. An increase in the zinc ion concentration in the body is a symptom related to many diseases such as Alzheimer’s disease, Parkinson’s disease, amyotrophic lateral sclerosis, and epilepsy. Our group developed a molecular probe based on a bipyridine derivative (GBC) that can selectively bind with Zn(II) ions and become two-photon-active.17 The bioimaging of this molecular probe was demonstrated in vitro as well as in vivo. In another design for zinc detection by appropriate location of donor–acceptor moieties on a molecular structure, the obtained architecture facilitates tuning of its three-photon-active cross section as well as its emission color. In the presence of exogenous Zn(II) ions in live HeLa cells, the emission color of the probe changes from green to red under excitation by an infrared laser at a wavelength of 1200 nm.18 On account of its low cytotoxicity and good cell permeability, this three-photon probe is a potential candidate for future real-time detection of Zn(II) ions in vivo.

2.1.2. Hydrogen Peroxide Detection. Hydrogen peroxide is a byproduct of various biological pathways on account of its ability to generate reactive oxygen species (ROS). Excess H2O2 may lead to different kinds of illnesses such as atherosclerosis, heart attack, and cancer. Therefore, a suitable quantitative assay for H2O2 sensing is of practical importance in
biomedicine. Nevertheless, rapid response and high sensitivity as well as in situ detection of H$_2$O$_2$ still remain great challenges. Metallic nanoparticles have been widely used in sensing systems. Taking advantage of these nanomaterials, periodic mesoporous silica (PMS)-coated reduced graphene oxide (RGO) embedded with gold nanoparticles (AuNPs) was synthesized, and the hybrid was then coated on a glassy carbon (GC) electrode (RGO-PMS@AuNPs/GC) for the detection of H$_2$O$_2$ (Figure 2). This system showed a rapid and selective response toward H$_2$O$_2$ in the presence of other common electroactive species, including glutathione, ascorbic acid, uric acid, and l-cysteine. The in vitro detection of H$_2$O$_2$ generated by cancer cells such as HeLa and HepG2 was also compared with that of normal HEK293 cells. H$_2$O$_2$ generated from cancer cell lines was clearly distinguished from that of normal cell lines, which proved the effectiveness of the RGO-PMS@AuNPs/GC probe in H$_2$O$_2$ sensing. Upon modification with the cancer-targeting ligand folic acid (FA) on the surface of the mesoporous silica layer, the obtained hybrid showed unprecedented peroxidase-like activity. This mimetic enzyme was successfully employed for H$_2$O$_2$- and ascorbic-acid-mediated therapeutics of cancer cells.

### 2.2. Bioimaging

Conventional medical imaging methods mainly rely on anatomical imaging. These clinical imaging techniques normally include X-ray computed tomography (CT), positron emission tomography (PET), fluorescence microscopy (FM), fluorescence reflectance imaging (FRI), bioluminescence imaging (BLI), ultrasound microscopy/imaging (UM/UI), photoacoustic microscopy/tomography (PAM/PAT), and MRI. A brief summary of these imaging techniques featuring the measuring signal, resolution, depth, and nanomaterials used is shown in Table 1. In order to identify the lesions, a database of different types of images is needed, especially in cases where the lesions are small and unable to be spotted by the naked eye. Conventional anatomical imaging techniques need further improvement to be suitable for the early detection of life-threatening diseases such as cancer, neurological diseases, and cardiovascular diseases. Thus, optical imaging is one of the promising solutions for early detection because of its capability for visualization at the cellular level, which dramatically enhances the specificity and resolution of the obtained images. In this context, nanoparticles have been highly anticipated for use as targeting imaging agents. Achieving this purpose usually requires nanosystems (e.g., liposomes) with short blood circulation half-lives and rapid exchange between biological compartments, which are facilitated through the enhanced permeability and retention (EPR) effect and active targeting. Different types of nanosystems, including organic nanoparticles, upconversion nanoparticles, and graphene-based hybrids, have been developed for bioimaging. These agents demonstrate flexibility for several types of imaging methods such as fluorescence, Raman, photoacoustic, and multiphoton imaging. Furthermore, dual-mode imaging methods have also been established to offer more accurate and multimode detection.

