Water-soluble gadolinium nanoparticles coated with DO3A-benzothiazole conjugate: Synthesis and application as a potential theragnostic agent

Min-Kyoung Kang[1], Hee-Kyung Kim[1], Ki-Hye Jung[2], Ji-Ae Park[3], Seung-Tae Woo[4], Joo-Hyun Kim[4], Eun-Young Jeon[5], Tae-Jeong Kim*[2], and Yongmin Chang*[1,6]

[1]Medical & Biological Engineering, Kyungpook National University, Daegu, Korea, [2]Applied Chemistry, Kyungpook National University, Daegu, Korea, [3]Molecular Imaging Research Center, Korea Institute of Radiological & Medical Science, Seoul, Korea, [4]Bayer Healthcare, Medical care, Seoul, Korea, [5]The Advanced Medical Technology Cluster for Diagnosis & Prediction, Kyungpook National University, Daegu, Korea, [6]Diagnostic Radiology and Molecular Medicine, Kyungpook National University, Daegu, Korea

Introduction

Magnetic resonance imaging (MRI) has proved to be a powerful non-invasive technique. The prominent advantage of MRI is a high spatial resolution and the ability to distinguish soft tissues. The contrast of the resulting image can be enhanced by injection of paramagnetic or superparamagnetic agents. Paramagnetic gadolinium (III) complexes, for example, have so far been most widely used in MRI as water relaxation agents to improve image contrast. In recent years, interest in gadolinium nanoparticles (GdNPs) has risen in the expectation that these systems would serve not only as MRI probes but also as therapeutic agents through Gd-based neutron therapy (Gd-NCT) and chemotherapy in conjunction with those possessing anti-tumor activity. One such example may include benzothiazoles which are known to possess potent antitumor properties in select breast, ovarian and renal cancer cell lines. In these studies are presented the synthesis, MR properties, tumor-targeting and anti-tumor activities of a series of GdNPs coated with DOTA and DO3A-benzothiazole conjugate.

Material and Methods

All reagents were purchased from commercial sources and used as received. GdNPs and Gd@SiO_2 were prepared according to the literature method. Characterization of new materials have been performed by analytical and various spectroscopic techniques (MRI, TEM). T_1 measurements were carried out using an inversion recovery method with variable inversion time (TI) at 1.5 T (64 MHz). T_1 relaxation times were obtained from the non-linear least square fit of the signal intensity measured at each TI value. T_2 relaxation times were obtained from the non-linear least squares fit of the mean pixel values for the multiple spin-echo measurements at each echo time (TE). Six-week-old male ICR (Institute of Cancer Research) mice with weights of 28 − 32 g were used for the MRI. The mice (n = 4) were anesthetized with 1.5 % isoflurane in oxygen. Measurements were made before and after injection of Gd@SiO_2-NHCO-DO3A via tail vein. The amount of CA per each injection is 0.1 mmol [Gd]/kg for MR images. Whole body MR images were obtained with a 1.5 T MR unit (GE Healthcare, Milwaukee, WI, U.S.) equipped with a homemade small animal rf coil. The coil was of the receiver type with its inner diameter being 50 mm. The imaging parameters for SE (Spin echo) are as follows: repetition time (TR) = 300.0 ms; echo time (TE) = 12.0 ms; 7.0 mm field of view (FOV); 192×128 matrix size; 1.2 mm slice thickness; number of acquisition (NEX) = 8. Images were obtained during 120 min after injection.

