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Catalytically Active Nanomaterials: A Promising Candidate for Artificial Enzymes

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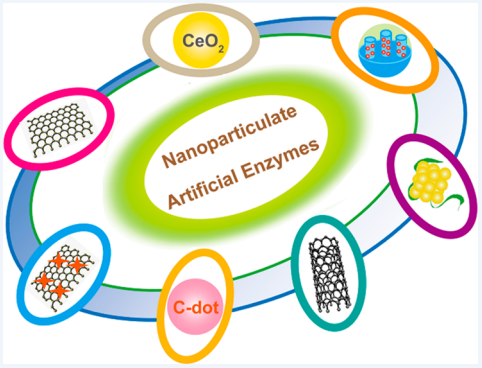
CONSPECTUS: Natural enzymes, exquisite biocatalysts mediating every biological process in living organisms, are able to accelerate the rate of chemical reactions up to 10^{19} times for specific substrates and reactions. However, the practical application of enzymes is often hampered by their intrinsic drawbacks, such as low operational stability, sensitivity of catalytic activity to environmental conditions, and high costs in preparation and purification. Therefore, the discovery and development of artificial enzymes is highly desired. Recently, the merging of nanotechnology with biology has ignited extensive research efforts for designing functional nanomaterials that exhibit various properties intrinsic to enzymes. As a promising candidate for artificial enzymes, catalytically active nanomaterials (nanozymes) show several advantages over natural enzymes, such as controlled synthesis in low cost, tunability in catalytic activities, as well as high stability against stringent conditions.



In this Account, we focus on our recent progress in exploring and constructing such nanoparticulate artificial enzymes, including graphene oxide, graphene-hemin nanocomposites, carbon nanotubes, carbon nanodots, mesoporous silica-encapsulated gold nanoparticles, gold nanoclusters, and nanoceria. According to their structural characteristics, these enzyme mimics are categorized into three classes: carbon-, metal-, and metal-oxide-based nanomaterials. We aim to highlight the important role of catalytic nanomaterials in the fields of biomimetics. First, we provide a practical introduction to the identification of these nanozymes, the source of the enzyme-like activities, and the enhancement of activities via rational design and engineering. Then we briefly describe new or enhanced applications of certain nanozymes in biomedical diagnosis, environmental monitoring, and therapeutics. For instance, we have successfully used these biomimetic catalysts as colorimetric probes for the detection of cancer cells, nucleic acids, proteins, metal ions, and other small molecules. In addition, we also introduce three exciting advances in the use of efficient modulators on artificial enzyme systems to improve the catalytic performance of existing nanozymes. For example, we report that graphene oxide could serve as a modulator to greatly improve the catalytic activity of lysozyme-stabilized gold nanoclusters at neutral pH, which will have great potential for applications in biological systems. We show that, through the incorporation of modulator into artificial enzymes, we can offer a facile but highly effective way to improve their overall catalytic performance or realize the catalytic reactions that were not possible in the past. We expect that nanozymes with unique properties and functions will attract increasing research interest and lead to new opportunities in various fields of research.

1. INTRODUCTION

Enzymes are extremely efficient at catalyzing a variety of reactions with high substrate specificity, activities, and yields under mild reaction conditions.[1,2] As a result, there is significant interest in utilizing enzymes for applications in biosensor, pharmaceutical processes, food industry, and agrochemical production. However, problems, including their low operational stability (denaturation and digestion), sensitivity of catalytic activity to environmental conditions, difficulties in recovery and recycling, and high costs in preparation and purification, greatly limit their applications.[3,4] To circumvent aforementioned limitations, artificial enzymes have been established as low-cost and highly stable alternatives to natural enzymes.[5−7] So far, design of artificial enzymes has very rapidly emerged as a lively field of research. Recently, the merging of nanotechnology with biology has also ignited intensive research efforts for designing functional nanomaterials that exhibit various properties intrinsic to enzymes.[6,8−25] A variety of nanoscale materials, such as cerium oxide nanoparticles,[8−10] magnetic nanoparticles,[3,11,12] gold nanoparticles (AuNPs),[13−18] V_2O_5,[19,20] PtPd−Fe_3O_4,[21,22] graphene oxide,[23] and carbon nanotubes,[24] have been discovered to possess unique enzyme-mimic catalytic activities. Due to the outstanding catalytic property of thiol monolayer protected nanogold, Scrimin, Pasquato, and co-workers called it "nanozyme" in analogy to the nomenclature of catalytic polymer (synzyme).[25] Here, to highlight enzyme-like activity of the nanomaterial, we adopt the term "nanozyme" to describe

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a man-made nanomaterial that is capable of simulating catalytic function demonstrated by natural enzyme.

In this Account, we will review our years' efforts in exploring, constructing, and improving nanoparticulate artificial enzymes, inspired by their advantages. The scope intends to cover not only the design and development of these nanozymes, but also their promising applications and the search for efficient modulators for promoting catalysis. In addition, the perspectives on main challenges and future opportunities are also discussed.

2. NANOPARTICULATE ARTIFICIAL ENZYMES

For ease of access, the examples treated in this Account are classified according to the structural characteristics of nanozymes: carbon-, metal-, and metal-oxide-based nanomaterials.

