Theranostic metal–organic framework core–shell composites for magnetic resonance imaging and drug delivery†

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Metal–organic frameworks (MOFs) have shown great potential in designing theranostic probes for cancer diagnosis and therapy due to their unique properties, including versatile structures and composition, tunable particle and pore size, enormous porosity, high surface area, and intrinsic biodegradability. In this study, we demonstrate novel MOF-based theranostic Fe₃O₄@UiO-66 core–shell composites constructed by in situ growth of a UiO-66 MOF shell on a Fe₃O₄ core for simultaneous drug delivery and magnetic resonance (MR) imaging. In the composites, the UiO-66 shell is devoted for encapsulating the drug, whereas the Fe₃O₄ core serves as a MR contrast agent. The Fe₃O₄@UiO-66 core–shell composites show good biocompatibility, high drug loading capacity, sustained drug release, and outstanding MR imaging capability, as well as effective chemotherapeutic efficacy, demonstrating the feasibility of designing theranostic Fe₃O₄@UiO-66 core–shell composites for cancer diagnosis and therapy.

Introduction

Metal–organic frameworks (MOFs) are an emerging class of organic–inorganic hybrid porous materials built from metal ion or cluster nodes and organic linkers, which have already been explored for a variety of applications, including separation, catalysis, and gas storage. In recent years, MOFs have been scaled down to nanometer sizes to form nanoscale MOFs and focused on their preliminary biomedical applications in drug delivery and bioimaging. MOFs have been successfully employed as drug delivery vehicles owing to their unique properties suitable for drug loading and release. Different from most of the existing pure organic and inorganic carrier materials, MOFs have exceptionally high surface area and enormous porosity, which are favorable for entrapment of large amounts of drugs, tunable pore size and hydrophilic–hydrophobic cavities to host a variety of drugs with different physico-chemical properties, controllable host–guest interactions and intrinsic biodegradability. Since Ferey’s group first reported the ability of iron-based MOFs as drug carriers to encapsulate ibuprofen molecules at unprecedented levels and deliver the drug continuously, with no burst effect, many researchers have developed other MOFs, such as UMCM-1, ZIF-8, MIL-53, MOF-74, Gd-MOF, UiO-66, Cu-BTC, and chiral MOFs, for drug delivery.

Paramagnetic metal ions-containing MOFs are also promising as contrast agents for magnetic resonance (MR) imaging. Compared with clinical small molecule contrast agents, the framework construction features ensure that MOFs not only possess large amounts of paramagnetic metal centers, but also exhibit enhanced per-metal-based relaxivity. Lin and co-workers first demonstrated the potential of Gd-based MOFs as MR contrast agents. The Gd-based MOFs show excellent longitudinal relaxivity; however, leaching of the free Gd³⁺ ions causes nephrogenic systemic fibrosis, which precludes their clinical applications. Given that Mn²⁺ and Fe³⁺ ions are also known as potent paramagnetic metal ions, with much lower toxicity than Gd³⁺ ions, low toxic Mn-based MOFs and non-toxic iron-carboxylate MOFs, have been developed for T₂/T₁-weighted MR contrast enhancement. The biocompatibility of Mn/Fe-MOFs-based MR contrast agents has been improved, but the moderate relaxivity of Fe/Mn-MOF still limits the imaging sensitivity, which hinders their practical applications. To circumvent the problem and make full use of the extraordinary drug encapsulation capacity of MOFs, the incorporation of nanoparticles with unique magnetic properties into MOFs is indeed an effective strategy.

