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ABSTRACT
In this paper, we reported a ZnO quantum dot (QD) based nano drug delivery system by surface modification of hydrophilic copolymer poly(methacrylate-co-N-isopropylacrylamide-co-polyethylene glycol methyl acrylate) (ZnO@PMNE) that was synthesized by the one step copolymerization method. The ZnO@PMNE nanoparticles (ZnO@PMNE NPs) have excellent stability and photoluminescence performance in water. Moreover, it is temperature responsive and biodegradable in an acid microenvironment. ZnO@PMNE NPs loaded with doxorubicin (DOX) will degrade to zinc ions (Zn$^{2+}$) and DOX at an acidic tumor microenvironment. Moreover, the cytotoxicity of cancer cells was significantly increased because the ZnO QDs exhibited cytotoxicity postdissolution compared to normal cells, which will achieve a synergistic antitumor effect to improve the therapeutic index.

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I. INTRODUCTION
Cancer threatens people’s health worldwide, and radiation therapy and chemotherapy are the most widely used cancer treatments, which, however, have severe side effects with great sequelae. In the past few decades, drug delivery systems based on nanotechnology have been widely researched to improve the cancer treatment route and avoid the damage of healthy cells. The nano drug delivery systems (NDDSs) are potential candidates in a variety of smart nanoscale platforms that carry drugs and labeling agents, which is because their size, shape, and surface combination can be precisely controlled. These NPs include graphene, metal oxide NPs, carbon NPs, carbon nanotubes, mesoporous silica NPs, rare earth NPs, and polymer NPs.

Quantum dots (QDs) have drawn huge attention in bioimaging applications because of their high quantum yield, photostability, and size tunability. In comparison with traditional nanoparticle drug delivery systems, the advanced ZnO NP drug delivery platform has many advantages. ZnO NPs are easy to prepare, are of low cost and less toxic, and display a response to acid. ZnO NPs have the ability of rapidly dissolving Zn$^{2+}$ and preferentially killing cancer cells at pH < 5.5. On the other hand, the ZnO NPs can load and release doxorubicin (DOX), which will lead to synergistic killing of tumor cells. However, there are two drawbacks, including low quantum yield and poor stability. In order to compensate for this shortcoming, the ZnO QDs were coated with the polymer, which could enhance stability and quantum yield. According to previous reports, ZnO QDs can be modified with polymethyl methacrylate (PMMA), polystyrene (PS), oleic acid, etc. Muhammad conjugated folic acid onto the surface of ZnO NPs and loaded DOX for cancer treatment. Cai reported that dicarboxyl-terminated poly(ethylene glycol) (PEG) and hyaluronic acid (HA) were introduced to modify ZnO NPs. The ZnO QDs of the surface were capped with poly[2-(dimethylamino) ethyl methacrylate] (PDMAEMA) that had been used for DNA delivery.

Poly(N-isopropylacrylamide) (PNIPAM) has been widely used in the biological domain. The molecular chains of PNIPAM have both hydrophilic acylamine and hydrophobic isopropyl,
making PNIPAM exhibit temperature-sensitive volume phase transition characteristics. In addition, poly(ethylene glycol) methyl ether methacrylate (PEGMA) is another excellent material in biomedical applications due to its good biocompatibility. PEGMA is a hydrophilic monomer, which was applied to increase the lower critical solution temperature (LCST) and decrease the protein adsorption of PNIPAM materials. The block copolymer (NIPAM-co-PEGMA) can improve the ZnO QDs of aqueous solubility, stability, and tumor targeting. The ZnO NDDS can easily reach the lesion by increasing the LCST and lowering the protein adsorption.

In this study, we developed a thermal and pH responsive NDDS, as shown in Fig. 1, which was based on the ZnO QDs functionalized with P(MAA-co-NIPAM-co-PEGMA) (PMNE). The ZnO@PMNE NPs have excellent water stability and fluorescence performance. DOX was selected as a model drug for this study and was loaded to the ZnO@PMNE NDDS by covalent interactions, which leads to the formation of the Zn$^{2+}$-DOX chelate complex. In addition, the performance of the ZnO@PMNE NDDS was investigated.