2.1. Carbon-Based Nanomaterials

Until now, carbon-based nanomaterials have been discovered to possess peroxidase-like[23,24,26−32] or superoxide dismutase-like[33] activity. Our lab contributed much effort to the peroxidase-like activity of graphene oxide,[23] graphene−hemin nanocomposites,[28,29] carbon nanotubes,[24,30] and carbon nanodots.[31]

As an atomically thick sheet of sp^2-hybridized carbon atoms, graphene-based material has recently emerged as a rapidly rising star.[34,35] Intriguingly, we made the surprising discovery that carboxyl-modified graphene oxide (GO−COOH) can serve as an effective peroxidase mimic, which catalyzes the reaction of peroxidase substrate 3,3,5,5-tetramethylbenzidine (TMB) in the presence of H_2O_2 to produce a blue color reaction.[23] Similar to horseradish peroxidase HRP, the catalytic activity of the GO−COOH was dependent on pH, temperature and H_2O_2 concentration.[23] After that, we investigated the mechanism of catalytic graphene oxide through studying the interactions between GO−COOH, H_2O_2 and TMB. First, the different absorption spectra show that a bathochromic shift occurs for GO−COOH upon the addition of H_2O_2.[23] The shifted absorbance suggests that electron transfer occurs from the top of the valence band of graphene to the lowest unoccupied molecular orbital (LUMO) of H_2O_2.[36,37] Meanwhile, since TMB can be absorbed on the surface of graphene and donates lone-pair electrons in the amino groups to graphene, TMB will confer an increase in electron density and mobility in graphene.[23] Such a charge-transfer n-type doping of graphene increases the Fermi level and thus the electrochemical potential from the LUMO of H_2O_2.[36] This accelerates the electron transfer from graphene to H_2O_2.[23] As a result, nitrogen enrichment in this way provides a higher density of catalytically active centers with low stereo hindrance for binding redox species.[23] On the basis of the intrinsic peroxidase property of GO−COOH, we designed a simple, cheap, and highly sensitive and selective colorimetric method for glucose detection by coupling graphene-based nanozyme with glucose oxidase (GOx) (Figure 1).[23] In our experiment, glucose could be detected as low as 1 μM, and the protocol exhibited excellent selectivity over other interferences.[23] More significantly, our method has shown great potential for analysis of glucose level in diluted blood and commercial fruit juices.[23] Thereafter, Huang and co-workers used this catalytically active nanomaterial for colorimetric detection of cancer biomarker prostate specific antigen.[38]

Apart from the intrinsic catalytic activity, graphene and its derivatives can also be utilized as good supports for heterogeneous catalytic processes due to their large specific

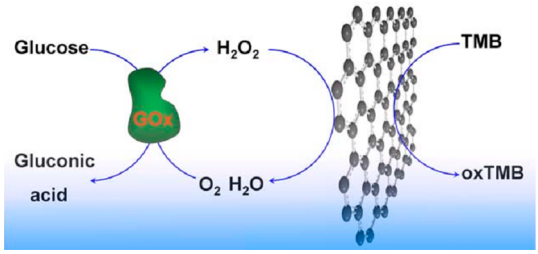

Figure 1. Colorimetric detection of glucose by using GOx and GO−COOH-catalyzed reactions.[23] Copyright 2010, Wiley-VCH.

surface areas.[26,27] Moreover, they possess a rich surface chemistry and have the potential to further promote the catalytic activity and stability of the supported molecular systems.[26,27] For instance, as a well-known natural metalloporphyrin, hemin can be assembled onto the surface of graphene through $\pi-\pi$ stacking.[26,27] As a result, the hemin−graphene nanocomposite (GH) can function as a highly effective catalyst in the oxidation reaction of peroxidase substrate.[26,27] Recently, we reported a colorimetric assay for quantitative and fast detection of cancer cells based on the synergetic peroxidase-like activity of folic acid conjugated graphene−hemin hybrid (GFH) (Figure 2).[28] Figure 2A

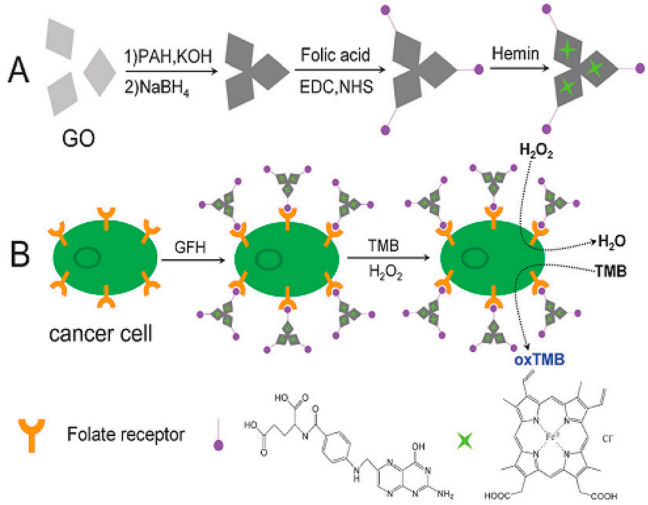

Figure 2. Schematic representation of (A) preparation of GFH and (B) cancer cell detection by using target-directed GFH.[28] Copyright 2011, Royal Society of Chemistry.