Iron oxide nanoparticles are prevalent in MR imaging due to their ability to significantly shorten transverse relaxation time...
and excellent biocompatibility. Moreover, various formulations of iron oxide nanoparticles, such as ferumoxsil, ferrixan and ferumoxide, have already been approved by the FDA as T₂-MR contrast agents for clinical use. Recently, Fe₃O₄-based core–shell nanoparticles, such as Fe₃O₄@Au, Fe₃O₄@CuInS₂, Fe₃O₄@PDA, Fe₃O₄@PPy, Fe₃O₄@Cu₂S, have also drawn considerable attention owing to their superior contrast effect in MR imaging. For these reasons, the employment of Fe₃O₄ as magnetic nanoparticles to design multifunctional MOF-based composites with high relaxivity, large drug payload and good biocompatibility is feasible. Recently, two groups have developed multifunctional Fe₃O₄@PAA/AuNCs/ZIF-8 NPs and RTIC-Fe₃O₄@IRMOF-3/FA NPs by coating Fe₃O₄ with two different Zn-based MOFs for imaging and drug delivery, which demonstrate the practicability of using MOF-based composites in biomedicine. However, the poor moisture stability of IRMOF-3 and the toxicity of Zn-based MOFs, originating from the ion channel/DNA damage caused by the competition of Zn²⁺ with Fe²⁺ and Ca²⁺, hinder their practical application in biomedicine. Furthermore, the synthesis procedures usually include multiple steps and the capability of MOF-based composites for imaging and drug delivery, as well as the toxicity should be further systematically investigated. Thus, further development of novel MOF-based theranostic agents via a simple method is necessary and of great interest.

Herein, we report our initial effort to simply incorporate Fe₃O₄ nanoparticles into the MOF UiO-66 to fabricate novel theranostic MOF core–shell composites (Fe₃O₄@UiO-66) for in vitro and in vivo MR imaging and drug delivery (Scheme 1). UiO-66, as one class of zirconium-based MOFs, is constructed with Zr(IV) and 2-amino-1,4-benzenedicarboxylate (NH₂-BDC) ligands and has received great attention in drug delivery due to its excellent chemical and solvent stability. The coordination of a Zr(IV)-cluster and linear ligands forms a cubic rigid 3D porous structure comprising octahedral cavities with a diameter of 1.1 nm and tetrahedral cavities with a diameter of 0.6 nm. The presence of Zr–O clusters, numerous open cavities, metal sites and amphiphilic character make UiO-66 advantageous for capturing and releasing anticancer drugs such as doxorubicin (DOX) based on the strong coordination interactions between the hydroxyl groups in DOX and Zr(IV) centers in UiO-66. In addition, the toxicity of UiO-66 has been demonstrated to be relatively low. Thus, UiO-66, which not only exhibits exceptional chemical and solvent stability, but also possesses good biocompatibility, was selected to fabricate the shell over the Fe₃O₄ nanoparticles for DOX delivery. Fe₃O₄@UiO-66 core–shell composites were synthesized via a facile in situ growth method based on the controllable growth of a UiO-66 shell on a carboxylate-terminated Fe₃O₄ core. The formed Fe₃O₄@UiO-66 composites simultaneously possessed the T₂-MR contrast properties of the Fe₃O₄ cores and the drug delivery ability originating from the MOF shells. As a result, the Fe₃O₄@UiO-66 composites show excellent stability, high drug loading capacity, low cytotoxicity, negligible in vivo toxicity and obvious MR signal attenuation effect. Furthermore, the DOX encapsulated composites show a sustained drug release and exhibit long-lasting and efficient anticancer therapeutic efficacy. All the results demonstrate that the developed Fe₃O₄@UiO-66 core–shell composites possess great potential as a novel theranostic agent for MR imaging and drug delivery in biomedicine.

**Results and discussion**

**Synthesis and characterization of Fe₃O₄ and Fe₃O₄@UiO-66**

Fe₃O₄ nanoparticles were synthesized through a hydrothermal method according to the literature. The as-synthesized Fe₃O₄ nanoparticles were spherical with a diameter of 150 nm (Fig. 1A) and in the cubic phase (JCPDS: 19-0629) (Fig. 1E). The Fe₃O₄@UiO-66 composites were then synthesized by an in situ self-assembly of UiO-66 on the surface of Fe₃O₄ to obtain the core–shell theranostic agent. In a typical process, Fe₃O₄ nanoparticles were directly dispersed into the synthetic precursor of UiO-66 composed of ZrCl₄, NH₂-H₂BDC and DMF, and the reaction was accomplished through a simple hydrothermal procedure. The developed method avoids time-consuming layer-by-layer MOF growth and further modification of core particles, which is much simpler than most of the previous methods of synthesizing MOF-based core–shell composites.