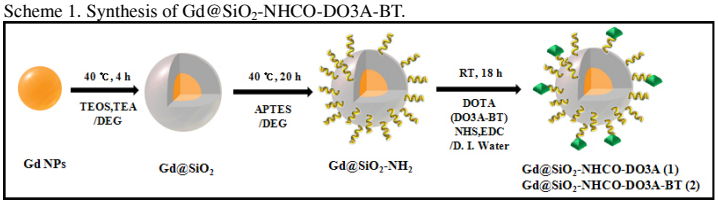
Results and Discussion

Scheme 1 shows the route leading to the formation of two gadolinium nano-systems coated with DOTA (Gd@SiO_2-NHCO-DO3A) and with DO3A-benzothiazole (Gd@SiO_2-NHCO-DO3A-BT) for use as a new family of multifunctional MRI/optical contrast agents (CAs). Both systems are completely soluble both in water and PBS with the gadolinium concentration [Gd] up to 38.6 mM^{-1} in water and 80.0 mM^{-1} in PBS (cf. Figure 1B). The R_1 relaxivities of Gd@SiO_2-NHCO-DO3A in water and PBS are 6.85 $mM^{-1}s^{-1}$ and 8.29 $mM^{-1}s^{-1}$, respectively (Table 1). An increase in R_1 in PBS as compared with that in water is rather contrasting in that quite an opposite observation is to be expected with low molecular-weight Gd-chelates. Namely, Gd-ions are known to be replaced by the phosphate ion in PBS to form the Gd-phosphate salt. Figure 1A shows R_1 and R_2 maps of Gd@SiO_2-NHCO-DO3A as functions of the gadolinium concentration. The R_1 and R_2 maps become the brightest with [Gd] = 1 mM. Figure 2 shows the coronal T_1-weighted images of mice, ICR with Gd@SiO_2-NHCO-DO3A. The pattern of *in vivo* MR images compares well with conventional low molecular-weight ECF MRI CAs based on Gd-chelates such as Dotarem® with signal enhancement in liver, kidney, and bladder. Enhancement in bladder supports the renal excretion. Further studies were carried out to investigate the antitumor activity of Gd@SiO_2-NHCO-DO3A-BT. Indeed, anti-tumor activity toward the breast cancer cells such as MCF-7 was clearly observed to make this system a new family of multifunctional theragnostic agents. These results will also be presented in detail in the poster.

Conclusions

The work describes the synthesis and characterization of Gd@SiO_2-NHCO-DO3A for use as a multifunctional theranostic agent. This system shows complete solubility in both water and PBS. The R_1 relaxivities are 6.85 $mM^{-1}s^{-1}$ and 8.29 $mM^{-1}s^{-1}$ in water and PBS, respectively. The pattern of *in vivo* MR images compares well with conventional low molecular-weight ECF MRI CAs based on Gd-chelates such as Dotarem® with signal enhancement in liver, kidney, and bladder. The same gadolinium nanoparticles, when modified with DO3A-BT, exhibit anti-tumor activity in the breast cancer cells such as MCF-7 as well as bimodal MR/optical imaging properties.

Scheme 1. Synthesis of Gd@SiO_2-NHCO-DO3A-BT.



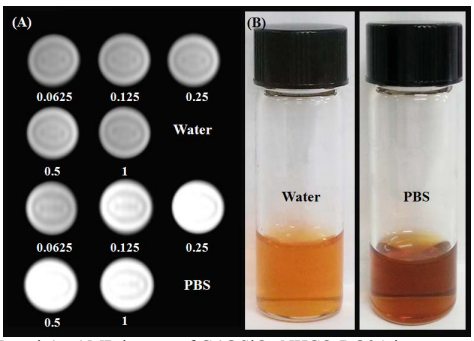


Figure 1. (A) T_1-weighted MR images of Gd@SiO_2-NHCO-DO3A in water and PBS: [Gd] = 0.0625 – 1.0 mM) at 1.5 T and 293 K. (B) Photographic images of Gd@SiO_2-NHCO-DO3A.

Table 1. Relaxivities of Gd@SiO_2-NHCO-DO3A at the 1 mM concentrations and 293 K.

Gd@SiO_2-NHCO-DO3A	R_1	R_2
Water	6.85 ± 0.19	7.08 ± 0.17
PBS	8.29 ± 0.13	9.33 ± 0.34

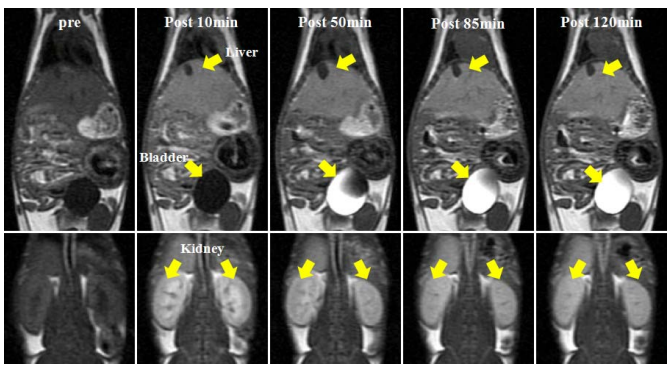


Figure 2. *In vivo* MR coronal images of mice obtained with Gd@SiO_2-NHCO-DO3A at 1.5 T and 293 K.