II. MATERIALS AND METHODS

A. Instruments

Fourier transform infrared (FT-IR) analysis was carried out using KBr disks in the region of 4000–500 cm$^{-1}$ using a Fourier transform infrared spectrophotometer (Shimadzu IR prestige-21, Japan). The photoluminescence (PL) spectra were recorded on a Shimadzu RF-5301 PC spectrofluorophotometer. Ultraviolet–visible (UV-Vis) absorption spectra were obtained on a Lambda 750 spectrophotometer (PerkinElmer, USA). The particle sizes of NPs were determined using a Britain Malvern PSA (NANO2590) submicron particle size analyzer with angle detection at 90°. The morphology and microstructure of the QDs complex were examined by high-resolution transmission electron microscopy (HRTEM) on a Philips Tecnai G2F20 microscope (Philips, The Netherlands) with an accelerating voltage of 200 kV. XRD [Rigaku D/MAX-2400 X-ray diffractometer with Ni-filtered Cu Ka radiation (λ = 1.540 56)] was used to investigate the crystal structure of the nanoparticles.

B. Materials

Methyl methacrylate (MAA) was purchased from Aladdin and distilled under reduced pressure prior to use. Zinc chloride, ethanol, triethylene glycol (TEG), and lithium hydroxide (LiOH⋅H$_2$O) were available from Qingdao Renhe Xing Experimental Technology Co., Ltd. and used without further purification. 2,2-azobisisobutyronitrile (AIBN) was obtained from Qingdao Zhengyge Experimental Technology Co., Ltd. and recrystallized from ethanol. N-isopropylacrylamide (NIPAM) and polyethylene glycol methyl acrylate (PEGMA) (Mn = 475) were obtained from Aladdin and used as received. DOX, in the form of a hydrochloride salt, was obtained from Aladdin. 3-(4,5-Dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide (MTT) and fetal bovine serum (FBS) were purchased from Alfa Aesar (Beijing, China). All the other reagents were purchased from Chinese National Medicine Corporation without further purification.

C. Synthesis of Zn(MAA)$_2$

Zn(MAA)$_2$ was synthesized as described by Zhang et al.$^{21}$ First, 3.2 g sodium hydroxide was dissolved in 20 ml deionized water. Then, 7.88 g methacrylic acid was added and kept stirring to ensure that they react completely. The pH was tuned to 7.0 followed by dropping ZnCl$_2$ solution. The solid precipitate was collected by vacuum filtration, washed several times with distilled water, and dried in a vacuum oven.

D. Preparation of ZnO QDs

A certain amount of Zn(MAA)$_2$ was dissolved in 20 ml deionized water under magnetic stirring at 70°C. After 3 h, it was cooled down to room temperature. Then, LiOH$\cdot$H$_2$O ethanol solution was added to the mixture with stirring for 2 h, and the ZnO quantum dots were obtained.

E. Preparation of ZnO@PMNE nanoparticles$^{22}$

A certain amount of Zn(MAA)$_2$ with was first dissolved in 20 ml TEG with stirring for 3 h, and then, it was cooled down to room temperature. After addition of AIBN (6 mg), NIPAM, and PEGMA, the mixture was stirred at 72°C for 10 min. After that, another 6 mg AIBN and LiOH$\cdot$H$_2$O were added the mixture and agitated for 2 h. The purified ZnO@PMNE hybrid nanoparticles were obtained by dialysis against deionized water for 3 days.
TABLE I. Structural parameters of a different NDDS.

| Sample       | Zn(MAA)/NIPAM/PEGMA (mg/mg/mg) | \(D_{h}\) (nm) | PDI | Zeta-potential (mV) |
|--------------|---------------------------------|----------------|-----|---------------------|
| ZnO QDs      | 100/0/0                          | 6.1            | 1.172 | −10.21             |
| ZnO@PMN      | 100/800/0                        | 14.8           | 0.298 | −6.25               |
| ZnO@PMNE     | 100/400/400                      | 15.2           | 0.314 | −0.26               |

\(D_{h}\) and PDI were measured by DLS.