To control the morphology of the core–shell composites, the concentrations of the precursors of UiO-66 were optimized. After mixing with different concentrations of UiO-66 precursor solutions, the Fe₃O₄@UiO-66 composites with different UiO-66 shell thickness (5, 25, 50 nm) were obtained (Fig. S1, ESI†). The Fe₃O₄@UiO-66 composites show uniform core–shell morphology with a 25 nm thickness of the UiO-66 shell when the ratio of Fe₃O₄ and UiO-66 precursor reached 25 mg in 37.5 mg ZrCl₄ (29 mg NH₂-H₂BDC/18 mL DMF) (Fig. 1B). Either the UiO-66 shell was too thick or no shell was formed if the ratio was higher or lower than the optimized ratio. Thus, the Fe₃O₄@UiO-66 core–shell composites synthesized under the optimized condition were used for the following experiments. Furthermore, high angle annular dark field scanning transmission electron microscopy (HAADF-STEM) and elemental mapping
Fig. 1 (A and B) TEM images of the synthesized Fe3O4 (A) and the Fe3O4@UiO-66 core–shell composites (B). (C) HAADF-STEM image and the corresponding elemental mapping of one core–shell Fe3O4@UiO-66 composite. (D) HAADF-STEM image of Fe3O4@UiO-66 composites and the corresponding EDS line scan. (E) XRD patterns of synthesized Fe3O4@UiO-66 composite. (F) XRD patterns of Fe3O4@UiO-66 composites (Fig. S2, ESI†).

The simultaneous existence of the characteristic peaks of Fe3O4 and UiO-66 in the X-ray diffraction (XRD) pattern of Fe3O4@UiO-66 indicated the successful formation of a UiO-66 shell on the surface of Fe3O4 nanoparticles without altering their crystallinity (Fig. 1E). The characteristic peaks of UiO-66 at 1570 cm⁻¹, 1435 cm⁻¹, and 1386 cm⁻¹ and Fe3O4 at 1651 cm⁻¹ and 595 cm⁻¹ in the Fourier transform infrared (FT-IR) spectrum of Fe3O4@UiO-66 indicated the formation of the Fe3O4@UiO-66 composites (Fig. S2, ESI†). The two peaks at 3416 cm⁻¹ and 3373 cm⁻¹ in the spectrum of Fe3O4@UiO-66, belonged to the asymmetric and symmetric stretching absorptions of the primary amine groups in the NH₂–H-BDC ligands, respectively, which further verified the successful growth of the UiO-66 shell on the Fe3O4 core. The contents of Fe and Zr elements in the prepared Fe3O4@UiO-66 were 30.6% and 0.9%, respectively. Thermogravimetric analysis results showed that the Fe3O4@UiO-66 composites decomposed at 340 °C, which is a similar breakdown temperature to that of the synthesized UiO-66 nanocrystals (Fig. S3, ESI†).

The drug loading capacity of Fe3O4@UiO-66 composites depends on the porosity of the UiO-66 shells. N₂ adsorption isotherms were used to characterize the surface area and porosity of Fe3O4@UiO-66 (Fig. 2A). The total pore volume ($V_{\text{total}}$) and BET surface area ($S_{\text{BET}}$) of the synthesized Fe3O4@UiO-66 were calculated to be 0.21 cm³ g⁻¹ and 149.75 m² g⁻¹, respectively, which are large for drug loading yet much lower than those of the pure UiO-66 due to the inner nonporous Fe3O4 core. In addition, the Fe3O4@UiO-66 composites possessed an interparticle mesopore at 3.5 nm (Table S1, ESI†), which makes them efficient for drug delivery.

Superparamagnetism is essential to MR imaging. Thus, the X-ray magnetic hysteresis curves demonstrated the strong superparamagnetic character of the as-synthesized Fe3O4@UiO-66 composites. Furthermore, the Fe3O4@UiO-66 composites could be quickly collected using the magnet (Fig. S5, ESI†).