illustrates the basic procedure for the fabrication of GFH. Since folate receptors are overexpressed on the surface of different types of cancer cells, GFH could selectively bind to the surface of cancer cells by targeting folate receptors, such as human cervical cancer cells (HeLa) and human breast cancer cells (MCF-7).[28] Owing to the large size and peroxidase-mimicking activity of GFH, selective GFH binding not only could be visualized under bright field microscopy, but also could be quantitatively determined by a colorimetric method (Figure 2B).[28] With the optimized protocol, as few as 1000 cells could be detected.[28] Later, we further developed a robust sensing strategy by utilizing the catalytic GH to detect a broad range of targets including metal ions, DNA, and small molecules (Figure 3).[29] This nearly "universal" biosensor approach is based on DNA-directed assembly of GH by targets.[29] In the absence of targets, GH is stable as DNA

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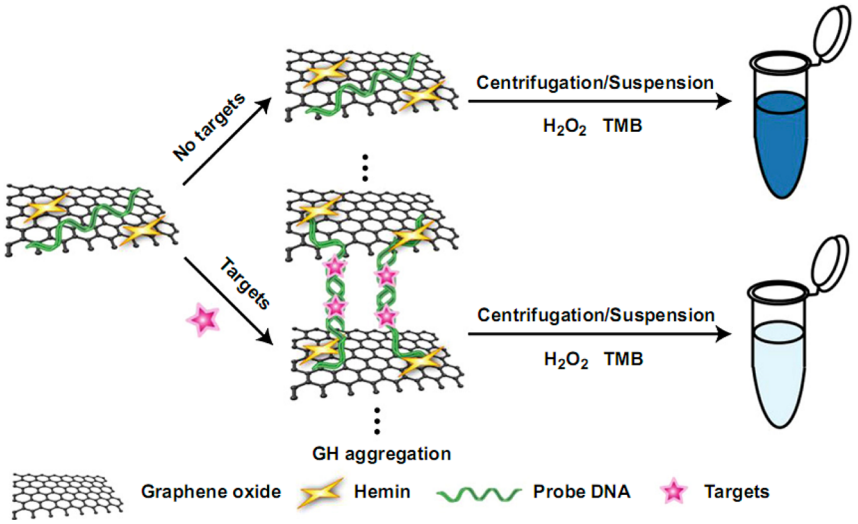

Figure 3. Schematic illustration of procedures for targets detection by using the catalytic GH and targets-induced assembly.[29] Copyright 2013, Elsevier.

hybridization between ssDNA probes does not happen. When a particular target is present, GH aggregate resulting from DNA hybridization will occur. Consequently, the colorimetric signal of the centrifugal supernatant will be significantly lower compared to that without targets.[29] This colorimetric "readout" offers great advantages of simple operation process, low-cost portable instrument, and easy-to-use applications.[29] More importantly, we can detect a broad range of other different targets, such as coralyne and DNA, only by changing label-free and target-specific ssDNA probes.[29] Therefore, such an apparently simple method holds great potential of becoming a routine tool for quantitative detection of a wide spectrum of analytes.

In recent years, carbon nanotubes (CNTs) have also attracted considerable attention.[39,40] They are classified in two main categories: single-wall carbon nanotubes (SWNTs) and multiwall carbon nanotubes (MWNTs), depending on the number of graphene layers.[40] Like COOH−GO, we demonstrated for the first time that SWNTs possess intrinsic peroxidase-like activity.[24] Inspired by the intrinsic peroxidase property of CNTs, we further reported a label-free colorimetric detection system for disease-associated single nucleotide polymorphism (SNPs) in human DNA (Figure 4A).[24] At optimum salt concentration, ssDNA could strongly adsorb on the SWNT surface and increase electrostatic repulsion due to the $\pi-\pi$ stacking interaction, thus resisting salt-induced SWNT aggregation. In contrast, dsDNA could not stably adsorb on SWNT, resulting in large aggregates under the same salt concentration.[24] Afterward, the SWNTs settled on the bottom of the vial after centrifugation, and the precipitate redispersed in phosphate buffer.[24] In the presence of TMB and H_2O_2, the colorimetric signal of the obtained SWNTs with dsDNA was remarkably higher compared to that without target DNA.[24] In addition, the target response proved to be sensitive, and our colorimetric detection approach could be used to distinguish SNPs in human DNA.[24] Besides the direct application of catalytic CNTs, we further developed a sensing and selective strategy for copper detection by click chemistry and combination of magnetic silica nanoparticles with CNTs (Figure 4B).[30]

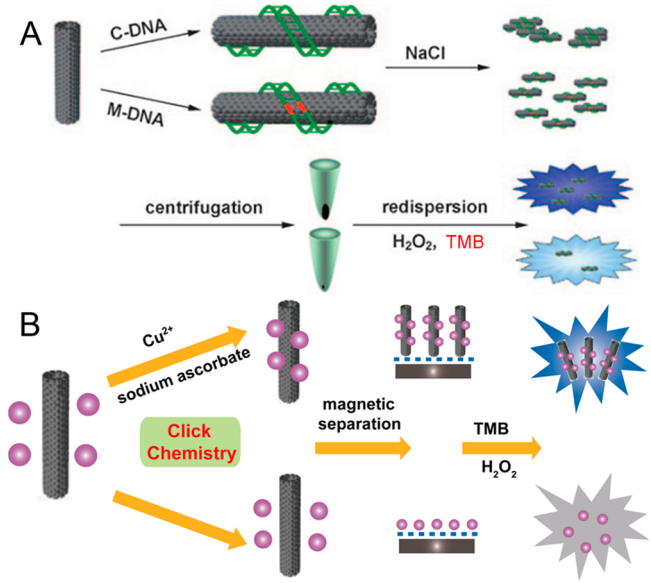

Figure 4. (A) Protocol for SNP detection of complementary (C-DNA) and mismatched (M-DNA) duplex DNA.[24] Copyright 2010, Wiley-VCH. (B) Protocol for sensing Cu^{2+} using click chemistry and peroxidase-like catalytic color reaction.[30] Copyright 2010, Royal Society of Chemistry.