F. Stability tests of the ZnO-based NPs

The ZnO NDDS was added into the solution of FBS (10%), assessing the resistance protein adsorption capability of ZnO NPs (1 mg ml\(^{-1}\)). The turbidity of ZnO NPs was determined by the UV absorption spectrum at 600 nm at different times using a UV-Vis spectrophotometer.

G. Test of lower critical solution temperature (LCST)

Phase transition behaviors were investigated in aqueous solutions (10 mg/ml) of ZnO@PMN and ZnO@PMNE. The turbidity of ZnO NPs was determined by the UV absorption spectrum at 600 nm at different times using a UV–Vis spectrophotometer. The solutions were kept at least 5 min at each temperature before measurement in order to reach the stable state. The solution was inactivated by heating, and the temperature at which the optical transmittance of solution was up to 50% reduction.

H. Drug loading and release

The ZnO@PMNE NPs (10 mg) were added into the solution of DOX solution (4 ml, 0.5 mg ml\(^{-1}\)) and kept for 12 h under dark conditions. The DOX loading was quantified by UV-Vis absorbance of DOX at 490 nm on the basis of the calibration curve obtained by the absorbance of DOX solutions with different concentrations under the same conditions. In vitro release experiments were performed at different pH values (PBS buffer). In detail, the dialysis bag (1000 Da) with 1 ml ZnO@PMNE-DOX was rinsed in 200 ml PBS buffer or acetate buffer and stirred at 24 h. 1 ml Samples were collected in cycles, and each of them was replaced with the same volume of fresh buffer. Finally, the amount of released DOX of the samples was measured by the UV absorption spectrum.

I. Cytotoxicity analysis

The cytotoxicity of NPs was evaluated by MTT experiments. HepG2 cells in a 96-well plate at 2 \(\times\) 10\(^4\) cells/well were incubated overnight. Then, ZnO QDs, corresponding concentrations of Zn\(^{2+}\) ions (ZnCl\(_2\)), free DOX, and various concentrations of ZnO@PMNE and ZnO@PMNE-DOX were incubated with HepG2 cells for 24 h, respectively. The medium in each well was replaced by 100 \(\mu\)l MTT (5 mg in 1 ml PBS and 9 ml FBS), and they were incubated for another 4 h. Finally, 100 \(\mu\)l DMSO was added after removing all medium, followed by shaking for 15 min. The microplate reader was used to measure the absorbance of MTT at 490 nm.

J. Confocal microscopy

A laser scanning confocal microscope (Zeiss LSM 800) was used to image the live cells. HepG2 cells were seeded onto the glass-bottomed culture dishes (35 mm, MetTek) at 5 \(\times\) 10\(^3\) cells/dish and incubated at 37 \(^\circ\)C in 5% CO\(_2\) overnight. Then, ZnO@PMNE (20 \(\mu\)g ml\(^{-1}\)) and ZnO@PMNE-DOX (DOX 0.5 \(\mu\)g ml\(^{-1}\)) were added to mix with the cells, respectively. After 4 h incubation, the cells were washed with PBS 3 times to remove the ZnO@PMNE or ZnO@PMNE-DOX complexes adsorbed on the outer surface of the cell membrane. The cells were imaged using the LSM microscope under the excitation wavelength of 360 nm.

