

Hybrid Dextran-gadolinium Nano-suitcases as High-relaxivity MRI Contrast Agents

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Abstract Dextran-poly(glycidyl methacrylate) (Dex-PGMA) nano-suitcases were synthesized efficiently *via* a graft copolymerization induced self-assembly (GISA) approach. On this basis, the Dex-PGMA nano-suitcases were modified with hydrazide, and the attachment of multiple chelated Gd(III) ions to the interior of the nano-suitcases affords nanoscale MRI contrast agents with high relaxivity values. The highly fenestrated dextran shell of the nano-suitcases assures water exchange which readily occurs between the surrounding environment and the Gd(III) ions encapsulated within the hybrid nano-suitcases. The complexation between the hydrophilic hydrazide interior of the nano-suitcases and Gd(III) ions results in an impressive Gd payload at 22.6 wt% in the hybrid nano-suitcases. The longitudinal relaxivity (r_1) of the hybrid nano-suitcases is reported as 44.4 L/(mmol·s), which is 9–14 folds of that of commercial Gd-DTPA agents. *In vivo* MRI studies demonstrate that the hybrid nano-suitcases accumulated in the lymph node of the rat due to their nanoscale dimensions and displayed strong signals *in vivo*. The results indicated that the hybrid nano-suitcases provide a promising platform for the diagnosis of lymph node related diseases.

Keywords Crosslinked polymer nanoparticles; Gadolinium(III); MRI contrast agents; Lymph node imaging

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INTRODUCTION

Magnetic resonance imaging (MRI) plays a critical role in the diagnosis of various diseases. Clinicians have widely employed this technology due to its excellent properties, such as its non-invasive nature, the use of non-ionizing radiation, high spatial resolution, and high soft tissue resolution in any imaging plane^[1, 2]. The most commonly used clinical MRI T_1 -shortening contrast agents are low molecular weight chelates based on gadolinium ions, such as Gd-diethylenetriaminepentaacetic acid (Gd-DTPA)^[1, 3, 4]. However, the low-molecular-weight Gd chelates generally have low T_1 relaxivity (r_1) and rapidly diffuse out of the blood in the veins, which results in potential toxicity, short imaging time windows, and poor image quality^[1, 5]. All of these points restrict the clinical applications of low-molecular-weight Gd chelates.

In this regard, various carriers have been developed for encapsulation and targeted delivery of the Gd chelates, which include polymeric vesicles, dendrimers, liposomes or micelles with the sizes varied from nanometre to micron

scales^[5–14]. Among them, some cases exhibit excellent MRI sensitivity, such as Gd-decorated polymer vesicles and liposomes. Based on reported examples, it has been widely recognized that the sizes of the carriers have a key influence on both the MRI relaxivity and the distributions of the cargos in different tissues^[15–17]. For example, particles with a diameter of 30–300 nm could accumulate selectively in the sentinel lymph node and thus were very efficient for specifically sentinel lymph node mapping^[6, 7]. In addition, the confinement of the Gd chelates in nanoscale carriers was confirmed to help improve the MRI relaxivity^[5]. However, the most of current Gd-loaded carriers, such as liposomes and micelles, are stabilized by non-covalent bonds and do not demonstrate enough stability in biological environments^[5, 8]. Besides, there is still space to achieve higher MRI resolution^[18–20]. Therefore, there remains a need for developing a versatile carrier that is both stable in biological environments and can provide high MRI relaxivity.

Dextran, as a typical carbohydrate, demonstrates an outstanding combination of attractive features, including biocompatibility, biodegradability and crucial biological functions^[21–25]. These characteristics make dextran a promising precursor for preparing Gd-loaded hybrid MRI enhancement agents. As being disclosed, dextran-based outer shell of the carriers may add more functionalities to the

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carriers, such as lymph node mapping^[7] and recognition capability for specific cells^[26]. Based on the consideration as above, here dextran-gadolinium hybrid nano-suitcases with high r_1 value were fabricated *via* the complexation between the multiple chelated Gd(III) ions (Gd-DTPA) and hydrazide modified dextran-poly(glycidyl methacrylate) (Dex-PGMA) nano-suitcases synthesized by a graft copolymerization induced self-assembly (GISA) approach. On the basis of fabricating dextran-gadolinium hybrid nano-suitcases, their size, morphology, cytotoxicity, longitudinal relaxivity (r_1) as well as the transverse relaxivity (r_2) were characterized. The potential of these hybrid nano-suitcases as MRI contrast enhancement agents for lymph node imaging was investigated. The *in vivo* studies indicated that these hybrid nano-suitcases demonstrated a high MR imaging efficiency for lymph nodes.

EXPERIMENTAL

Materials and Methods

Dextran ($M_w = 4 \times 10^4$ Da), ceric ammonium nitrate (CAN, $\geq 99\%$), hydrazinium hydroxide (85%) and dimethyl sulfoxide were purchased from Shanghai Sinopharm Chemical Reagent Co., Ltd. Glycidyl methacrylate (97%, Aladdin) and diallyl disulfide (DADS, 80%, Sigma Aldrich) were used after purification by vacuum distillation. Gadopentetic acid dimeglumine salt injection (Gd-DTPA) was purchased from Bayer Vital GmbH (Berlin, Germany).

The structure of the nano-suitcases was characterized *via* proton nuclear magnetic resonance spectroscopy ($^1\text{H-NMR}$, Avance III 400 MHz spectrometer, Bruker, Switzerland). The hydrodynamic diameters of the nano-suitcases were evaluated using the dynamic light scattering measurement (DLS, Zetasizer Nano ZS90 instrument, Malvern, U.K.). The morphology of the nano-suitcases was observed by transmission electron microscopy (TEM, Tecnai G2 spirit Biotwin, 120 kV, FEI, U.S.A.). The element analysis of Dex-PGMA-Gd(III) nano-suitcases was carried out using an energy-dispersive detector (EDS, Tecnai G2 spirit Biotwin, 200 kV, FEI, U.S.A.).