In addition, Huang et al. and our groups also found that photoluminescent carbon nanodots possess high peroxidase-like activity.[31,32] Using this enzyme-mimicking activity, we further developed a simple, cheap, and colorimetric assay for glucose in real samples.[31]

2.2. Metal-Based Nanomaterials

In addition to carbon-based nanomaterials, metal nanomaterials have also been explored to mimic the functions of natural enzymes.[13−18,41] Our interests were mainly put into GOx- or/and peroxidase-mimicking activities of gold-based nanomaterials (i.e., mesoporous silica-encapsulated gold nanoparticles[42] and gold nanoclusters[43]).

Gold colloids have fascinated scientists for over a century and now have been heavily utilized in various applications.[44,45] Historically, gold has been regarded as being chemically inert,

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but in recent years AuNPs with different surface modifications have been found to exhibit GOx- or peroxidase-like activity.[14−16,46] After that, Fan et al. have constructed a new sensor platform for sensitive detection of DNA and microRNA by utilizing this GOx-like activity.[17] However, the potential of AuNPs as enzyme mimics is limited by their relatively low catalytic activities and stability.[17] In addition, single-component AuNPs cannot simultaneously possess dual enzyme-like functionalities, as unsupported AuNPs with different surface properties only keep one enzyme-like functionality active while the other catalytic activity can be completely blocked.[15,46]

Recently, the emergence and recent advance of nanotechnology opens new opportunities for the development of nanoparticles with stable and high catalytic activity.[47,48] With this in mind, we set out to investigate the fabrication of AuNPs encapsulated in expanded mesoporous silica support (EMSN) (Figure 5A).[42] EMSN as the skeleton assists the synthesis of a high density of very small and dispersed AuNPs, hinders the aggregation of neighboring particles, and facilitates the catalytic reaction by providing mesoporous diffusion channels. TEM images reveal that a high density of very small AuNPs immobilized on EMSN is well dispersed (Figure 5B).[42] Since the catalytic properties of AuNPs are strongly dependent on their particle size and stability,[47] the small and stable AuNPs may exhibit much higher catalytic activity. As expected, the resulting EMSN-AuNPs showed high GOx- and peroxidase-like catalytic activities (Figure 5C); whereas citrate-capped AuNPs had very little activities at the same concentration.[42]

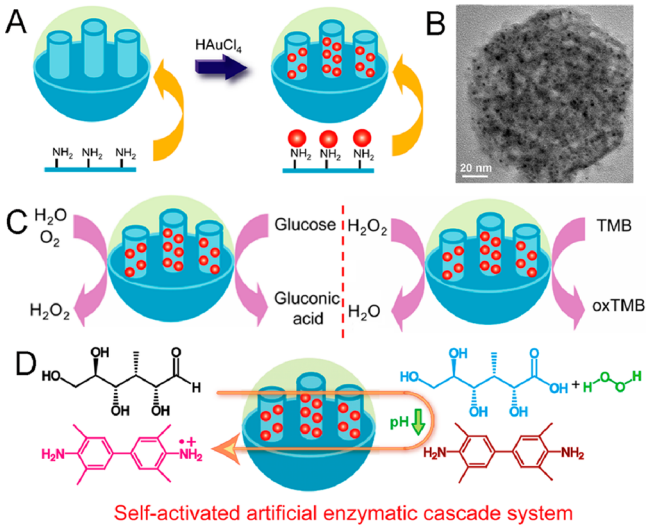

Figure 5. (A) Schematic illustration for the synthesis of EMSN-AuNPs.[42] (B) TEM image of EMSN-AuNPs.[42] (C) Schematic illustration for dual enzyme-like activities of EMSN-AuNPs.[42] (D) Schematic representation of EMSN-AuNPs as intelligent enzyme mimics for realizing artificial catalytic cascade.[42] Copyright 2013, Elsevier.

Although the prepared nanocomposites can be served as dual artificial enzymes with high activities and stability, it is still a big challenge to scale them up for assembling an enzymatic cascade system due to the incompatibility of various reactions operating. In our catalytic system, the optimal pH for their GOx-like activity was 7.4, while this value for peroxidase-mimic activity was about 4.0.[42] Fortunately, as the first reaction product gluconic acid is one of the organic acids, its production in the system could decrease the ambient pH under the low concentration of a buffer solution.[42] Therefore, we offered an effective way to eliminate the incompatibility of different reactions and piece different activities together into a self-organized artificial cascade reaction. Initially, EMSN-AuNPs catalytically oxidized glucose by oxygen to yield gluconic acid and hydrogen peroxide in 0.5 mM phosphate buffer, pH 7.4.[42] Next, gluconic acid produced in the system decreased the ambient pH, which could activate the peroxidase-like activity of EMSN-AuNPs.[42] In the presence of TMB, EMSN-AuNPs could produce a blue color reaction owing to the oxidation of TMB by cumulative product H_2O_2 (Figure 5D).[42] This is the first example using nanomaterials alone as dual artificial enzymes for mimetic cascade catalysis.[42] Therefore, the proof-of-principle results take an important step forward in developing enzyme mimics for realizing more complex functions.