III. RESULTS AND DISCUSSION

Highly luminescent ZnO@PMNE NPs were prepared by the one-step copolymerization method.\(^23\) The ZnO@PMNE NPs with different NIPAM/PEGMA ratios are summarized in Table I. It is obvious after being modified with polymers, the hydrodynamic diameter \((D_{h})\) of the NPs increased from 6.1 nm to 15 nm, while the polymer dispersity index (PDI) decreased from 1.172 to around 0.314. The zeta potential of ZnO QDs was \(-10.21\) mV and increased to \(-6.25\) mV after the modification of PMN. The zeta potential of ZnO@PMNE was decreased to \(-0.26\) mV, which is due to the PEG shell shielding of the surface charge of the nanoparticles.

As shown in the TEM images of Fig. 2(a), the size of ZnO@PMNE NPs is 4–5 nm, corresponding to the hydrodynamic diameter. The HRTEM image [Fig. 2(b)] shows that the distance between ZnO@PMNE lattices is about 0.25 nm,\(^24\) which indicated...
that the ZnO@PMNE NPs have a good crystalline structure. The XRD patterns of ZnO@PMNE are consistent with the standard pattern of ZnO (JCPDS 36-1451), which indicated the ZnO@PMNE NPs of the crystalline structure according to the literature (Fig. S1).

The surface modification of ZnO NPs was confirmed by FT-IR spectroscopy (Fig. 3). In Fig. 3(a), the absorption at around 470 cm$^{-1}$ was attributed to the Zn–O bond of ZnO QDs. As shown in Fig. 3(b), the characteristic absorption of PNIPAM appears at ν(NH–) vibration (1650 cm$^{-1}$), C–N (1460 cm$^{-1}$), indicating that the PNIPAM was successfully decorated on the surface of ZnO QDs. Figure 3(c) shows new absorption peaks at 1720 cm$^{-1}$, which were corresponding to C=O groups, indicating that PEGMA was also successfully modified on the surface of ZnO QDs and ZnO@PMNE NPs was formed.

Conventional ZnO quantum dots are unstable in water. However, as shown in Fig. S2, the UV-Vis absorption spectra of the ZnO@PMNE NPs in aqueous solution remained basically the same during the two-week storage, proving that the ZnO@PMNE NPs were very stable in the aqueous solution at pH = 7. The aggregation behavior of ZnO NPs was investigated in 10% FBS to assess their protein adsorption capacity, which is shown in Fig. 4(a). The UV absorbance spectra of ZnO@PMNE, ZnO@PMNE-DOX, and free DOX are shown in Fig. 4(b). The DOX was loaded to ZnO@PMNE NPs successfully, which was verified by the UV-absorbance peak of ZnO@PMNE-DOX at 500 nm.