Fabrication of Dex-PGMA Nano-suitcases

Dex-PGMA nano-suitcases were fabricated by following our published procedures^[27–29] with minor modifications. Briefly, 1.500 g of dextran was dissolved in 30 mL of water in a 100 mL 3-neck round-bottomed flask. The solution was kept under N_2 protection at 30 °C. Subsequently, the initiator CAN (0.6993 g, pre-dissolved in 0.75 mL of 0.1 mol/L HNO_3) was injected *via* a syringe, and 5 min later the monomer, GMA (1.224 mL, dissolved in 2.5 mL of DMSO) was added to the solution. After 30 min, the crosslinker DADS (0.144 mL, dissolved in 0.5 mL of DMSO) was added into the system. The reaction was allowed to proceed for 4 h. Finally, the Dex-PGMA nano-suitcases were purified by dialysis using Milli-Q and then lyophilized to obtain the required nanoparticles denoted as Dex-PGMA nano-suitcases. The yield was calculated as follows using Eq. (1):

$$\text{Yield (\%)} = \frac{W_{\text{Dex-PGMA}}}{W_{\text{Dex}} + W_{\text{GMA}} + W_{\text{DADS}}} \times 100\% \quad (1)$$

where W_{Dex} , W_{GMA} , W_{DADS} , $W_{\text{Dex-PGMA}}$ are the weights of dextran, GMA, DADS and Dex-PGMA nano-suitcases, respectively.

Fabrication of Dex-PGMA-NHNH₂

Dex-PGMA nano-suitcases (0.800 g) were thoroughly dispersed in 110 mL of water in a 250 mL 3-neck round-bottomed flask, and heated to 100 °C. After 5 min, 14.3 mL of 85% *V/V* hydrazinium hydroxide was added dropwise to the solution. The reaction solution was refluxed and mechanically stirred at a speed of 500 r/min for 3 h. Finally, the sample solution was purified by dialysis using Milli-Q water and then freeze-dried to obtain the hydrazide modified Dex-PGMA nanoparticles denoted as Dex-PGMA-NHNH₂ nano-suitcases.

Fabrication of Dex-PGMA-Gd(III) Nano-suitcases

Dex-PGMA-NHNH₂ nano-suitcases (0.040 g) were dispersed in 40 mL of water. Subsequently, 15 mL of gadopentetic acid dimeglumine salt injection (0.5 mol/L) was added dropwise to the solution. The reaction solution was stirred for 24 h at 25 °C. Finally, the solution was purified by dialysis using Milli-Q water and then lyophilized to obtain the Gd ions modified Dex-PGMA nanoparticles defined as Dex-PGMA-Gd(III) nano-suitcases.

Solution Stability of Gd-DTPA in Hybrid Dex-PGMA-Gd(III) Nano-suitcases

The release rate of Gd(III) ions from hybrid dextran-gadolinium nano-suitcases was investigated to evaluate the stability of sample. The Dex-PGMA-Gd(III) nano-suitcases stored at 4 °C for 2 days were used to test the *in vitro* Gd(III) ions release rate. The test was conducted *via* the dialysis method. The detailed procedure is as follows: 1 mL of a 50 mmol/L Dex-PGMA-Gd(III) nano-suitcases in normal saline solution stored at 4 °C for 2 days was transferred into a dialysis bag (molecular weight cut-off = 1.4×10^4 Da) and was then immersed into 9 mL of normal saline for 6 h. The weight of the Gd(III) ions in dialyzate and total weight in Dex-PGMA-Gd(III) nano-suitcases before dialysis were determined by ICP-AES respectively. The release content of Gd(III) ions in 2 days was calculated as follows using Eq. (2):

$$\text{Release content of Gd(III) (\%)} = \frac{W_{\text{dialyzate}}}{W_{\text{nano-suitcases}}} \times 100\% \quad (2)$$

where $W_{\text{dialyzate}}$ and $W_{\text{nano-suitcases}}$ are the weights of the Gd(III) ions released into dialyzate and contained in Dex-PGMA-Gd(III) nano-suitcases before dialysis.

Cell Viability Assays

The lymphatic endothelial cells (LECs) were purchased from the Shanghai Institute of Cell Biology, the Chinese Academy of Sciences (Shanghai, China). LECs were seeded at a density of 8000 cells/well in 96-well cell culture plates under 5% CO_2 at 37 °C for 24 h. Subsequently, the fresh culture medium with various concentrations of the Dex-PGMA-Gd(III) was added to each well to replace the old medium ($n = 5$). After being co-incubated with Dex-PGMA-Gd(III) for 24 h, the cell viability was measured by the standard 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide (MTT) assays.

Relaxivity

The relaxivity of Dex-PGMA-Gd(III) nano-suitcases was evaluated with a 3.0 T PHILIPS Ingenia scanner. T_1 and T_2 values of Dex-PGMA-Gd(III) nano-suitcases in agar solution with various concentrations were measured. The parameters of the T_1 measurement were as follows: spin echo sequence, TE = 9 ms, TR = 100, 200, 300, 400, 500, 600, 700 ms. The parameters of the T_2 measurement were as follows: multi-echo sequence, TR = 2000 ms, TE = 20, 40, 60, 80, 100, 120, 140, 160 ms. The concentration of the Gd ions in the nano-suitcases was confirmed by an inductively-coupled plasma emission spectrometer (ICP-AES, iCAP 6300, Thermo Fisher Scientific Inc., U.S.A.).

In Vivo MR Imaging

Female Wistar rats aged 10–12 weeks (acquired from West Poole Will Kai Experimental Animal Co. Ltd, Shanghai, China) were maintained under specific pathogen free conditions. All experiments employing rats were performed in accordance with the Chinese NIH guidelines for the care and use of laboratory research animals. For *in vivo* experiments, according to previous reported methods^[7], 50 mL of Dex-PGMA-Gd(III) nano-suitcases (20 mmol/L) at the dose of 0.1 mmol/kg was intradermally injected into the forepaw of Wistar rats, and its *in vivo* MRI was recorded using a PHILIPS Ingenia scanner. The T_1 -weighted image was obtained using the following parameters: TR 6 ms, TE 3 ms, image matrix 240×240 , flip angle 25° , FOV 120 mm, slice thickness 0.7 mm.