Recently, noble-metal nanoclusters have also become a burgeoning area of scientific interest.[49] Intriguingly, Wang et al. discovered that bovine serum albumin-stabilized gold clusters (BSA-AuNCs) possess intrinsic peroxidase-like activity.[50] Inspired by these initial results, our group developed an easy prepared fluorometric and colorimetric dual channel probe for dopamine by using BSA-AuNCs.[43] The as-prepared BSA-AuNCs exhibited strong fluorescence and high peroxidase-like activity.[43] In the presence of dopamine, the fluorescence intensity of the AuNCs decreased significantly through a photoinduced electron transfer process (Figure 6A).[43] Meanwhile, as the catalytic activity of AuNCs is extremely sensitive to surface properties, their enzyme-like activity could be efficiently restrained after their interaction with dopamine (Figure 6B).[43] Both methods exhibited high sensitivity and excellent selectivity toward dopamine over other interfering substances.[43] More importantly, we demonstrated the application of the present method in hydrochloride injection sample, human serum sample and PC12 cells.[43]

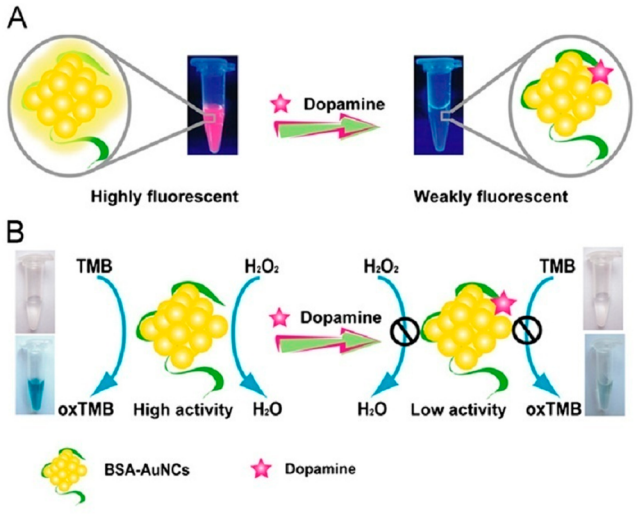

Figure 6. (A) Schematic representation of the fluorescence response of the BSA-AuNCs to dopamine.[43] (B) Schematic illustration of peroxidase-mimicking catalytic color reaction for sensitive sensing of dopamine.[43] Copyright 2013, Elsevier.

2.3. Metal Oxide-Based Nanomaterials

Like carbon- and metal-based nanomaterials, metal-oxides-based nanomaterials have also emerged as efficient enzyme

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mimics.[3,8−12,19,20] Among them, cerium oxide nanoparticles[8−10] and magnetic nanoparticles[3,11,12] are two most widely used metal oxide catalysts. But this introduction will focus on our interests in making use of CeO_2 nanoparticle based ones.[51]

CeO_2 nanoparticles, which exist in a mixed valence state (Ce^{3+}, Ce^{4+}), possess many unique properties that have proven to be of high utility in biomedical and catalytic applications.[8,52−57] Recently, they have been reported to possess multienzyme, such as SOD,[10,54] catalase,[9] oxidase,[8,56] and phosphatase,[57] mimetic properties. For instance, Perez's group reported that nanoceria has an intrinsic oxidase-like activity at acidic pH values, as it can quickly oxidize a series of colorimertic dyes without any oxidizing agent.[8] Following these early works, we found that the pH-tunable oxidation ability of nanoceria to ferrocenecarboxylic acid (Fc-COOH).[51] After incubation nano-CeO_2 with Fc-COOH solution at pH 7.4, there was no significant change in absorption or color.[51] While after incubation at pH 4.5, the mixture solution displayed a yellow to green color change with a new absorption band appeared at 632 nm, which suggested the ferrocenyl moieties could be oxidized to positively charged ferrocenium ions by nano-CeO_2 under acidic pH (Figure 7A).[51] More interestingly,