As shown in Fig. 5(a), the fluorescence spectra of ZnO@PMNE NPs in aqueous solution have typical characterization bands at 550 nm. With increasing concentrations of DOX (10–70 μg ml$^{-1}$) added into 1 mg ml$^{-1}$ of ZnO@PMNE aqueous solution at pH = 7.5, the fluorescence spectrum intensities of ZnO@PMNE were decreased. On the contrary, the similar phenomena are shown in Fig. 5(b), the fluorescence spectrum of DOX relevantly decreased with the increasing amount of ZnO@PMNE (0–350 μg ml$^{-1}$) mixed with DOX (40 μg ml$^{-1}$, pH = 7.5). The successful loading of DOX onto the ZnO@PMNE surfaces can be verified by the fluorescence quenching of DOX. The ZnO@PMNE surface has a lot of Zn-ligand coordination bond and transition metal ions (Zn$^{2+}$) to load DOX, which possesses pH sensitivity. As shown in Fig. 5(c), an increasing quantity of ZnO@PMNE (0–350 μg ml$^{-1}$) was mixed with a fixed amount of DOX (40 μg ml$^{-1}$) under mildly acidic condition (pH = 5.5), the fluorescence of DOX is not totally quenched and only slightly decreased, which is due to an increase of Zn$^{2+}$. Owing to the protonation of DOX’s phenolic group, the coordinate bond between Zn$^{2+}$ and DOX was decomposed at pH = 5.5. The color change at pH = 5.5 was verified, which is shown in Fig. 5(d). The aqueous solution of ZnO@PMNE is transparent and colorless.
FIG. 5. (a) Fluorescence spectra of ZnO@PMNE (1 mg ml\(^{-1}\)) with the increase in the DOX concentration from 0, 10, 20, 30, 40, 50, 60 to 70 \(\mu\)g ml\(^{-1}\). The excitation wavelength is 365 nm, and the pH is 7.5. (b) Fluorescence spectra of DOX (50 \(\mu\)g ml\(^{-1}\)) with the increase in ZnO@PMNE concentration from 50, 100, 150, 200, 250, 300 to 350 \(\mu\)g ml\(^{-1}\). The excitation wavelength is 480 nm, and the pH is 7.0. (c) Fluorescence spectra of DOX (50 \(\mu\)g ml\(^{-1}\)) with the increase in ZnO@PMNE concentration from 50, 100, 150, 200, 250, 300 to 350 \(\mu\)g ml\(^{-1}\). The excitation wavelength is 490 nm, and the pH = 5.5. (d) Photographs of aqueous solutions of (d-1) ZnO@PMNE, (d-2) free DOX, (d-3) ZnO@PMNE-DOX, and (d-4) ZnO@PMNE-DOX at pH = 5.5. [Fig. 5(d–1)]. When it added to DOX aqueous solution, the color of solution is orange [Fig. 5(d–2)] and will change to pink with the addition of DOX [Fig. 5(d–3)]. The color of ZnO@PMNE-DOX solution restored from pink to orange [Fig. 5(d–4)] with the pH value reduced below 5.5, which suggested that there was coordinate bond cleavage between the drug and metal ion and the DOX molecules would release from the surface of ZnO QDs. As a result, acidic environment-triggered drug release can achieve the DOX fluorescence quenching and recovery.\(^{27}\)

[27] We also proved the temperature responsiveness of the ZnO@PMNE NPs. As shown in Fig. 6(a), the average hydrodynamic diameter of ZnO@PMNE NPs was about 15.2 nm at 20 °C. With the solution temperature increased to 40 °C, the particle size decreased to 7.6 nm [Fig. 6(b)]. When the temperature rises, the hydrogen bonding force between PNIPAM copolymer chains and water molecules becomes weaken. The PNIPAM copolymer molecular chain shrinks, and the solubility becomes worse in water solution. When the temperature decreases, the hydrogen bonding force...
The phase transition behavior of ZnO@PMN and ZnO@PMNE in aqueous solutions is shown in Fig. 7(a). The lower critical solution temperature (LCST) was determined from the temperature at which the transmittance reduced by 50%. The LCST of ZnO@PMNE was about 37.8 °C, higher than ZnO@PMN NPs, which was about 33.4 °C. This is due to the presence of the hydrophilic chain from PEGMA. As is shown in Fig. 7(b), the optical photographs of ZnO@PMNE NPs in aqueous solution have significant changes from transparent to turbid with temperatures rising from room temperature to 40 °C, which is attributed to the poor hydrophilicity of PNIPAM above LCST. Thus, when the ZnO@PMNE NDDS arrived at the tumor tissue, the ZnO@PMNE NDDS will shrink and aggregate at the condition of tumor microenvironmental temperature. This could be achieved due to transition temperature
(LCST = 37.8 °C) that was higher than physiologic body temperature (Tb = 37 °C).