The strong superparamagnetism of the Fe3O4@UiO-66 composites ensured their ability to act as a T₂ contrast agent for MR imaging. T₂-weighted MR images of Fe3O4@UiO-66 showed an obvious concentration-dependent darkening effect with a high transverse relaxivity ($r₂$) of 255.87 mM⁻¹ s⁻¹ (Fig. 1F), and the $r₂$ values decreased as the thickness of the UiO-66 shell increased due to the reduced ratio of Fe3O4 to UiO-66 in the composite (Table S2 and Fig. S6, ESI†). Furthermore, the $r₂$ value of the prepared Fe3O4@UiO-66 (1396 mg⁻¹ mL s⁻¹) was much higher than that of Fe3O4@PAA/AuNCs/ZIF-8 NPs (53.79 mL mg⁻¹ s⁻¹) and several clinical Fe-based T₂-weighted contrast agents such as ferumoxsil (72 mM⁻¹ s⁻¹), ferumoxtran-10 (63.12 mM⁻¹ s⁻¹), ferumoxtran-10 (63.12 mM⁻¹ s⁻¹), and ferumoxsil (72 mM⁻¹ s⁻¹).
Drug loading/release of Fe₃O₄@UiO-66 composites

The stability of the nanocarrier in a wide pH range from basic to acidic is essential for drug loading/release. Thus, we investigated the stability of the as-synthesized Fe₃O₄@UiO-66 composites at different pH values (4.0, 5.0, 6.0, 7.4, 8.0). No significant changes in the XRD pattern (Fig. 3A) or the morphology (Fig. S7, ESI†) of the Fe₃O₄@UiO-66 composites at these pH values were observed. The results indicated that the composites had super stability.

To evaluate the drug loading capacity of the Fe₃O₄@UiO-66 composites, a commonly used anticancer drug, DOX, was mixed with Fe₃O₄@UiO-66 in PBS at pH 8.0 for 24 h. After removing the excess unloaded DOX, the drug loaded composites Fe₃O₄@UiO-66-DOX were obtained. UV-Vis-NIR spectra of Fe₃O₄@UiO-66 before and after encapsulation with DOX were measured. The appearance of the characteristic peak of DOX at 500 nm in the UV-Vis-NIR spectra of Fe₃O₄@UiO-66-DOX and the increase of the characteristic peak with the added DOX confirmed the successful loading of DOX into the Fe₃O₄@UiO-66 composites (Fig. 3B). Moreover, the presence of characteristic absorption bands (indicated by green dashed rectangle) of DOX in the spectra of Fe₃O₄@UiO-66-DOX also indicated the incorporation of DOX molecules into Fe₃O₄@UiO-66 (Fig. 3C).

The drug loading capacity increased from 2.5 to 66.3 wt% as the amount of DOX increased from 0.15 to 15 mg (Fig. 3D). Furthermore, the drug loading capacity increased with the thickness of the UiO-66 shell (Table S2, ESI†). The DOX loading capacity of Fe₃O₄@UiO-66 could reach 66.3 wt% with an ultra-high loading content of 2.0 mg DOX per mg composites, which is almost the highest DOX payload among the MOFs carriers.25,44-46 The impressive result was meaningful for clinical applications, because the administration of high dosages could be realized using a small amount of composites.

The high drug loading capacity of Fe₃O₄@UiO-66 composites was probably attributed to the large surface area of the UiO-66 shell and the interactions between DOX and UiO-66. A remarkable fluorescence quenching of DOX along with color change from brown to wine for the Fe₃O₄@UiO-66-DOX dispersion confirmed the strong interaction between DOX and Fe₃O₄@UiO-66 (Fig. S5 and S8, ESI†). Potential interactions include π–π stacking between the aromatic anthracycline of DOX and the aromatic pore walls of UiO-66, hydrogen bonding between the oxygen atoms of DOX and the amino groups in UiO-66, van der Waals forces, electrostatic interactions, and coordination bonding.47 Among the above interactions, stable coordination bonding between the deprotonated hydroxyls in DOX and the numerous Zr sites in the UiO-66 framework played the leading role in drug loading, as confirmed by ultraviolet-visible (UV-Vis) spectroscopy and X-ray photoelectron spectroscopy (XPS) (Fig. 3E and S9, ESI†). Red shift of the UV-Vis spectrum of Fe₃O₄@UiO-66-DOX in comparison with that of DOX after DOX loading into Fe₃O₄@UiO-66 indicates the formation of a DOX-Zr complex in the Fe₃O₄@UiO-66 framework, which is also supported by the similar spectral modification of DOX with free Zr(iv) ions (Fig. S9T). XPS results showed that the binding energy of Zr 3d shifted from 182.76 and 185.12 eV for Fe₃O₄@UiO-66 to 182.40 and 184.76 eV for Fe₃O₄@UiO-66-DOX (Fig. 3E). The binding energy of Zr 3d shifting to lower levels after DOX encapsulation could be attributed to electron transfer due to binding of DOX to active Zr sites.48-50 The formation of coordination bonds between DOX and metal centres (Zr, Fe, Zn) has also been described previously.44,51-53 Owing to the strong coordination bonding of DOX–Zr, the Fe₃O₄@UiO-66 showed a remarkable and efficient DOX payload.