RESULTS AND DISCUSSION

Fabrication of Dex-PGMA Nano-suitcases

Dextran-poly(glycidyl methacrylate) (Dex-PGMA) nano-suitcases were synthesized efficiently *via* a graft copolymerization induced self-assembly (GISA) approach. In our previous work, we had demonstrated that dextran-based nanogels could be efficiently prepared through a graft copolymerization induced self-assembly (GISA) approach induced by hydrogen bonding^[7, 27, 29]. Inspired by this work on one hand, and the unprecedented potential for biomedical applications provided by dextran-based nano-suitcases on the other hand, we report the successful combination with a view of forming Dex-PGMA nano-suitcases with hydrophobic PGMA inner crosslinked cores and narrowly dispersed diameters for the delivery of multiple chelated Gd(III) ions. The synthetic route and mechanism of Dex-PGMA nano-suitcase formation are proposed as depicted in Scheme 1. Here we respectively selected dextran, glycidyl methacrylate

(GMA), ceric ammonium nitrate (CAN), and diallyl disulfide (DADS) as the backbone, graft precursor, initiator, and crosslinker in an aqueous reaction medium, as water is a selective solvent that is good for the dextran backbone chain, but poor for the PGMA grafts, and CAN is efficient for initiating free radical graft polymerization of various vinyl monomers from polysaccharide chains. The self-assembly of PGMA containing amphiphilic copolymers in aqueous solution is well-understood^[30] and it was therefore anticipated that at a threshold degree of polymerization, the PGMA grafts would become insoluble, and thus GISA would occur.

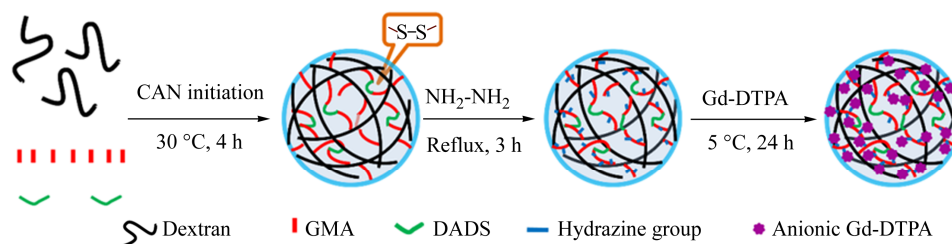
The successful preparation of Dex-PGMA nano-suitcases was confirmed by $^1\text{H-NMR}$ spectroscopy (Fig. 1a). The proton signals in the range of 3.0–5.0 and 0.5–3.0 ppm corresponded to the dextran moiety and PGMA chains, respectively^[7, 31]. The molar ratio of GMA to the glucose repeat units of Dex-PGMA was *ca.* 0.7 to 1, which was calculated based on $^1\text{H-NMR}$ spectra. The Dex-PGMA nano-suitcases exhibited a spherical shape with a diameter of 50–80 nm in the dry state under TEM observation as shown in Fig. 2(a). The DLS characterization (Fig. 2b) revealed that Dex-PGMA nano-suitcases had a hydrodynamic diameter $\langle D_h \rangle$ of *ca.* 138 nm. The particle dispersion index (PDI) of the sample was 0.208. Furthermore, the yield of Dex-PGMA nano-suitcases was 78%, and solid content of the Dex-PGMA nano-suitcases in reactant medium reached up to 40 mg/mL.

Fabrication of Dex-PGMA-NHNH₂ Nano-suitcases

The Dex-PGMA nano-suitcases were modified by hydrazine groups to facilitate their loading with Gd-DTPA. In this regard, Dex-PGMA-NHNH₂ nano-suitcases were prepared *via* the procedure as shown in Scheme 1. A $^1\text{H-NMR}$ spectrum of the sample is shown in Fig. 1(b). The disappearance of proton signals corresponding to the epoxide group in the range of 2.5–3.0 ppm indicated the successful synthesis of Dex-PGMA-NHNH₂ nano-suitcases. The values of the hydrodynamic diameter and PDI (Fig. 2d) of the Dex-PGMA-NHNH₂ nano-suitcases are similar to those of Dex-PGMA. TEM images as shown in Fig. 2(c) revealed that Dex-PGMA-NHNH₂ nano-suitcases still maintained the spherical morphology.

Fabrication of Dex-PGMA-Gd(III) Hybrid Nano-suitcases

It is known that electrostatic interactions occur between the carboxyl group of DTPA and the amino group in aqueous media^[3]. With this in mind, we propose a hypothesis that Gd-DTPA may bind to Dex-PGMA-NHNH₂ nano-suitcases due to the non-covalent interactions between the hydrazine group of the nano-suitcases and the carboxyl group of DTPA.



Scheme 1 Schematic illustration depicting the fabrication of Dex-PGMA-Gd(III) nano-suitcases