The thermosensitive polymer chain on the surface of ZnO also affects the photoluminescence (PL) properties. Figure 7(c) shows the PL spectra of ZnO QDs, ZnO@PMN and ZnO@PMNM, at different temperatures. The ZnO@PMN and ZnO@PMNE fluorescence intensity gradually decreased when the temperature increased from 20 °C to 40 °C. The reduction rate of the ZnO@PMN PL intensity is the fastest when temperatures rise from 30 °C to 35 °C. This is consistent with the LCST of ZnO@PMN (33.4 °C). However, the reduce rate of the ZnO@PMNE PL intensity is the fastest when temperatures rise from 35 °C to 40 °C. This is consistent with the LCST of ZnO@PMNE (37.8 °C). Furthermore, the fluorescence intensity of ZnO@PMNE shows a reversible change from 20 °C to 40 °C [Fig. 7(d)]. After several repeated cycles, the photoluminescence does not show any obvious change. The temperature-dependent fluorescence change is mainly due to two factors. One is ascribed to the polymer modification leading to the aggregation of ZnO QDs. When the temperature rises, the hydrogen bonding force between the polymer (PMN or PMNE) chains and water molecules becomes weaken, while the hydrogen bonding force between the polymer chains themselves becomes stronger. It may cause the aggregation of ZnO QDs and fluorescence diminished. Another is ascribed to the variation of the strain at the surface of ZnO QDs. When the temperature rises, the PNIPAM chain shrinks and produces stress. Thus, the elastic tension will propagate to the surface of the ZnO QDs, producing surface quenching states.

In vitro DOX loading and release experiments were performed to study the potential application of ZnO@PMNE NPs. The drug loading content of DOX to ZnO@PMNE is calculated to be 21.2%. The typically high loading efficiency indicated the potential prospects of the ZnO@PMNE system as NDDS. In Fig. 8, the in vitro drug release experiments at different pH values were carried out over 30 h. At pH = 7.5 (simulating the physiological normal cells condition), only 11.4% DOX was released. When the pH was dropped to 6.5, an obvious increase in release speed was observed due to ZnO QDs beginning to decompose. When the pH was tuned to pH = 5.5 (simulating the physiological tumor cell condition), the decomposition speed of ZnO was improved, resulting in largely increased release speed with ~83% DOX released from the NDDS.

DOX was loaded on the surfaces of ZnO NPs via complexation with Zn2+ ions. Complexation is a pH dependent process, thus, the dissociation of metal ion–ligand coordination bonds and the release of DOX in an acidic microenvironment. As a result, ZnO@PMNE NPs would realize a fast release of drugs inside tumor cells, which is in favor of killing tumor cells. Above all, it is concluded that ZnO@PMNE can serve as a promising candidate for the pH responsive NDDS.

Confocal laser scanning microscope (CLSM) was carried out to verify the intracellular release of DOX from ZnO@PMNE-DOX (Fig. 8). HepG2 cells were incubated with 10 μg ml–1 ZnO@PMNE and ZnO@PMNE-DOX for 3 h. As shown in Fig. 9(a), yellow fluorescence was clear, which indicated the successful penetration of ZnO@PMNE into the tumor cells via endocytosis. Although it was verified that ZnO QDs would decompose under mildly acidic conditions, they still had a certain time to manifest bioimaging properties before decomposing. In Fig. 9(b), red fluorescence in HepG2 cells was noticed, demonstrating the release of DOX molecules from the ZnO@PMNE-DOX into cytosols ascribed to the disintegration of ZnO QDs in intracellular acidic environments of endosomes and lysosomes (pH = 6.5–4.5).