The DOX release behavior of the Fe₃O₄@UiO-66-DOX composites was investigated at different pH values (4.0, 5.0, 6.0, and 7.4). The time-dependent accumulated drug release (ADR) curves showed a slow and sustained release pattern without any burst effect (Fig. 3F). The release rate would generate a stable drug concentration and provide sufficient time for the Fe₃O₄@UiO-66-DOX to accumulate at the tumor site. As shown in Fig. 3F, about 36.1% and 21.6% of the DOX were released in 41 days at pH 4.0 and 5.0, respectively, whereas 17.1% and 13.8% of DOX were released at pH 6.0 and 7.4, indicating the sensitivity of Fe₃O₄@UiO-66-DOX to acidic tumor microenvironments. The pH-responsive DOX release was controlled by the

(98.3 mM⁻¹ s⁻¹) and Resovist (150 mM⁻¹ s⁻¹).43 These results show the great potential of Fe₃O₄@UiO-66 in T₂-MR imaging.
drug–matrix interactions under acidic conditions. At acidic pH, the amino group in DOX was easily protonated, giving DOX a positive charge. Moreover, the surface zeta potential of Fe₃O₄@UiO-66 became less negative in acidic conditions (−9.8 ± 1.1 mV at pH 4.0 and −25.7 ± 5.2 mV at pH 7.4). Thus, the electrostatic interactions between Fe₃O₄@UiO-66 and DOX were weakened, which promoted the drug release in acidic conditions. Moreover, the breakage of the coordination bonds between the protonated hydroxyls in DOX and Zr sites under acidic conditions would accelerate the DOX release. In addition, the pH dependent DOX release, the tendency of endogenous phosphate salts in the endosomes to coordinate to the Zr sites also facilitates the release of DOX from the Fe₃O₄@UiO-66 composites. These results indicate that Fe₃O₄@UiO-66 nanocarriers could deliver the drug to tumor tissue sustainably and effectively, which would be beneficial for the diminishing of toxic side effects and decreasing of patient discomfort.

**In vitro cytotoxicity**

*In vitro* cell viabilities of different concentrations of Fe₃O₄@UiO-66, Fe₃O₄@UiO-66-DOX and free DOX on HeLa cells were evaluated by MTT assay to study the bio-toxicity of Fe₃O₄@UiO-66 and the therapeutic effect of Fe₃O₄@UiO-66-DOX. The HeLa cells treated with Fe₃O₄@UiO-66 showed no obvious toxicity (nearly 100% cell viability), even at a concentration up to 500 mg L⁻¹, indicating the good biocompatibility of the synthesized Fe₃O₄@UiO-66 composites (Fig. 4A). In contrast, the Fe₃O₄@UiO-66-DOX exhibited significant increase in anticancer activity against HeLa cells with the increase of the loaded DOX concentration (Fig. 4B). Nearly 60% of HeLa cells were killed after incubation with Fe₃O₄@UiO-66-DOX, even at a low DOX loading concentration of 20 mg L⁻¹ and the Fe₃O₄@UiO-66-DOX had similar cell toxicity to that of free DOX. In addition, the Fe₃O₄@UiO-66-DOX showed greater lethality against HeLa cells after incubating for a longer time (48 h), indicating the long-term and sustained DOX release from the Fe₃O₄@UiO-66 composites.