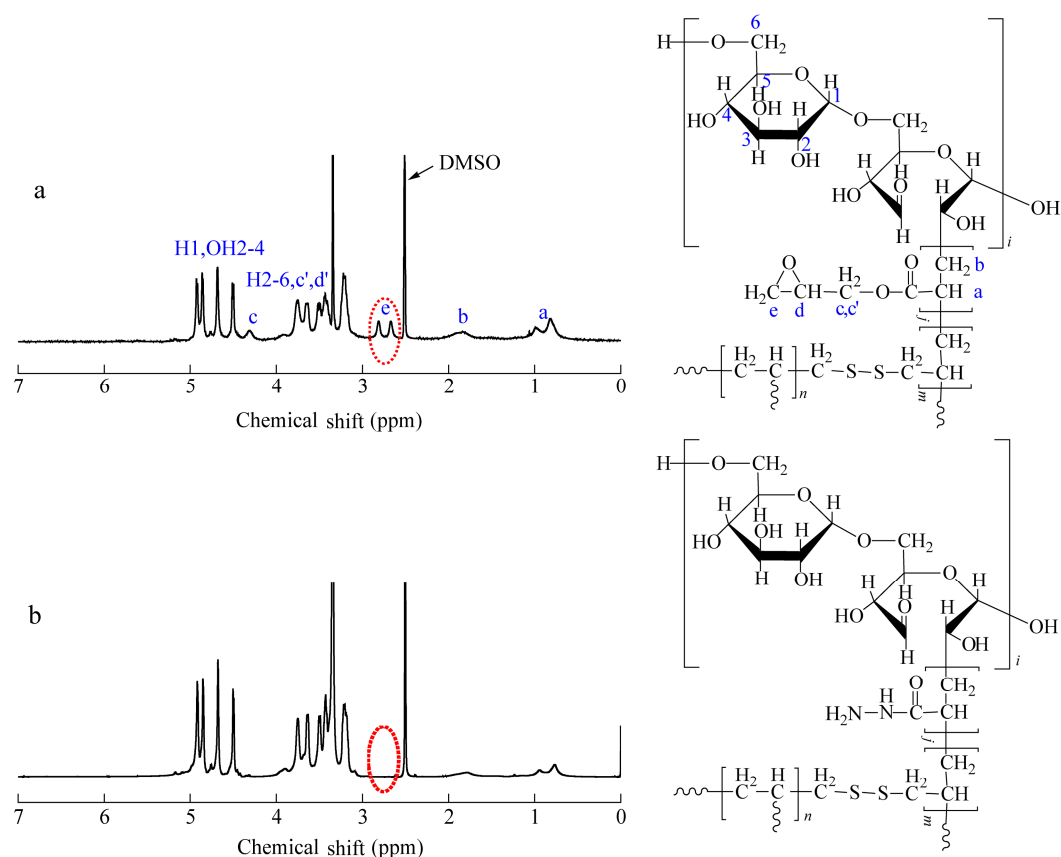


Fig. 1 ^1H -NMR spectra (DMSO- d_6) of Dex-PGMA nano-suitcases (a) and Dex-PGMA-NHNH $_2$ nano-suitcases (b)

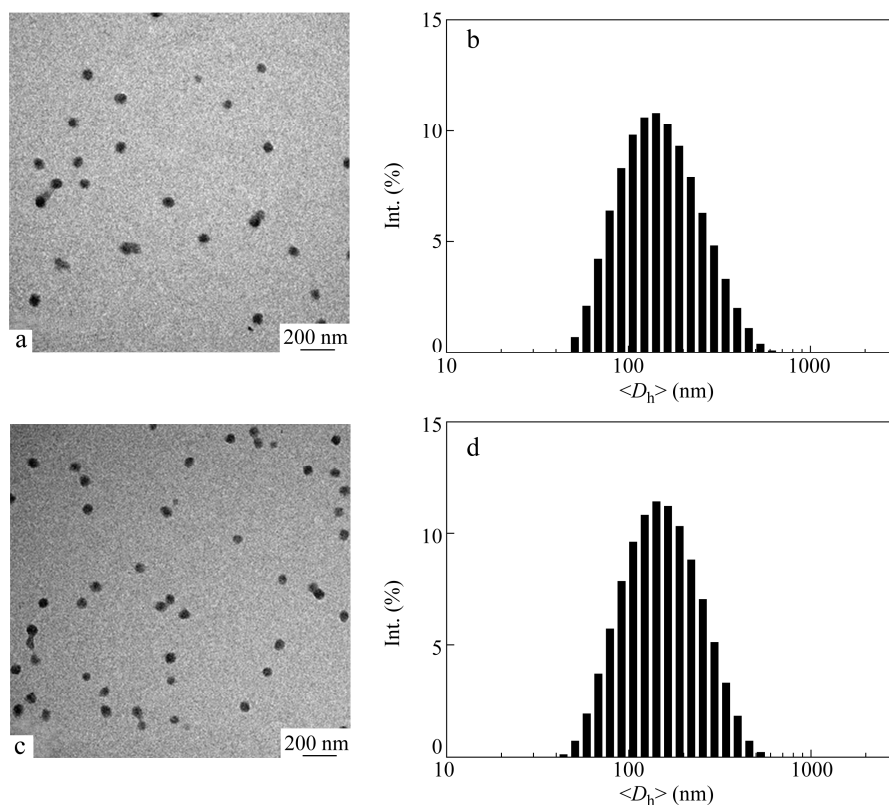


Fig. 2 TEM images of Dex-PGMA nano-suitcases (a) and Dex-PGMA-NHNH $_2$ nano-suitcases (c), DLS measurements for Dex-PGMA nano-suitcases (b) and Dex-PGMA-NHNH $_2$ nano-suitcases (d)

The prepared Dex-PGMA-Gd(III) hybrid nano-suitcases were characterized by TEM, DLS and element analysis. As shown in Fig. 3, the results revealed that the Dex-PGMA-Gd(III) hybrid nano-suitcases were spherical with a diameter of 120–160 nm in the dry state and had a hydrodynamic diameter $\langle D_h \rangle$ of *ca.* 185 nm in aqueous solution, which is slightly larger than that of Dex-PGMA-NHNH₂ nano-suitcases. The element analysis characterization showed that Gd-DTPA was mainly distributed on the outer layer of the Dex-PGMA-NHNH₂ nano-suitcases. The content of the Gd(III) in the hybrid nano-suitcases was 22.6 wt%, as confirmed by ICP-AES.

Stability of Gd-DTPA in Hybrid Dex-PGMA-Gd(III) Nano-suitcases

The stability of Gd containing Dex-PGMA-Gd(III) nano-suitcases is crucial for clinical application. In this regard, the stability of the nano-suitcases in both the solution and dry states was studied by using DLS and ICP-AES. The release content of Gd(III) ions from hybrid dextran-gadolinium nano-suitcases after being stored in solution state for 2 days was determined as 6.1%. The result indicates that the hybrid nano-suitcases can be stored in solution state for a short term of 2 days. Besides, our study disclosed good stability of the Dex-PGMA-Gd(III) nano-suitcases in their sizes when being stored in dry state ($\langle D_h \rangle$ of 185 nm after preparation and 171 nm after being stored in dry state for 17 days). The sizes of the nano-suitcases determined freshly after being prepared and after being stored in dry state for 17 days are shown in Fig. 3(b). On this basis, the long-term storage of the Dex-PGMA-Gd(III) nano-suitcases can be proceeded in dry state.