The concentration-dependent cytotoxicity was evaluated by the MTT experiment with HepG2 cells (Fig. 10). As shown in Fig. 10(a), ZnO QDs and zinc ions (ZnCl2) performed a distinct antitumor effect when the zinc ion dosage surpassed 20 μg ml–1. Meanwhile,
more than 90% of the cell could survive with the concentration of zinc ions lower than 5 μg ml⁻¹, which was very important considering that enriched zinc ions were not cytotoxic in healthy cells but fatal for acidic cancer cells. The cytotoxicity of the ZnO@PMN and ZnO@PMNE was also tested, and the results showed that the HepG2 cell viability was more than 90% at a concentration of 40 μg ml⁻¹ of ZnO@PMNE. This is because that the PEG polymer encapsulates and shields the ZnO QDs, which could reduce the cytotoxicity. Similarly, the cytotoxicity of ZnO@PMNE-DOX and free DOX was also studied with HepG2 cell lines, which was shown in Fig. 10(b). According to the literature, Zn²⁺ ions involve in peroxidation of lipid and damage of DNA, resulting in its cytotoxicity. This figure demonstrated that the cell viability of ZnO@PMNE-DOX decreased to 35.7% at 5 μg ml⁻¹, which is more effective than free DOX. We think that this is due to both the loaded DOX and the dissolution of ZnO QDs, exhibiting a synergistic antitumor effect. However, the cytotoxicity of DOX is higher than that of zinc ions at lower concentrations, so DOX plays the major role and Zn²⁺ secondary.
Calcine-AM is a nonfluorescent, cell-permeable dye, widely used to determine the cell viability of cells (green). PI, a nucleic acid binding dye producing red fluorescence, cannot penetrate the membrane of viable cells, but it can readily enter the apoptotic/dead cells due to the loss of membrane integrity.2,25 Therefore, AM-PI staining is able to distinguish viable cells (green) and dead cells (red). As shown in Fig. 11(a), the number of red cells increased when treated with Zn\textsuperscript{2+} (20 μg ml\textsuperscript{-1}) compared to HepG2 cells that without added Zn\textsuperscript{2+}. The result shows that the zinc ion has certain cytotoxicity. A significant amount of red fluorescent cells is exhibited when treated with DOX (0.5 μg ml\textsuperscript{-1}) or 20 μg ml\textsuperscript{-1} Zn\textsuperscript{2+} + 0.5 μg ml\textsuperscript{-1} DOX. The result shows that Zn\textsuperscript{2+} has an antitumor effect and achieved synergistic therapy with DOX. As shown in Fig. 11(b), a few red spots are observed when treated with biocompatible ZnO@PMNE, the results show that the ZnO@PMNE exhibits excellent compatibility compared to control groups of HepG2 cells. An amount of red fluorescent cell is exhibited obviously when treated with DOX or ZnO@PMNE-DOX at 0.5 μg ml\textsuperscript{-1} DOX concentration. The result is consisted with the MTT assay and proved that cancer cells can be inhibited or killed by the ZnO@PMNE-DOX. In conclusion, the ZnO@PMNE NDDS has been successfully fabricated by the one-step copolymerization method. The fluorescence of ZnO@PMNE was temperature and pH responsive. The ZnO@PMNE-DOX remained stable at physiological pH, then the rapid release of DOX and Zn\textsuperscript{2+} was achieved through the dissociation of ZnO QDs in acidic tumor environments, resulting in synergistic killing of tumor cells due to the incorporation of the antitumor effect of Zn\textsuperscript{2+} and DOX. Above all, we demonstrate that ZnO@PMNE-DOX will be a valuable NDDS to improve the therapeutic index by DOX and Zn\textsuperscript{2+}.

IV. CONCLUSIONS

In conclusion, the ZnO@PMNE NDDS has been successfully fabricated by the one-step copolymerization method. The fluorescence of ZnO@PMNE was temperature and pH responsive. The ZnO@PMNE-DOX remained stable at physiological pH, then the rapid release of DOX and Zn\textsuperscript{2+} was achieved through the dissociation of ZnO QDs in acidic tumor environments, resulting in synergistic killing of tumor cells due to the incorporation of the antitumor effect of Zn\textsuperscript{2+} and DOX. Above all, we demonstrate that ZnO@PMNE-DOX will be a valuable NDDS to improve the therapeutic efficiency of cancer.

SUPPLEMENTARY MATERIAL

See the supplementary material for the crystalline structure and stability of the ZnO@PMNE by XRD and UV-Vis absorption spectra.

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