To demonstrate the low toxic and side effects of DOX loaded Fe₃O₄@UiO-66 on normal cells, we further evaluated the biocompatibility of different concentrations of Fe₃O₄@UiO-66 and Fe₃O₄@UiO-66-DOX on 3T3 cells by MTT assay (Fig. 4C). As expected, the 3T3 cells treated with Fe₃O₄@ UiO-66 showed high viability, further indicating the good biocompatibility of the Fe₃O₄@UiO-66 composites. Furthermore, the DOX loaded Fe₃O₄@UiO-66 also showed negligible toxicity on 3T3 cells, demonstrating the low side effect of Fe₃O₄@UiO-66-DOX on normal cells. Although Fe₃O₄@UiO-66-DOX could be phagocytized by both normal cells and cancer cells, the tremendous difference in cell viability between the HeLa cells and the 3T3 cells may be attributed to the different DOX release rates determined by the cellular microenvironments. The fast reproduction of cancer cells makes the cellular microenvironment acidic, which facilitates DOX release from Fe₃O₄@UiO-66-DOX, and thus leads to high toxicity to cancer cells. These results demonstrate the availability of the Fe₃O₄@UiO-66 composites as drug carriers for cancer cell killing.

**In vitro and in vivo MR imaging**

The high transverse relaxivity of the synthesized Fe₃O₄@UiO-66 core–shell composites gives them potential as a contrast agent for cancer diagnosis. The T₂-weighted MR images of HeLa cells incubated with different concentrations of Fe₃O₄@UiO-66 composites (0, 25, 50, 100, 150 and 200 mg L⁻¹) became much darker with increasing of the concentration of Fe₃O₄@UiO-66 due to the dose-dependent cellular uptake. The results demonstrate the capability of Fe₃O₄@UiO-66 composites as a T₂-weighted MR contrast agent for *in vitro* MR imaging (Fig. 5A).

The feasibility of the Fe₃O₄@UiO-66 composites for *in vivo* MR imaging was also tested. A significant darkening effect was observed in the liver region of the Kunming mouse at 10 min post-injection of Fe₃O₄@UiO-66 composites (400 μL, 5 mg mL⁻¹, 24 mg Fe per kg), indicating the ability of Fe₃O₄@UiO-66 to enhance *in vivo* T₂-weighted images (Fig. 5B). The dramatic T₂ signal intensity decrease is probably due to the phagocytosis of the Fe₃O₄@UiO-66 composites by the liver macrophage cells in reticuloendothelial systems (RES). Considering the excellent
The potential toxic effects of nanomaterials are a major concern for their biomedical applications. Thus, the metabolism, biodistribution and long-term toxicity of the as-synthesized Fe₃O₄@UiO-66 core–shell composites were systematically investigated. For metabolism study, the T₂-weighted images of the Kunming mouse injected with the composites were collected after 1, 7, 14 and 30 days of injection. Strong darkening effect after injection and signal recovery at 30 days post-injection were observed in the liver region, but no time-dependent darkening effect was observed in the urinary bladder, indicating that Fe₃O₄@UiO-66 was not excreted from the kidneys but from the liver (Fig. 5B). For biodistribution study, the major organs (heart, liver, spleen, lung, kidney) of the mice treated with Fe₃O₄@UiO-66 for 1, 7 and 30 days were collected, weighed and solubilized by aqua regia for AAS measurement of Fe and ICP-MS determination of Zr concentrations, respectively. As expected and consistent with the T₂-weighted MR images, high levels of Fe and Zr contents mainly accumulated in mononuclear phagocyte systems such as the spleen and liver (Fig. 6A and S10, ESI†). The Fe levels in all measured organs constantly decreased as the post-injection time prolonged and nearly dropped back to the normal levels after 30 days, except for the liver, in which the Fe content was slightly higher than the control. It was noteworthy that Zr was not detected in the heart, lung or kidney due to the absence of Zr in the organism and no accumulation of Fe₃O₄@UiO-66 in these organs. In addition, the Zr levels in the liver and spleen could be gradually metabolized over time, which was consistent with the biodistribution of Fe.