Relaxivity

Relaxivity (r_1) is a parameter to evaluate the MRI enhancement capability of MRI agents. The solvent relaxation rate is linearly dependent on the concentration of gadolinium ions^[24]. A curve of solvent relaxation rate $1/T_1$

versus the concentration of Gd(III) was plotted, with the relaxivity corresponding to the slope of this curve. According to the curve shown in Fig. 4, the longitudinal relaxivity (r_1) of the Dex-PGMA-Gd(III) nano-suitcases was 44.4 L/(mmol·s), which is between 9- and 14-fold that of commercial Magnevist (Gd-DTPA, $r_1 = 3.12\text{--}4.8$ L/(mmol·s))^[4, 32–37], and much higher than that of many nanoscale contrast agents^[38]. The transverse relaxivity (r_2) of the Dex-PGMA-Gd(III) nano-suitcases was 9.54 L/(mmol·s). The low r_2/r_1 ratio of 0.21 indicated that it was a positive contrast agent and had a very promising r_1 effect^[39, 40]. The excellent r_1 relaxivities might be due to the high water diffusion coefficient of the crosslinked inner hybrid cores and the nanoscaled sizes of the Dex-PGMA-Gd(III) nano-suitcases^[5, 8, 14, 41, 42]. The highly fenestrated dextran shell of the nano-suitcases assures water exchange between the surrounding environment and the Gd(III) ions encapsulated within the hybrid nano-suitcases. The complexation between the hydrophilic hydrazide interior of the nano-suitcases and the Gd(III) ions resulted in the impressive payload of Gd at 22.6 wt% in the hybrid nano-suitcases. In a confined nanoscale space, Dex-PGMA-Gd(III) nano-suitcases exhibited an effective r_1 relaxation rate, indicating that they have promising potential as MRI contrast agents for the detection and diagnosis of various diseases.

In Vitro MTT Assays

The biosafety of these Gd-loaded nano-suitcases is the most critical factor determining suitability of a biomedical material. Therefore, the *in vitro* effect of Gd-loaded nano-suitcases on the growth of healthy cells, lymphatic endothelial cells (LECs), was evaluated *via* the MTT assay method, and the cytotoxicity is shown in Fig. 5. The viability of LECs incubated with Dex-PGMA-Gd(III) nano-suitcases at a high Gd(III) concentration (0.5 mmol/L) was 96.56% relative to the control. This finding indicated that Dex-PGMA-Gd(III) nano-suitcases had low cytotoxicity and high biocompatibility with LECs.

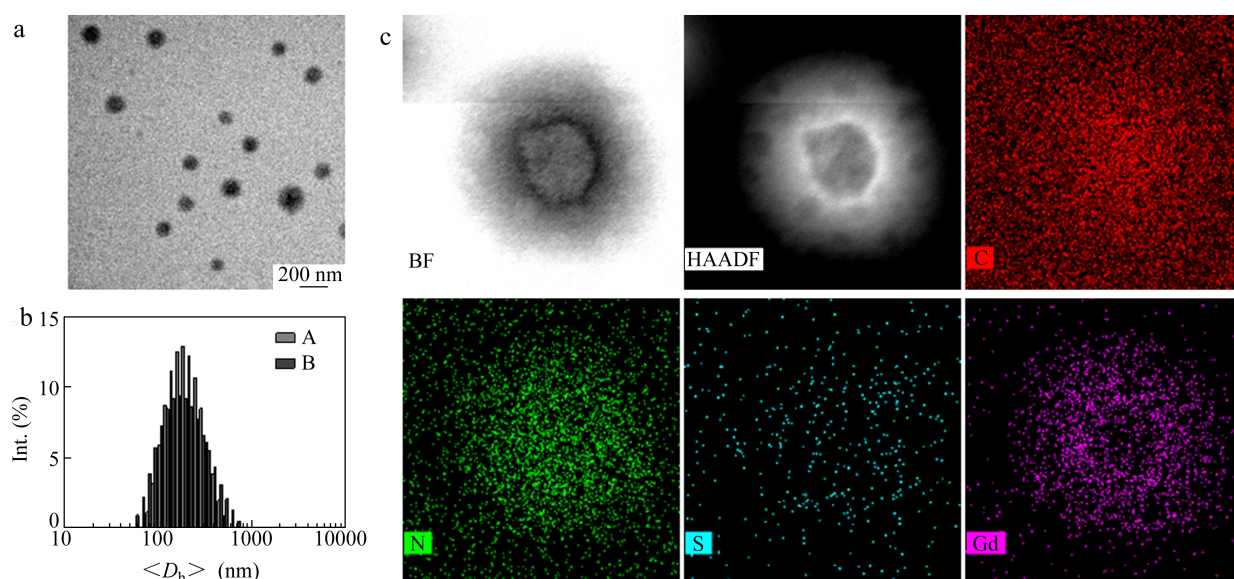


Fig. 3 TEM image of Dex-PGMA-Gd(III) nano-suitcases (a); DLS measurements (b) for Dex-PGMA-Gd(III) nano-suitcases freshly after being prepared (A) and after being stored for 17 days in dry state (B); The element analysis results of Dex-PGMA-Gd(III) nano-suitcases (c)