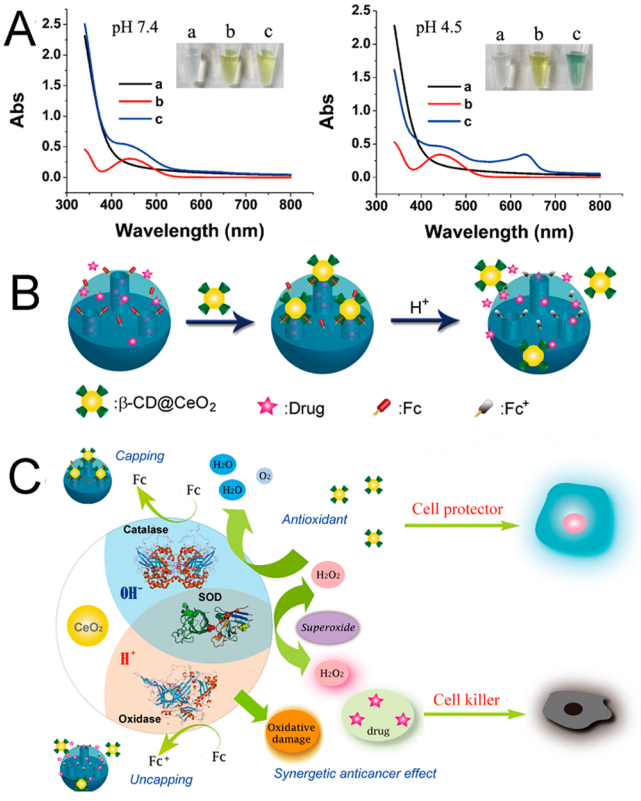

Figure 7. (A) UV−vis spectra and photograph of (a) nanoceria alone, (b) only Fc-COOH, and (c) their mixture at pH 7.4 and pH 4.5.[51] (B) Schematic illustration for pH-triggered release of the anticancer drug from β-CD@CeO_2 capped Fc-MSN.[51] (C) Enzymatic activities of CeO_2 at different pH values.[51] Copyright 2013, Wiley-VCH.

ferrocene residue could tightly bind to β-cyclodextrin (β-CD) via host−guest interactions, whereas the oxidized, positively charged ferrocenium ion did not.[51]

Inspired by these unique features, we demonstrated a pH stimuli-responsive vehicle for intracellular drug delivery by using the β-CD-modified CeO_2 as the capping agent (Figure 7B).[51] To create this nanogated ensemble, ferrocene groups were first introduced onto the outlet of mesoporous silica.[51] Therefore, β-CD-functionalized CeO_2 nanoparticles could cap onto ferrocene-modified mesoporous silica through host−guest interactions.[51] After internalization into A549 cells by a lysosomal pathway, the ferrocenyl moieties were oxidized to ferrocenium ions by CeO_2 lids, which could trigger uncapping of the CeO_2 and cause drugs release.[51] Moreover, as cancer cells have a more acidic cytosolic pH than normal ones, CeO_2 with oxidase-like activity could oxidize some intracellular and extracellular components to induce cancer cell apoptosis (Figure 7C).[51] This proof of concept provides a novel route for using switchable enzymatic activity of CeO_2 as capping agents in the field of versatile controlled delivery nanodevices.

3. EXPLORING EFFICIENT MODULATORS

To keep up with the overall performance of natural enzymes, one major activity is searching more efficient nanozymes, as reflected by numerous recent publications.[8−25] An alternative strategy is exploring modulators to further enhance the performance of existing artificial enzymes. Below, we introduce three exciting push in the use of efficient modulators on artificial catalytic systems.[58−60]

3.1. Ionic Liquid

In spite of the fascinating features of present peroxidase mimics, one of the main shortcomings that these enzyme mimics (including EMSN-AuNPs) suffer is that their catalytic performance at high temperature is far below expectations.[3,23,58] As they usually exhibit superior thermal stability to natural enzyme,[3,23,58] we rule out the possibility of this phenomenon caused by nanozymes themselves, and point out that the thermally induced instability of enzymatic product $ABTS^{\bullet+}$ inhibit their catalytic activity at high temperature (Figure 8A).[58] Inspired by the unique properties of the ionic liquid, we reasoned that it could serve as a stabilizing agent for improving thermal stability of $ABTS^{\bullet+}$, and subsequently enabling high-temperature reaction that were not working efficiently in buffer solution. Indeed, in the presence of ionic liquid, their catalytic activity and product stability at high temperature were much

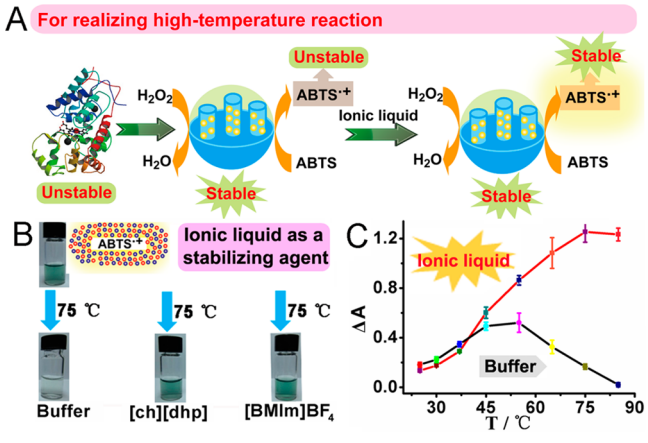

Figure 8. (A) Schematic illustration for the realization of high-temperature catalysis by using thermally stable EMSN-AuNPs and ionic liquid.[58] (B) Thermal stability of enzymatic product in different fluids.[58] (C) Catalytic activities of EMSN-AuNPs as a function of incubation temperature in different fluids.[58] Panels (A)−(C) reprinted with permission from ref 58. Copyright 2013 American Chemical Society.