#### 2.2.1. Fluorescence Imaging

Conventional imaging agents usually come in the form of organic dyes that can be highly affected by the internal environment of living systems, such as hydrolysis and nucleophile attack, which dramatically reduce the efficiency as well as targeting effect of these dyes. In order to protect these dyes in a dynamic environment, GO-wrapped MSNs were developed as efficient dye-protecting vessels. The zwitterionic dye molecule bis(2,4,6-trihydroxyphenyl)squaraine was used as a cargo model. After the dye is loaded into the mesopores of the MSNs, electrostatic interactions between GO and the MSNP surface bring the GO layers closer, and herein the GO layers serve as a blanket to seal the pores and prevent the dye from attacking. The clear accumulation of dye-loaded hybrids in HeLa cell cytoplasm was observed via fluorescent imaging (Figure 3), demonstrating a promising application of these vessels in cellular bioimaging.

A color-tunable fluorescent dye was developed via hierarchical self-assembly of a cyanostilbene–naphthalimide dyad. The dual fluorescent properties originate from the Z-to-E photoisomerization of the cyanostilbene unit upon light irradiation. This transformation causes morphological disorder in the molecular self-assembly along with gradual emission changes from yellow through green and finally to blue. The color-tunable bioimaging of HeLa cells was successfully carried out using this unimolecular system. The present results pave the path for the future design of smart systems with tunable luminescent color.

#### 2.2.2. Photoacoustic Imaging

Although many imaging agents have been developed, the quest for deep-tissue imaging

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**Table 1. Measuring Signal, Resolution, Depth, and Nanomaterials Used in Common Clinical Imaging Techniques**

<table>
<thead>
<tr>
<th>modality</th>
<th>measuring signal</th>
<th>resolution</th>
<th>depth</th>
<th>commonly used nanomaterials</th>
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</thead>
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<tr>
<td>X-ray CT</td>
<td>X-rays</td>
<td>50 μm</td>
<td>no limit</td>
<td>iron oxide-doped nanomaterials, iodinated nanoparticles</td>
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<tr>
<td>PET</td>
<td>positrons from radioactive nuclides</td>
<td>1–2 mm</td>
<td>no limit</td>
<td>nanoparticles with radioisotopes such as $^{18}$F, $^{11}$C, and $^{44}$Cu</td>
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<tr>
<td>UM/UI</td>
<td>sound</td>
<td>50 μm</td>
<td>mm to cm</td>
<td>microbubbles, emulsions, polystyrene beads</td>
</tr>
<tr>
<td>PAM/PAT</td>
<td>sound</td>
<td>mm to cm</td>
<td>mm to cm</td>
<td>carbon nanotubes, dye-loaded nanomaterials, gold nanomaterials</td>
</tr>
<tr>
<td>MRI</td>
<td>magnetic field variations</td>
<td>25–100 μm</td>
<td>no limit</td>
<td>iron oxide nanoparticles, Gd(III)-doped nanoparticles</td>
</tr>
<tr>
<td>FM</td>
<td>light</td>
<td>1–3 mm</td>
<td>&lt;2 mm</td>
<td>dye-loaded nanoparticles, upconversion nanomaterials, carbon-based materials</td>
</tr>
<tr>
<td>FRI</td>
<td>light</td>
<td>&lt;1 cm</td>
<td>&lt;1 cm</td>
<td></td>
</tr>
<tr>
<td>BLI</td>
<td>light</td>
<td>cm scale</td>
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agents that can provide high contrast and good spatial resolution is still ongoing. Photoacoustic imaging as a noninvasive imaging modality to achieve structural and functional imaging relies on the photoacoustic effect. Upconversion nanoparticles have been applied in bioimaging on account of their high luminescence efficiency and good biocompatibility. However, strong tissue absorption is the main drawback for the luminescence signal to be used in deep-tissue imaging. Upon modification of oleic acid-stabilized upconversion nanoparticles with α-cyclodextrin (α-CD), the luminescence of the α-CD modified upconversion nanoparticles (UC-α-CD) was quenched severely, and at the same time, thermal expansion of UC-α-CD in water was observed. 27 A combination of solvent-induced nonradiative relaxation and increased thermal conductivity was proposed for the enhancement in the photoacoustic signal generation of UC-α-CD. Photoacoustic imaging using a 980 nm nanosecond pulsed laser with a power density of 0.5 mJ cm\(^{-2}\) demonstrated excellent in vivo imaging capability of UC-α-CD in live mouse (Figure 4).