For in vivo long-term toxicity, the body weight and the blood analysis were evaluated. The body weights of the control group and the experimental group treated with Fe₃O₄@UiO-66 maintained similar increases over 30 days, and no death or significant body weight drop were observed in the experimental group, illustrating that the injection of the Fe₃O₄@UiO-66 composites did not perceivably interfere with the growth of the mice (Fig. S11, ESI†). Blood analysis parameters, including liver function markers (total protein, TP; albumin, ALB; globulin GLB; alanine aminotransferase, ALT; aspartate aminotransferase, AST; alkaline phosphatase, ALKP; gamma glutamyl transaminase, GGT; total bilirubin, TBIL; direct bilirubin, DBIL) and kidney function markers (urea, UREA; creatinine, CREA; uric acid URIC), at different time points post-injection of Fe₃O₄@UiO-66 appeared to be normal compared with those in the control group, indicating that no obvious liver or kidney disorders were induced by the injection of Fe₃O₄@UiO-66 (Fig. 6B). All these results demonstrated that the as-synthesized Fe₃O₄@UiO-66 was a relatively safe theranostic agent for biomedical applications.

**In vivo antitumor efficacy**

Biocompatibility of Fe₃O₄@UiO-66 and in vitro anticancer effect of the DOX-loaded Fe₃O₄@UiO-66, the in vivo...
chemotherapy performance of Fe₃O₄@UiO-66-DOX was investigated using the HeLa tumor-bearing nude mice. As shown in Fig. 7A, the tumors in the mice treated with PBS grew quickly. In contrast, the tumor growth on mice treated with Fe₃O₄@UiO-66-DOX was effectively inhibited due to the preferential DOX accumulation and release at the tumor site via the EPR effect. Furthermore, the images and sizes of the tumor blocks isolated 30 days after being treated showed that the mice treated with Fe₃O₄@UiO-66-DOX had the smallest tumor size (Fig. 7B and C). All these results make a proof of concept that Fe₃O₄@UiO-66 can efficiently carry and release drugs into tumors, leading to effective antitumor efficacy.

Although visualized tumor size could be observed from the images of the mice and tumor blocks, adopting non-invasive MR imaging to monitor tumor development is necessary to realize precision medicine. Thus, monitoring the tumor change of the mice after being treated with Fe₃O₄@UiO-66-DOX via MR imaging was carried out to accurately evaluate the antitumor efficacy. The MR images of HeLa-tumor bearing mice injected with Fe₃O₄@UiO-66-DOX (200 μL, 5 mg mL⁻¹) showed obvious darkening effect in the tumor region, indicating that the Fe₃O₄@UiO-66-DOX could passively accumulate in the tumor at 7 days post-injection (Fig. 7D and S12, ESI†). Continuous monitoring of tumor development by MR imaging was performed and efficient tumor growth inhibition in the mice treated with Fe₃O₄@UiO-66-DOX was observed through the obtained MR images at 21 days post-injection. In contrast, the tumors of the mice treated with PBS grew rapidly, as revealed by MR images. All the results not only further convincingly demonstrated the efficient antitumor efficacy of Fe₃O₄@UiO-66-DOX in terms of MR imaging, but also indicated the great potential of Fe₃O₄@UiO-66 in imaging-guided therapy.

Conclusions

In conclusion, we developed a novel Fe₃O₄@UiO-66 theranostic agent by in situ growth of a UiO-66 MOF shell on a Fe₃O₄ core. The obtained Fe₃O₄@UiO-66 core–shell composites can serve as nanocarriers and contrast agents for simultaneous drug delivery and T₂-weighted MR imaging. The exceptionally high drug loading capacity (≈63 wt%, 2.0 mg DOX per mg composites), and sustained and effective drug release make Fe₃O₄@UiO-66 an excellent drug delivery carrier. Moreover, high transverse relaxivity (255.87 mM⁻¹ s⁻¹) revealed that Fe₃O₄@UiO-66 has the ability to act as a contrast agent for MR imaging. The cytotoxicity assay, biodistribution and in vivo toxicity studies demonstrated that the Fe₃O₄@UiO-66 composites possess low toxicity and good biocompatibility, which inspired us to explore their antitumor efficacy and MR imaging capability in vitro and in vivo. High cancer cell mortality, remarkable tumor size inhibition and significant darkening effect were obtained after treatment with Fe₃O₄@UiO-66 or Fe₃O₄@UiO-66-DOX in vitro and in vivo. All the results indicate that the presented novel multifunctional MOF-based composites should be very promising in cancer therapy and diagnosis due to their effective drug delivery and MR imaging.

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Notes and references