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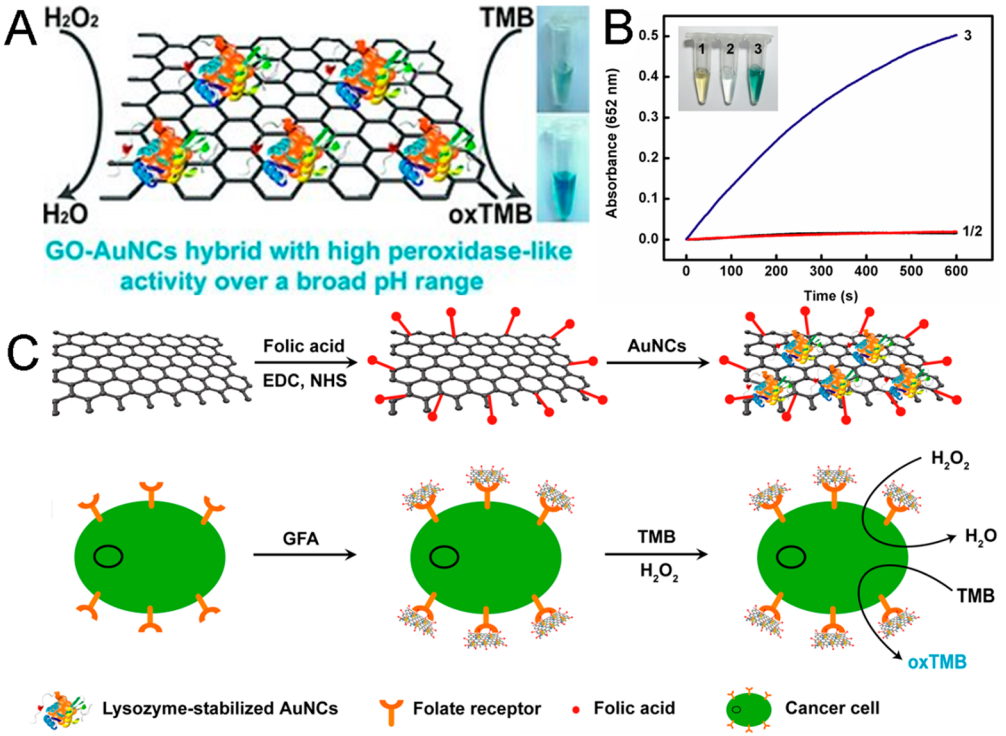

Figure 9. (A) Schematic illustration of the high catalytic activity of synergistic GO-AuNCs hybrid in a fairly broad range of operating pH.[59] (B) Time-dependent absorbance changes at 652 nm in the presence of (1) GO, (2) AuNCs, and (3) the GO-AuNCs hybrid at pH 7.0.[59] (C) Schematic representation of preparation of GFA and cancer cell detection by using target-directed GFA.[59] Copyright 2013, Wiley-VCH.

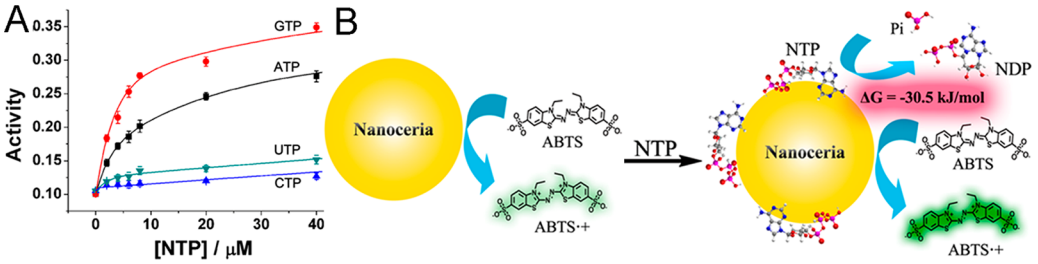

Figure 10. (A) Increased catalysis of nucleoside triphosphates (NTPs) to oxidase-like activity of nanoceria.[60] (B) Schematic illustration of the coupling of the oxidative reaction with the NTP hydrolysis reaction.[60] Copyright 2013, Wiley-VCH.

greater than that obtained in the absence of ionic liquid (Figure 8B, C).[58] Such a positive effect is possibly connected with their weak interactions with product, a large number of cations and anions, and their greater solvation power.[58] Our findings pave the way to applying ionic liquid as a positive modulator in nanozyme-based catalytic reactions.

3.2. Graphene Oxide

Compared with other nanomaterial-based peroxidase mimics, AuNCs as nanozymes are more prominent for bioanalysis due to their small size, excellent stability, and biocompatibility.[50] Despite these favorable properties, AuNCs usually suffer one main shortcoming: its optimum reaction occurs in acidic solution, which seriously restrict its applications in biological systems where a near neutral pH is required. To overcome this drawback, we demonstrated that GO could serve as the enzyme modulator to regulate the peroxidase-like activity of lysozyme-stabilized AuNCs (Figure 9A).[59] The most exciting feature of the synergistic GO-AuNCs hybrid is that it exhibits high catalytic activity over a broad pH range, even at neutral pH.[59] As shown in Figure 9B, hybrid catalyst exhibited excellent catalytic activity at neutral pH, whereas, both GO and AuNCs showed almost no activity.[59] Significant activity enhancement indicated that GO played an important role in modulating the catalytic activity of the AuNCs. Initially, TMB could be absorbed onto GO efficiently as GO possessed high surface-to-volume ratios as well as high affinity for hydrophobic molecules.[23] After that, the active site of AuNCs and substrate TMB were confined in the same nanoscale region, which could greatly enhance the catalytic activity of AuNCs.[59] This mechanism was similar to that of natural enzymes in which the extraordinarily high catalytic efficiency was largely due to the ability to bring substrates into proximity with their active sites.[2] Consequently, the hybrid catalyst showed an excellent peroxidase-like activity over a broad pH range, thus opening this novel catalytic system for a multitude of potential applications in biological systems.[59] For instance, upon conjugation of folic acid to the hybrid, we have utilized the target-functionalized nanohybrid (GFA) as a robust nanoprobe for selective, quantitative, and fast colorimetric detection of cancer cells at physical pH, which would provide a drastic positive effect on diagnosis and prognosis (Figure 9C).[59]