In addition, a method to produce contrast agents for photoacoustic signal enhancement was developed by coating GO sheets on silica-covered AuNPs. 28

2.2.3. Multimodal Imaging. Multimodal imaging with two or more imaging modalities integrated together could provide synergistic strengths of the individual modalities while overcoming their limitations. This method enables us to precisely visualize and delineate structural information on the studied subject. As an example, we have synthesized a nanosandwich hybrid capable of two-photon and photoacoustic dual-mode imaging. 29 A two-photon-active dye, 4-(4-diethylaminostyryl)-1-methylpyridinium iodide, was loaded into the mesopores of silica-coated GO to afford the final hybrid, denoted as PAA@NS1. This hybrid was then sealed with poly(acrylic acid) to prevent dye leakage. High-resolution two-photon imaging of the hybrid in HeLa cells was demonstrated, and its photoacoustic imaging at depths of 4–8 mm in a real tissue model was also realized (Figure 5).

Another example of multimodal imaging using the combination of in vitro Raman imaging and fluorescence imaging was developed with a pillararene–GO hybrid. 30 The assembly of amphiphilic pillaranere onto the GO surface provides a cavity that can be used for loading of dye molecules based on host–guest complexation. Effective in vitro Raman and fluorescence imaging elucidated the potential of the hybrid for dual-mode imaging. By varying the combination of imaging agents and therapeutic agents, one can rationally design novel theranostic nanoparticles.

3. THERAPY

The advantage of using nanoparticles in therapeutics relies on their effectiveness in targeted drug delivery and controlled drug release, which dramatically reduce side effects, a drawback using conventional medicines. Together with the development of genetic analysis, nanoparticles stand out as unique drug carriers. In this section, we discuss several therapeutic methods such as photodynamic therapy as well as controlled delivery of drugs/gens using various types of nanoparticles. Other types of therapeutic approaches including photothermal therapy and gene delivery could also be achieved using hybrid nanoparticles. 31,32

3.1. Photodynamic Therapy

Reactive oxygen species generated by different types of nanoparticles have been demonstrated for their efficiency in inducing cell apoptosis, which can be employed for cancer treatment. Single-component phosphorescent polymer dots containing an Ir(III) complex have been developed by us. 33 Upon irradiation with 488 nm light, energy transfer between...
the triplet state of the Ir(III) complex and the ground state of an oxygen molecule leads to the generation of singlet oxygen (\(1O_2\)) by means of the triplet–triplet annihilation mechanism (Figure 6). To the best of our knowledge, this is the first example of the use of single-component polymer dots to produce \(1O_2\) that further promotes cancer cell apoptosis under low-power light irradiation.