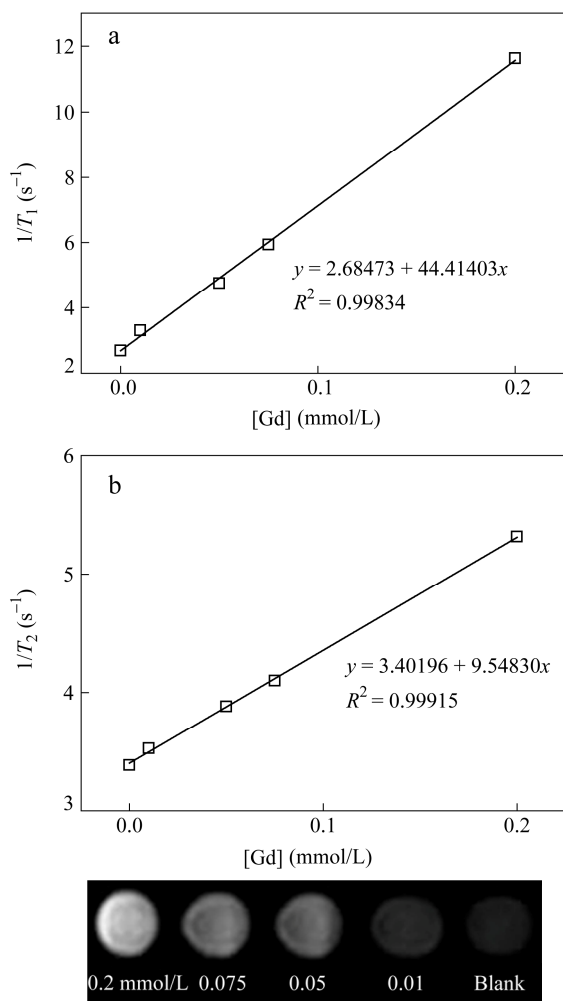


Fig. 4 r_1 relaxivity curve (a), r_2 relaxivity curve (b) and T_1 -weighted MRI (c) of Dex-PGMA-Gd(III) hybrid nano-suitcases at various Gd concentrations

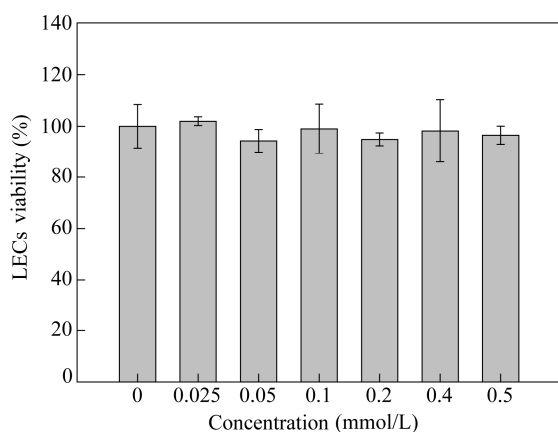


Fig. 5 LECs viability of Dex-PGMA-Gd(III) nano-suitcases

In Vivo MR Imaging

The accurate localization of the sentinel lymph node plays a major role in cancer detection to determine if tumor metastasis has occurred through the biopsy of sentinel lymph nodes^[7]. Our DLS study disclosed that Dex-PGMA-Gd(III) nano-suitcases had hydrodynamic diameters of 185 nm, which is a suitable particle size for lymph node imaging^[6, 7].

With this in mind, the possibility that the lymph node is accurately located through *in vivo* MR imaging using Dex-PGMA-Gd(III) nano-suitcases was investigated (Fig. 6). An *in vivo* study showed that these Dex-PGMA-Gd(III) nano-suitcases rapidly accumulated in the lymph node of the rat at a high level, which was visualized within 1 min after injection, indicating its high imaging efficiency and confirming that accurate localization of lymph nodes could be achieved using these Dex-PGMA-Gd(III) nano-suitcases. These hybrid nano-suitcases selectively diffused into the lymph node rather than blood vessels because of their matching particle size ($\langle D_h \rangle$ of ca. 185 nm).

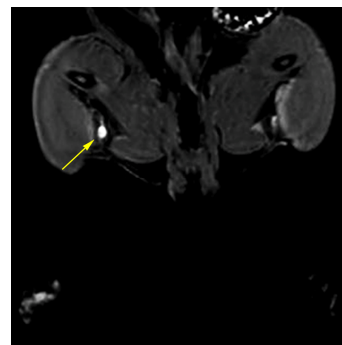


Fig. 6 T_1 -weighted MR imaging of lymph node of rat injected with Dex-PGMA-Gd(III) nano-suitcases ([Gd] = 0.1 mmol/L. The arrow refers to the lymph node.)

CONCLUSIONS

In summary, Gd(III)-loaded hybrid nano-suitcases with excellent MRI enhancement capability and a suitable size for lymph node targeting, dextran-poly(glycidyl methacrylate) (Dex-PGMA) nano-suitcases, were prepared *via* a facile GISA strategy. The r_1 relaxivity and r_2 relaxivity of the Dex-PGMA-Gd(III) nano-suitcases were 44.4 and 9.54 L/(mmol·s), respectively. The Dex-PGMA-Gd(III) nano-suitcases exhibited higher imaging efficiency than both commercial Gd-DTPA and most of the other reported Gd(III)-loaded nanoscale contrast agents. The *in vivo* MRI of the lymph node demonstrated that Dex-PGMA-Gd(III) nano-suitcases have the potential to be used as a contrast agent for the detection of lymph node related diseases.