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Table 1. Comparison between Natural Enzymes and Nanozymes[a]

	main advantages	main disadvantages
natural enzymes	(1) high efficiency	(1) low operational stability
	(2) high specificity	(2) high cost
	(3) high selectivity	(3) sensitivity of catalytic activity to environment
	(4) various catalytic types	(4) difficulties in recovery and recycling
nanozymes	(1) high operational stability	(1) low efficiency
	(2) low cost	(2) low specificity and low selectivity
	(3) facile preparation	(3) limited catalytic types
	(4) robustness against stringent conditions	(4) difficulties in designing efficient nanozymes
	(5) *large surface area for further modification*	
	(6) *other specific functions of nanozymes besides catalysis (e.g., magnetic property of magnetic nanoparticles for recycling)*	

[a]Items in italic font are unique for nanozymes.

3.3. Nucleoside Triphosphates

Very recently, we reported that nucleoside triphosphates (NTPs) could be used as coenzymes improve the oxidase-like activity of nanoceria and that activity enhancement is related with the type of NTPs (Figure 10A).[60] Since the nanoceria has both oxidase-like[8] and phosphatase-like[57] activities, such increased catalysis resulted from the coupling of the oxidative reaction with the NTP hydrolysis reactions (Figure 10B).[60] In addition, the difference of the improvement effect reflected the different dephosphorylation catalytic activities of nanoceria to the NTP used.[2] Based on these intriguing results, we also developed series effective and high-throughput colorimetric assays for single-nucleotide polymorphism (SNP) typing.[60]

4. CHALLENGES AND FUTURE OPPORTUNITIES

To better understand the challenges and opportunities of nanozymes, the main advantages and disadvantages of nanozymes and natural enzymes are presented in Table 1. Under comparison, artificial enzymes are less vulnerable to denaturation, low-cost, easy to obtain, and more stable to biodegradation. In addition to the above advantages shared with other enzyme mimics, nanozymes also possess their unique features, including large surface area for further modification and other specific functions besides catalysis (e.g., magnetic property of magnetic nanoparticles).

Despite these advantages, there are four important challenges that remain for efficiently directing the development of nanomaterial-based artificial enzymes (Table 1). (1) Nanozymes usually exhibit the relatively low catalytic activity, in comparison with natural enzymes. In this respect, the development of enzyme mimics to be able to show excellent enzyme-like activity will have great potential for the next generation of mimetic enzyme systems. Alternatively, our recent works have started to address this issue by exploring efficient modulators.[58−60] (2) Nanozymes often have a lower binding affinity and lower specificity for substrate than natural enzymes. In contrast, due to their unique catalytic microenvironments, enzymes exhibit high binding affinity and substrate specificity. In their active site, a pocket is available for substrate recognition and catalysis. For this reason, the exterior surfaces of nanomaterials may be coated with functional groups similar to those exposed by enzymes,[6] thus enhancing their binding affinity and substrate specificity. Meanwhile, it can accelerate the chemical transformations by bringing substrates into proximity with active sites.[2] (3) The types of nanozyme catalytic reactions are limited to only redox type reactions and hydrolytic reactions, whereas enzymes are known to catalyze various types of biochemical reactions. Therefore, further investigations are indeed essential to construct novel nanozymes for catalyzing other types of reactions. (4) Rational design of efficient nanozymes is still remains a big challenge. A solution to this problem may be offered by the rapidly growing field of nanotechnology.[47,48]

Besides resolving those problems mentioned above, future work in nanozymes technology is likely to continue to focus on the exploitation of their potential applications and piece these synthetic nanocomponents together into organized functional systems.

5. CONCLUSIONS

Over the past few decades, natural enzymes have been a constant source of inspiration for chemists in their efforts to create synthetic structures that mimic their functions and promote catalysis. In this Account, we summarized our recent development in the field of nanozymes. The studies featured herein attempt to understand and apply nanomaterials as enzyme mimics and, more significantly, shed light on the improvements of these catalytic nanomaterials and their promising applications in biomedical diagnosis, environmental monitoring, and therapeutics.

Despite the fact that there are still many unresolved issues and challenges, the unique properties and functions of these enzyme mimics and the promising results exhibit that this field will continue to thrive and mature in the years to come.

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Notes

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Xiaogang Qu received his Ph.D. from the Chinese Academy of Sciences (CAS) in 1995 with CAS President's Award. He moved to the United States afterward and worked with Professor J. B. Chaires at the Mississippi Medical Center and Nobel Laureate Professor Ahmed. H. Zewail at the California Institute of Technology. Since late 2002, he is a professor at Changchun Institute of Applied Chemistry, CAS. From 12/2006 to 05/2007, he visited the group of Nobel Laureate Professor Alan J. Heeger at UCSB. His current research is focused on ligand-nucleic acids or related protein interactions, and biofunctional materials for advanced medical technology.

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