Another example of photodynamic therapy utilizes the nanochannels of MSNPs for the disassembly of zinc(II) phthalocyanine (ZnPc) dye, leading to increased efficiency in \(1O_2\) generation. In this case, adamantane (Ada) was used to modify the nanochannels of MSNPs to increase the intercalating effect in order to prevent the aggregation of ZnPc inside the pores.\(^3\)\(^4\) In vitro experiments in HeLa cells showed efficient apoptosis through photodynamic therapy. In order to improve the selectivity of the nanoparticles for cancer cells over normal cells, the surface of the MSNPs was modified with an FA-targeting ligand. In further studies, the photosensitizer precursor 5-aminolevulinic acid (5-ALA) was loaded into HMSNP carriers.\(^3\)\(^5\) Once 5-ALA was released inside skin cancer cells, it was converted to protoporphyrin IX (PphIX), a strong photosensitizer for effective photodynamic therapy. The decomposition rate of PphIX inside cancer cells (12–24 h) is 6 times higher than that in healthy cells (2–4 h), thus facilitating the therapeutic efficacy.

3.2. Drug Delivery and Controlled Release

3.2.1. pH-Triggered Release. Since MSNPs offer a great platform for drug loading as well as targeted drug delivery, controlled-release systems based on functionalized MSNPs specifically for the delivery of the anticancer drug doxorubicin (Dox) were developed.\(^3\)\(^6\)\(^3\)\(^7\) MSNPs were functionalized with several components having various functions: (1) a poly(ethylene glycol) (PEG)-coated surface to enhance the nanoparticle stability under physiological conditions; (2) folate ligands for cancer targeting; (3) perdiamino-\(\beta\)-cyclodextrin (\(\beta\)-CD(NH\(_2\))\(_7\)) conjugation to provide positive charges on the nanoparticle surface under acidic conditions in order to facilitate the transfer of the nanoparticles from endosomes to the cytoplasm; and (4) \(\beta\)-CD(NH\(_2\))\(_7\) grafted on the mesopore orifices to control drug loading and release by electrostatic interactions under \(pH\) changes.\(^7\) The study showed that Dox-loaded MSNPs could be efficiently internalized by HeLa cells through receptor-mediated endocytosis and that the loaded drugs could then be released into the cells triggered by acidic endosomal \(pH\).

In order to investigate the effect of the nanoparticle size in cancer therapy, functional MSNPs with different sizes of 48, 72,
and 100 nm were synthesized. Increasing the reaction temperature to 95°C accelerated the nucleation process. Meanwhile, introducing a short chain of PEG-silane (MW = 575–750) on the nanoparticle surface inhibited silica condensation, thus leading to smaller MSNPs. The functionalyzed nanoparticles were loaded with Dox to afford Dox@PEG-MSNPs48-CD-PEG-FA, Dox@PEG-MSNPs72-CD-PEG-FA, and Dox@PEG-MSNPs100-CD-PEG-FA with different diameters. MDA-MB-231 tumor-bearing Balb/c mice injected with different nanoparticles were analyzed at different post-injection durations of 24, 48, and 72 h. The concentrations of Si in different major organs and the tumor were evaluated to determine the targeting effect of various MSNPs (Figure 7). The obtained results revealed that the highest drug accumulation in the tumor was achieved in the case of Dox@PEG-MSNPs48-CD-PEG-FA 48 h postinjection. Remarkably, obvious tumor shrinkage (ca. 80%) was observed after 3 week injection of Dox@PEG-MSNPs48-CD-PEG-FA, demonstrating a high tumor-targeting effect and therapeutic efficacy.

3.2.2. Redox-Triggered Release. Glutathione (GSH) is an important antioxidant in the human body, preventing damage to cellular components caused by ROS. In particular, the GSH concentration in the intracellular environment is about 103 times higher than that in the extracellular matrix. Therefore, GSH can be a potential trigger for drug release within cancer cells. Given the fact that disulfide bonds are susceptible to GSH reductant,38 smart designs to incorporate S–S linkages into nanoparticles can lead to redox-triggered release.