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REFERENCES

- Kim, J. H.; Park, K.; Nam, H. Y.; Lee, S.; Kim, K.; Kwon, I. C. Polymers for bioimaging. *Prog. Polym. Sci.* 2007, 32(8), 1031–1053.
- de Leon-Rodriguez, L. M.; Lubag, A. J. M.; Malloy, C. R.;

- Martinez, G. V.; Gillies, R. J.; Sherry, A. D. Responsive MRI agents for sensing metabolism *in vivo*. *Acc. Chem. Res.* 2009, 42(7), 948–957.
- 3 Darras, V.; Nelea, M.; Winnik, F. M.; Buschmann, M. D. Chitosan modified with gadolinium diethylenetriaminepentaacetic acid for magnetic resonance imaging of DNA/chitosan nanoparticles. *Carbohydr. Polym.* 2010, 80(4), 1137–1146.
 - 4 van der Elst, L.; Raynaud, J. S.; Vives, V.; Santus, R.; Louin, G.; Robert, P.; Port, M.; Corot, C.; Muller, R. Comparative relaxivities and efficacies of gadolinium-based commercial contrast agents. *Proceedings of the 21st Annual Meeting of ISMRM*. 2013.
 - 5 Soleimani, A.; Martinez, F.; Economopoulos, V.; Foster, P. J.; Scholl, T. J.; Gillies, E. R. Polymer cross-linking: a nanogel approach to enhancing the relaxivity of MRI contrast agents. *J. Mater. Chem. B* 2013, 1(7), 1027–1034.
 - 6 Noh, Y. W.; Kong, S. H.; Choi, D. Y.; Park, H. S.; Yang, H. K.; Lee, H. J.; Kim, H. C.; Kang, K. W.; Sung, M. H.; Lim, Y. T. Near-infrared emitting polymer nanogels for efficient sentinel lymph node mapping. *ACS Nano* 2012, 6(9), 7820–7831.
 - 7 Dai, T.; Zhou, S.; Yin, C.; Li, S.; Cao, W.; Liu, W.; Sun, K.; Dou, H.; Cao, Y.; Zhou, G. Dextran-based fluorescent nanoprobe for sentinel lymph node mapping. *Biomaterials* 2014, 35(28), 8227–8235.
 - 8 Li, Y.; Beija, M.; Laurent, S.; Elst, L. V.; Müller, R. N.; Duong, H. T.; Lowe, A. B.; Davis, T. P.; Boyer, C. Macromolecular ligands for gadolinium MRI contrast agents. *Macromolecules* 2012, 45(10), 4196–4204.
 - 9 Caravan, P. Strategies for increasing the sensitivity of gadolinium based MRI contrast agents. *Chem. Soc. Rev.* 2006, 35(6), 512–523.
 - 10 Casali, C.; Janier, M.; Canet, E.; Obadia, J. F.; Benderbous, S.; Corot, C.; Revel, D. Evaluation of Gd-DOTA-labeled dextran polymer as an intravascular MR contrast agent for myocardial perfusion. *Acad. Radiol.* 1998, 5, S214–S218.
 - 11 Jacques, V.; Desreux, J.; New classes of MRI contrast agents. *Contrast Agents I* 2002, 123–164.
 - 12 Major, J. L.; Meade, T. J. Bioresponsive, cell-penetrating, and multimeric MR contrast agents. *Acc. Chem. Res.* 2009, 42(7), 893–903.
 - 13 Liu, Q.; Zhu, H.; Qin, J.; Dong, H.; Du, J. Theranostic vesicles based on bovine serum albumin and poly(ethylene glycol)-block-poly(L-lactic-co-glycolic acid) for magnetic resonance imaging and anticancer drug delivery. *Biomacromolecules* 2014, 15(5), 1586–1592.
 - 14 Liu, Q.; Chen, S.; Chen, J.; Du, J. An asymmetrical polymer vesicle strategy for significantly improving T_1 MRI sensitivity and cancer-targeted drug delivery. *Macromolecules* 2015, 48(3), 739–749.
 - 15 Shalgunov, V.; Zaytseva-Zotova, D.; Zintchenko, A.; Levada, T.; Shilov, Y.; Andreyev, D.; Dzhumashev, D.; Metelkin, E.; Urusova, A.; Demin, O. Comprehensive study of the drug delivery properties of poly(L-lactide)-poly(ethylene glycol) nanoparticles in rats and tumor-bearing mice. *J. Control. Release* 2017, DOI: <https://doi.org/10.1016/j.jconrel.2017.06.006>
 - 16 Zhang, Q. L.; Wang, H. Y.; Ge, C. C.; Duncan, J.; He, K. H.; Adeosun, S. O.; Xi, H. X.; Peng, H. T.; Niu, Q. Alumina at 50 and 13 nm nanoparticle sizes have potential genotoxicity. *J. Appl. Toxicol.* 2017, 37, 1053–1064.
 - 17 Wang, L. Y.; Huang, J.; Chen, H. B.; Wu, H.; Xu, Y. L.; Li, Y. C.; Yi, H.; Wang, Y. A.; Yang, L.; Mao, H. Exerting enhanced permeability and retention effect driven delivery by ultrafine iron oxide nanoparticles with T_1 - T_2 switchable magnetic resonance imaging contrast. *ACS Nano* 2017, 11(5), 4582–4592.
 - 18 Taylor, K. M. L.; Kim, J. S.; Rieter, W. J.; An, H.; Lin, W.; Lin, W. B. Mesoporous silica nanospheres as highly efficient MRI contrast agents. *J. Am. Chem. Soc.* 2008, 130(7), 2154–2155.
 - 19 Godin, B.; Tasciotti, E.; Liu, X. W.; Serda, R. E.; Ferrari, M. Multistage nanovectors: from concept to novel imaging contrast agents and therapeutics. *Acc. Chem. Res.* 2011, 44(10), 979–989.
 - 20 Chen, K. J.; Wolahan, S. M.; Wang, H.; Hsu, C. H.; Chang, H. W.; Durazo, A.; Hwang, L. P.; Garcia, M. A.; Jiang, Z. K.; Wu, L. A small MRI contrast agent library of gadolinium(III)-encapsulated supramolecular nanoparticles for improved relaxivity and sensitivity. *Biomaterials* 2011, 32(8), 2160–2165.
 - 21 Liu, Z. H.; Jiao, Y. P.; Wang, Y. F.; Zhou, C. R.; Zhang, Z. Y. Polysaccharides-based nanoparticles as drug delivery systems. *Adv. Drug Deliv. Rev.* 2008, 60(15), 1650–1662.
 - 22 Hu, Y.; Li, Y.; Xu, F. J. Versatile Functionalization of polysaccharides *via* polymer grafts: from design to biomedical applications. *Acc. Chem. Res.* 2017, 50(2), 281–292.
 - 23 Mizrahy, S.; Peer, D. Polysaccharides as building blocks for nanotherapeutics. *Chem. Soc. Rev.* 2012, 41(7), 2623–2640.
 - 24 Du, H. J.; Shen, Y. C.; Liu, Y. P.; Han, L.; Zheng, Y.; Yan, G. P.; Tu, Y. Y.; Wu, J. Y.; Guo, Q. Z.; Zhang, Y. F.; Xia, X. T.; Lan, X. L.; Xia, X. T. Dextran gadolinium complex containing folate groups as a potential magnetic resonance imaging contrast agent. *Chinese J. Polym. Sci.* 2015, 33(9), 1325–1333.
 - 25 Klaveness, J. Water-soluble polysaccharides as carriers of paramagnetic contrast agents for magnetic resonance imaging: synthesis and relaxation properties. *Carbohydr. Res.* 1991, 214(2), 315–323.
 - 26 Pustynnikov, S.; Sagar, D.; Jain, P.; Khan, Z. K. Targeting the C-type lectins-mediated host-pathogen interactions with dextran. *J. Pharm. Pharm. Sci.* 2014, 17(3), 371–392.
 - 27 Zhou, S.; Dou, H.; Zhang, Z.; Sun, K.; Jin, Y.; Dai, T.; Zhou, G.; Shen, Z. Fluorescent dextran-based nanogels: efficient imaging nanoprobe for adipose-derived stem cells. *Polym. Chem.* 2013, 4(15), 4103–4112.
 - 28 Zhou, S.; Min, X.; Dou, H.; Sun, K.; Chen, C. Y.; Chen, C. T.; Zhang, Z.; Jin, Y.; Shen, Z. Facile fabrication of dextran-based fluorescent nanogels as potential glucose sensors. *Chem. Commun.* 2013, 49(82), 9473–9475.
 - 29 Wang, H.; Dai, T.; Zhou, S.; Huang, X.; Li, S.; Sun, K.; Zhou, G.; Dou, H.; Self-assembly assisted fabrication of dextran-based nanohydrogels with reduction-cleavable junctions for applications as efficient drug delivery systems. *Sci. Rep.* 2017, 7, DOI: [10.1038/srep40011](https://doi.org/10.1038/srep40011).
 - 30 Li, Q. L.; Gu, W. X.; Gao, H.; Yang, Y. W. Self-assembly and applications of poly (glycidyl methacrylate)s and their derivatives. *Chem. Commun.* 2014, 50(87), 13201–13215.
 - 31 Sousani, A.; Moghadam, P. N.; Hasanzadeh, R.; Motiei, H.; Bagheri, M. Synthesis of poly glycidylmethacrylate grafted azobenzene copolymer: photosensitivity and nonlinear optical properties. *Opt. Mater.* 2016, 51, 232–240.
 - 32 Lauffer, R. B. Paramagnetic metal complexes as water proton relaxation agents for NMR imaging: theory and design. *Chem. Rev.* 1987, 87(5), 901–927.
 - 33 Zheng, J. P.; Liu, Q. L.; Zhen, M. M.; Jiang, F.; Shu, C. Y.; Jin, C.; Yang, Y.; Alhadlaq, H. A.; Wang, C. R. Multifunctional imaging probe based on gadofulleride nanoplateform. *Nanoscale* 2012, 4(12), 3669–3672.
 - 34 Borges, M.; Yu, S.; Laromaine, A.; Roig, A.; Suárez-García, S.; Lorenzo, J.; Ruiz-Molina, D.; Novio, F. Dual T_1/T_2 MRI contrast agent based on hybrid SPION@ coordination polymer nanoparticles. *RSC Adv.* 2015, 5(105), 86779–86783.

- 35 Park, J. Y.; Kim, S. J.; Lee, G. H.; Jin, S.; Chang, Y.; Bae, J. E.; Chae, K. S. Various ligand-coated ultrasmall gadolinium-oxide nanoparticles: water proton relaxivity and *in-vivo* T_1 MR image. *J. Korean Phys. Soc.* 2015, 66(8), 1295–1302.
- 36 Le, W.; Cui, S.; Chen, X.; Zhu, H.; Chen, B.; Cui, Z. Facile synthesis of Gd-functionalized gold nanoclusters as potential MRI/CT contrast agents. *Nanomaterials* 2016, 6(4), DOI: 10.3390/nano6040065
- 37 Zhang, J.; Hao, G.; Yao, C.; Hu, S.; Hu, C.; Zhang, B. Paramagnetic albumin decorated CuInS 2/ZnS QDs for CD133+ glioma bimodal MR/fluorescence targeted imaging. *J. Mater. Chem. B* 2016, 4(23), 4110–4118.
- 38 Della Rocca, J.; Liu, D.; Lin, W. Nanoscale metal-organic frameworks for biomedical imaging and drug delivery. *Acc. Chem. Res.* 2011, 44(10), 957–968.
- 39 Favier, A.; D'Agosto, F.; Charreyre, M. T.; Pichot, C. Synthesis of *N*-acryloxysuccinimide copolymers by RAFT polymerization, as reactive building blocks with full control of composition and molecular weights. *Polymer* 2004, 45(23), 7821–7831.
- 40 Chen, Z.; Yu, D.; Liu, C.; Yang, X.; Zhang, N.; Ma, C.; Song, J.; Lu, Z. Gadolinium-conjugated PLA-PEG nanoparticles as liver targeted molecular MRI contrast agent. *Drug Target.* 2011, 19(8), 657–665.
- 41 Liu, Q.; Song, L.; Chen, S.; Gao, J.; Zhao, P.; Du, J. A superparamagnetic polymersome with extremely high T_2 relaxivity for MRI and cancer-targeted drug delivery. *Biomaterials* 2017, 114, 23–33.
- 42 Song, S.; Guo, H.; Jiang, Z.; Jin, Y.; Wu, Y.; An, X.; Zhang, Z.; Sun K.; Dou, H. Self-assembled microbubbles as contrast agents for ultrasound/magnetic resonance dual-modality imaging. *Acta Biomater.* 2015, 24, 266–278.