Figure 7. (a–c) Biodistribution of different-sized PEG-MSNPs-CD-PEG-FA after intravenous injection into mice (dose = 20 mg kg⁻¹). Si concentrations were measured by ICP-MS and converted to the Si percentage of injected dose per gram of tissue (% ID/g). (d) MDA-MB-231 solid tumors from different treatment groups excised on the 31st day. Scale bars: 5 mm. Reproduced with permission from ref 37. Copyright 2013 Wiley-VCH Verlag GmbH & Co. KGaA, Weinheim.

Figure 8. Representative TEM images showing (a) a HepG2 cell on tissue-culture polystyrene (control group) and (b–d) HepG2 cells treated with (b) HMSNPs, (c) HMSNs-S-S-Ada/β-CD, and (d) HMSNs-S-S-Ada/β-CD-LA after 12 h. Reproduced with permission from ref 40. Copyright 2014 Elsevier Ltd.
release systems. Taking advantage of this mechanism, we designed and synthesized redox-responsive nanoparticles by utilizing different drug delivery platforms such as MSNPs, HMSNPs, and copolymer micelles. Redox-responsive HMSNPs were constructed by immobilizing removable Ada functional moieties onto silica nanocontainers connected through disulfide bonds. CDs serving as the gates were capped on the nanoparticle surface to control the drug loading and release (HMSNPs-S-S-Ada/β-CD). The Dox loading capacity of the resulting delivery system was up to 20 wt %, which is much higher than that of MSNPs (<10%). FA or lactobionic acid (LA) was grafted onto the HMSNPs for their effectiveness in targeting different cancer cells. Dox-loaded HMSNPs functionalized with FA could effectively inhibit the growth of HeLa subcutaneous tumors, while Dox-loaded and LA-functionalized HMSNPs (HMSNPs-S-S-Ada/β-CD-LA) showed superb therapeutic efficacy toward HepG2 tumors after 21 days of treatment. Thus, this design presents a novel type of redox-responsive drug delivery system with tumor specificity (Figure 8).

3.2.3. Light-Triggered Release. In addition to intra-cellular triggers such as pH and GSH, external triggers are also important options to control the drug delivery process. Among various external stimuli, light is chosen as a trigger on account of its rapid activation and low invasiveness to biological systems. An organic-nanoparticle-based drug delivery system was developed using perylene-3,4,9,10-tetrayltetramethanol (Pe(OH)4) as a photoremovable protecting group. This protecting group was coupled with the anticancer drug chlorambucil (Cbl) to form photocaged Pe(Cbl)4 nanoparticles. The Pe(Cbl)4 nanoparticles showed high drug loading capacities of up to ca. 79 wt %. The green emission of Pe(Cbl)4 nanoparticles at 558 nm was quenched by photolysis upon irradiation with visible light, and at the same time the Pe(Cbl)4 nanoparticles released the Cbl drug. This is a good example of a real-time-monitoring drug release system. Real-time monitoring of some serious diseases (e.g., cancer and heart attack) by monitoring of certain biomarkers is important for early detection of the diseases.

Another useful organic molecule that is well-known for its photoresponsive properties is azobenzene. Its isomerization between the cis and trans conformations upon light irradiation and heating was utilized in the drug loading and release process. Photothermal-responsive α-CD/azobenzene [2]-rotaxane-mechanized MSNPs were achieved. The controlled drug release was based on the back-and-forth movement of the CD ring on the azobenzene dumbbell. Upon heating or visible-light irradiation, curcumin-loaded rotaxane-mechanized MSNPs could effectively release curcumin against heart failure in zebrafish larvae (Figure 9), shedding light on future applications of light-triggered treatment.

4. HYBRID NANOPARTICLES FOR COMBINED THERANOSTICS

The combination of diagnostics and therapeutics provided by hybrid nanoparticles has been highly sought in biomedical science, as it can provide real-time monitoring of the treatment process. For instance, the biocompatible pillarenne-based assembly can be loaded with different fluorescent dyes and drugs in its cavities. The capability to load different types of dyes and anticancer drugs and then to deliver them in vitro has paved the way for utilizing these biocompatible carriers in theranostics. Another example in which GO-wrapped gold nanoparticles (Au@NGO) were used for Raman imaging and hydrophobic drug delivery was demonstrated. The Raman signal was enhanced by the integration between GO and AuNPs to facilitate the imaging process, whereas the π conjugation system of GO was used for Dox loading by π–π stacking interactions. Dox was later released when the hybrid entered an acidic endosome.

Concurrently, genetic engineering presents great promise and opportunities in personalized medicine. A new DNA sequence is transported to the host organism to develop the desired property by replication, which may allow personalized clinical treatment. In order to deliver these therapeutic oligonucleotides to specific targets, a delivery system is needed.
By using the above-mentioned disulfide bond modification on MSNPs, we were able to carry out redox-responsive codelivery of drugs and single-stranded DNA or small interfering RNA (siRNA). The ability to deliver model genes into HeLa cells using the fabricated delivery systems demonstrates their feasibility in controlled gene delivery. On the other hand, pH-triggered release of siRNA was also achieved via HMSNPs-based carriers. The versatility of fluorescent MSNP delivery systems was further proven by the intracellular delivery of the antisense peptide nucleic acid PNA(Ac-2). Bcl-2 protein expression in HeLa cells after PNA(Bcl-2) delivery was semi-quantified by the Western blot assay, providing a foundation for the future development of gene therapy.

5. CONCLUSION AND OUTLOOK

Upon incorporation of different functional groups, ligands, and biomolecules, multifunctional nanoparticles have shown their superb application capability in diagnosis and therapeutics. Several hybrid nanoparticles have already exhibited promising application potential during clinical trials. For instance, functionalized magnetic iron oxide nanoparticles including dextran-coated iron oxide nanoparticles (Sienna+) for the detection of sentinel lymph nodes, amnisilane-coated iron oxide nanoparticles (MFL AS1) for hyperthermia therapy, and siloxane-coated iron oxide nanoparticles (Ferumoxsil) as efficient oral MRI contrast agents have presented clinical utility. These encouraging results are driving the research field forward quickly.

Various designs and synthesis methods have been developed to obtain specific hybrid nanoparticles with targeted and controlled drug delivery towards practical uses. Different aspects of nanoparticles, from investigations of the effects of various types of nanoparticles and their sizes on the therapeutic efficiency in biological environments to studies of specific ligand targeted drug/gene delivery have been carefully performed. As an example, our research has revealed that smaller-sized functionalized MSNPs can lead to better tumor accumulation and that the use of HMSNPs-based carriers can dramatically enhance the drug loading capacity. Despite all of the advantages of nanoparticle-based systems discussed in this Account, there is always room for further improvements in this field. The toxicity of the nanoparticle carriers is always a major concern when they are used in real patients. Reducing the dosage of nanocarriers and enhancing their biodegradability in vivo are the most promising solutions. Taking MSNP-based nanocarriers as an example, one of the possible methods is to dope zinc or iron elements into the silica network, which may facilitate excretion of the nanoparticles from body after drug release. In view of the significant research efforts being dedicated to the field, it could be expected that humanity will greatly benefit from nanomedicine in the near future.

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Notes

The authors declare no competing financial interest.

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Kim Truc Nguyen received her B.Sc. (Honors) degree in Chemistry from Nanyang Technological University (NTU) Singapore in 2011 and obtained her Ph.D. degree there in 2015 under the supervision of Prof. Yanli Zhao.

Yanli Zhao is currently an associate professor at NTU. He received his B.Sc. and Ph.D. degrees from Nankai University. He was a postdoctoral scholar with Professor Sir Fraser Stoddart at UCLA and subsequently at Northwestern University as well as with Professor Jeffery Zink at UCLA. His current research focuses on biocompatible nanoparticles for diagnostics and therapeutics and porous materials for energy storage and catalysis.

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